Thesis 1419

AN EVALUATION OF STRATEGIES FOR PRODUCTION OF NILE TILAPIA (OREOCHROMIS NILOTICUS L.) FRY SUITABLE FOR HORMONAL TREATMENT

A thesis presented for the degree of

Doctor of Philosophy to the University of Stirling

bу

Institute of Aquaculture
University of Stirling
Stirling
Scotland

1989

The work presented in this thesis is the result of my own investigations and has neither been accepted nor is being submitted for any other degree.

Daniel Mainten Supervisor 25/2/90 Date

CONTENTS

		<u>Page</u>
ACKNOWLE	DGEMENTS	Ť
LIST OF	TABLES	iii
LIST OF F	FIGURES	хi
LIST OF A	ABBREVIATIONS	xv
ABSTRACT		xvi
CHAPTER 1	: GENERAL INTRODUCTION	
1.1	General introduction	1
1.2	Intensive hatchery production of commercial fish species - the tilapia as a special problem	7
1.3	A review of existing fry production systems	14
1.3.1	Reproduction under natural conditions	14
1.3.2	Reproduction in culture systems	16
1.4	Major factors affecting <u>Oreochromis</u> fry production under culture conditions	23
1.4.1	Breeding intensity	23
1.4.2	Incubation success	26
1.4.3	Harvest efficiency	29
1.5	Scope of research and nomenclature	32
CHAPTER 2	: BROODSTOCK EXCHANGE STRATEGIES IN TANKS AND	HAPAS
2.1	Background	34
2.2	General methodology	46
2.2.1	Experimental system design	46
2.2.1.1	Concrete tanks within a recirculation system	46
2.2.1.2	Nylon hapas in earthen ponds	51

		<u>Page</u>
2.2.2	Experimental methods	53
2.2.2.1	Concrete tanks	53
2.2.2.2	Nylon hapas	54
2.2.2.3	Seed storage and analysis	55
2.3	The use of intensive broodstock management strategies for seed production in concrete tanks	58
2.3.1	Experimental methodology	58
2.3.2	Results	69 .
2.3.3	Discussion	78
2.4	Effect of conditioning environment and exchange strategy on seed production in tanks and hapas	89
2.4.1	Background	89
2.4.2	Methods	90
2.4.3	Results	96
2.4.3.1	System data	96
2.4.3.2	Data for individual fish	102
2.4.4	Discussion	113
2.5	The effect of duration of female broodfish conditioning and selective female and male broodfish exchange on seed production in large nylon hapas	123
2.5.1	Methods	123
2.5.2	Results	128
2.5.3	Discussion	136
CHAPTER 3	: EFFECT OF EARLY BROODSTOCK PRODUCTION AND ST DENSITY IN SPAWNING TANKS AND HAPAS ON SEED PRODUCTION	FOCKING
3.1	Broodstock production	144
3.1.1	Background	144

		<u>Page</u>
3.1.2	Methods	149
3.1.3	Results	154
3.1.4	Discussion	155
3.2	Effect of broodstock density on tilapia seed production	157
3.2.1	Background	157
3.2.2	Methods	162
3.2.3	Results	169
3.2.4	Discussion	184
CHAPTER 4	: SWIM-UP FRY PRODUCTION IN EARTHEN PONDS	
4.1	Background	195
4.2	Methods	200
4.2.1	Pond preparation	200
4.2.2	Broodstock	200
4.2.3	Selection and pond management	201
4.2.4	Fry removal	202
4.2.5	Estimation of fry numbers	204
4.2.6	Broodstock harvest	204
4.3	Results	212
4.3.1	Effect of harvest intensity	212
4.3.2	Effect of broodstock size	215
4.4	Discussion	219
CHAPTER 5	: INCUBATION	
5.1	Background	226
5.2.1	Methods	233
5.2.1.1	Experimental system	233

•

		<u>Page</u>
5.2.1.2	Experimental methods	234
5.2.2	Results	239
5.3	Discussion	243
5.4	General discussion of systems investigated	251
CHAPTER 6	: ECONOMIC ANALYSIS OF FRY PRODUCTION	
6.1	Introduction	260
6.2	Methods and assumptions of analysis	268
6.2.1	The strategies	268
6.2.1.1	The production of swim-up fry for hormone treatment	268
6.2.1.2	Hormone treatment to produce MT-fry	276
6.2.1.3	The use of MT-fry for grow-out	279
6.2.2	Analysis	281
6.2.2.1	Framework of methodology	281
6.2.2.2	Determination of costs	283
6.2.2.3	Determination of revenues	289
6.3	Determination of production costs	295
6.3.1	Swim-up fry	295
6.3.1.1	Central Thailand farms	295
6.3.1.2	Northeast Thailand farms	300
6.3.2	Hormone treatment costs	304
6.4	Determination of revenues	306
6.4.1	Fry output	306
6.4.2	Fry price	306
6.4.2.1	Determination of different levels of fry price	307
6.4.3	Value of by-products of fry production	315

		<u>Page</u>
6.5	Results	318
6.5.1	Central Thailand farms	318
6.5.1.1	Calculation of Net Present Value (NPV)	318
6.5.1.2	Marginal analysis of undominated treatments	318
6.5.1.3	Sensitivity analysis	320
6.5.2	Northeast Thailand farms	323
6.5.2.1	Calculation of Net Present Value (NPV)	323
6.5.2.2	Sensitivity analysis	324
6.6	Discussion	325
6.6.1	Comparison between hatchery systems	325
6.6.2	Comparison between production of hapa	
	MT-treated tilapia fry in Central and Northeast Thailand	327
6.6.3	Adoption of new seed technologies by farmers	332
6.6.4	Recommendations	337
CHAPTER 7:	: CONCLUSIONS	339
REFERENCES		343
APPENDICES		362

ACKNOWLEDGEMENTS

This piece of work has been completed only with the assistance of a large team of people at the Asian Institute of Technology and the constant support and encouragement from friends and colleagues both in Thailand and the U.K.

I would particularly like to thank Dr. Don Macintosh for his efforts to supervise this thesis, especially during the final stages.

My initial and continued presence at AIT was only possible due to the efforts and patience of Professor Peter Edwards and his support and friendship throughout are sincerely appreciated.

Dr. Hermann Waibel spent many hours unravelling the mysteries of economic analysis for which I am sincerely grateful.

I would like to thank Khun Chintana Boonthumjinda both for her direct role in the microbiological analysis and more generally for her unbridled enthusiasm and energy and Khun Manoj Yomjinda for monitoring water quality throughout the study.

I am extremely grateful to the many people who diligently counted tilapia eggs, a most boring chore. The practical incompetence of the author were happily balanced by the skill and savvy of Khun Manu and the patience and reliability of many of his brothers, particularly Khun Manop, are readily acknowledged.

I thank Mike Skladany and Bob Rode as friends and collaborators on the survey and analysis of small-scale hatcheries in Northeast Thailand; the staff of Udorn Patina farm for their

past and continuing role in the authors education and Nick Innes-Taylor and the rest of the AIT Aquaculture team for their patience.

I am especially grateful to Khun Paisinee for help with the statistics.

Secretarial assistance was provided by Khun Yupa, Khun Nee Khun Kanchana and Khun Kung at various times and graphics by Khun Aurai and AIT's drafting department.

Both David Sampson and Amaranate Yakupitiage are thanked for their critical input at various times during the study.

At Stirling the many talents of Hiliary Duggua were continually in demand and Dr. Krishan Rana's comments and access to valuable literature much appreciated.

I am greatly indebted to the EEC and the Overseas Development Administration for the financial support that made this study possible.

Last, but not least, many thanks to Dorrit for sticking out the ordeal.

LIST OF TABLES

<u>Table</u>		<u>Page</u>
1.1	Status of fry production of major commercially farmed fish in Asia	6
1.2	Strategies used for the production of tilapia fry	15
1.3	A comparison of swim-up fry from the three major types of system (Pond, Hapa and Tank) using intensively managed broodstock in terms of different productivity measures	18
1.4	Summary of research scope	31
2.1	Physical parameters of the recirculated water tank system in the EEC hatchery at AIT used for tank experiments (T1, T2, T3, H1)	48
2.2	The experimental design for investigating the effect of exchange of female broodstock on spawning in concrete tanks (T1)	57
2.3	Mean 5 day output of ssed from concrete tanks (Area = 12.57 m^2) before and after change in frequency of harvest from 10 to 5 days, for different strategies of broodfish disturbance and exchange (T1)	64
2.4	The percentage of total seed production by female broodfish in concrete tanks observed at harvest as stage 3-4 and stage 1-1.5 after a change of frequency of harvest from 10 to 5 days for different strategies of female broodfish exchange (T1)	66
2.5	Growth parameters of broodfish at stocking and at harvest after a period (t = 155 days) of seed production in concrete tanks (Area = 12.57 $\rm m^2$) for different strategies of broodstock exchange and disturbance (T1)	67
2.6	Percentage change in body weight of mouthbrooding female broodfish during (a) the interharvest period (10 days) and (b) the conditioning period for different strategies of female broodfish exchange (T1)	68
2.7	The degree of association (r) between the change in body weight of female broodfish during the interharvest period (10 days) and after a subsequent period of conditioning and stage of seed recovered at harvest for three different strategies of female broodfish exchange (T1)	72

<u>Table</u>		<u>Page</u>
2.8	The degree of association (r) between mean reproductive traits and body weight for <u>O. niloticus</u> females under different exchange strategies	75
2.9	The relationship of seed clutch size (all stages) and body length for female broodfish spawned in concrete tanks (A = 12.57 m²) under different exchange strategies (T1). Seed removed from the mouth of individual females every 10 days; comparative data given for Oreochromis niloticus from other workers	85
2.10	The experimental design for investigation of the effect of environmental change and female broodfish exchange strategy on spawning in nylon hapas suspended in earthen ponds (H1)	87
2.11	The experimental design for investigation of the effect of environmental change and female broodfish exchange strategy on spawning in concrete tanks (T2)	88
2.12	Mean total seed output per spawning unit per harvest (5 days) in tanks (Area = 12.57 m^2 , T2) and hapas (Area = 40 m^4 , H1) for different female exchange strategies and conditioning environments	92
2.13	Early spawned seed as a percentage of total seed production harvested every 5 days from (a) spawning units stocked with female broodfish managed under a range of different exchange and conditioning strategies (b) spawning hapas stocked with females conditioned in tanks or hapas and (c) all spawning tank and hapa treatments	95
2.14	Intercepts, a, $(\pm \text{ S.D.})$; regression coefficient, b, $(\pm \text{ S.D.})$; coefficient of determination (r^i) of the linear regression analysis for reproductive traits in <u>O. niloticus</u> female broodfish maintained in tanks and hapas under different exchange and conditioning regimes	97
2.15	Summary of seed production parameters of female broodfish spawned in tanks and hapas under different exchange and conditioning regimes	98
2.16	Parameters of seed production for female broodfish spawned in tanks (Area = 12.57 m ² , T2) and hapas (Area = 40 m ² , H1) for different female exchange strategies and conditioning environments	100

<u>Table</u>		<u>Page</u>
2.17	Effect of spawning frequency on size characteristics at final harvest of tagged female broodfish maintained in tanks (all treatments combined, tagged females 30% of total number of fish at stocking)	103
2.18	Mean total and reproductive production of female broodfish maintained in tanks and hapas under different exchange and conditioning regimes (all tank and hapa treatments combined, respectively)	104
2.19	Observed interspawning interval (ISI) of individual, tagged female broodfish maintained in tanks and hapas under different exchange and conditioning regimes	108
2.20	Gonadosomatic index at final harvest of female broodfish maintained under different exchange and conditioning regimes (a) in spawning and conditioning tanks and hapas (b) after exchange of either all or spawned females and (c) of unspawned females harvested from spawning tanks and hapas	110
2.21	Crude protein, total lipid and moidsture content of whole ovaries from spawned and unspawned broodfish spawned in tanks and hapas under different exchange and conditioning regimes at final harvest	111
2.22	Mean ISI of all females held in spawning units under different exchange and conditioning regimes	119
2.23	Experimental design used to study effect of conditioning period and male and female broodstock exchange on spawning in nylon hapas suspended in earthen ponds	122
2.24	Seed production in hapas under different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)	125
2.25	Mean percentage of total seed harvested as uneyed eggs (stage 1) from spawning hapas stocked with broodfish managed under different conditioning and exchange regimes	129
2.26	Parameters of seed production for female broodfish spawned in hapas after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)	130
2.27	Mean frequency of seed harvest and ISI of female	

.

<u>Table</u>		<u>Page</u>
	broodfish spawned in hapas after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)	132
2.28	Individual size and growth characteristics at (a) stocking and (b) final harvest of broodfish spawned in hapas after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange	133
3.1	Mean size of female fish raised in fertilised earthen ponds using three different supplementary feeds prior to their use as broodfish in concrete tanks at different densities (T3)	151
3.2	Mean total net yield and survival of fish raised on three different supplementary feeds in earthen ponds prior to their use as broodfish in concrete tanks at different densities (T3)	152
3.3	Frequencies of observed and expected harvest seed clutches from females fed different supplementary diets prior to their use as broodfish in concrete tanks at different densities (T3)	153
3.4	Experimental design used to study effect of broodstock density on seed production in circular tanks (Area = 12.57 m^2)	163
3.5	Experimental design used to study effect of broodstock density on seed production in rectangular hapas (Area = 40 m^2)	164
3.6	Productivity of tank and hapa seed production systems over a range of broodstock densities (T3, H2)	167
3.7	Productivity of tank and hapa seed production systems over a range of broodstock densities (T3, H2)	168
3.8	Variation in clutch size of seed harvested from broodfish in tanks and hapas (a) seed harvested of different stages (mean of all density treatments) and (b) seed harvested from broodstock spawned at different densities (T3, H2)	171
3.9	Synchrony of spawning in (a) tanks and hapas stocked with broodfish at different densities and (b) in hapas over a range of stocking densities for different periods within the experimental period	172

<u>Table</u>		<u>Page</u>
3.10	Mean frequency of females in tanks and hapas stocked at different densities as a percentage of fish stocked per spawning period (5 days)	173
3.11	Mean Interspawning Interval (ISI) of female broodfish stocked at different densities in tanks and hapas	177
3.12	Body characteristics of female broodfish stocked at different densities in tanks and hapas at final harvest	178
3.13	Mean feeding rate, actual feed input and biomass of broodfish stocked at different densities in tank and hapa systems	179
4.1	Mean number of fry per 5 days of two size classes removed by dip-net from the shallow margin of large earthen ponds (Area = 1740 m²)	204
4.2	Oreochromis <u>niloticus</u> fry suitable for hormone treatment as a percentage of total fry harvested by dip-net from the shallow margin of large earthen ponds (Area = 1740 m²) over a study period of 105-116 days	208
4.3	Efficiency of removal of two size classes of fry by dip-net from the shallow margin of large earthen ponds (Area = 1740 m^2) at two levels of harvest intensity (t = 116 days)	209
4.4	Characteristics of female broodfish at harvest following fry production in large earthen ponds (Area = 1740 m^2) using (a) different harvest intensity (P1) and (b) broodfish of different size class (P2) (t, days; P1 =116, P2 =105)	211
4.5	Synchrony of fry production in large earthen ponds (Area = 1740 m^2) as indicated by the proportion of total fry harvest removed by dip-net in the shallow pond margins during the first 33 days (Experimental period; P1 = 116 days , P2 = 105 days)	216
5.1	Percentage survival to swim-up of <u>Oreochromis</u> seed of different stage harvested from (a) tanks and (b) hapas and placed in 20 1 downwelling plastic incubators supplied with recirculated, slow-sand filtered water in three sequential trials (n = 3)	235
5.2	(a) Number of seed and percentage survival to	

<u>Table</u>		Page
	spawning tanks and hapas (combined) and incubated in 20 l plastic incubators supplied with recirculated, slow-sand filtered water in 10 sequential trials (b) Maximum density of seed during incubation in the same vessel	236
5.3	Total bacteria in incubation water as measured by standard plate count during a complete incubation cycle in four 20 l plastic vessels supplied with recirculated, slow-sand filtered water	237
5.4	Physico-chemical parameters of the system water during incubation of <u>Oreochromis</u> seed in 20 1 plastic vessels supplied with recirculated, slow-sand filtered water	238
5.5	Range of water flow rate and residence time in downwelling 20 l plastic vessels supplied with recirculated, slow-sand filtered water in the present trial with comparative data from other studies	240
5.6	Summary of methodology and effectiveness of artificial incubation of <u>Oreochromis</u> seed compared to natural incubation	242
5.7	Summary of estimated swim-up fry production from experimental treatments in concrete tanks, nylon hapas-in-ponds and earthen ponds	250
5.8	Indicators of system efficiency for experimental treatments in concrete tanks, nylon hapas-in-ponds and earthen ponds	254
6.1	Number of tilapia and carp seed produced annually per unit weight of broodstock raised (seed kg ⁻¹) and unit area of nursery pond (seed m ⁻²) in 3 hatcheries in Northeast Thailand	262
6.2	Production of the common, herbivorous fish species in Central and Northeast Thailand	267
6.3	Comparison of size and productivity of five hatcheries in Northeast Thailand	269
6.4	Nomenclature of economic models for Northeast and Central Thailand using experimental treatments as basis for analysis	271
6.5	Definition of main categories of fixed costs in economic analysis of swim-up fry production in Central Thailand model	284

<u>Table</u>		<u>Page</u>
6.6	Major categories of variable cost for swim-up fry production	285
6.7	Discounted fixed cost (investment period 10 years) by category for 29 swim-up fry production strategies in Central Thailand	291
6.8	Initial fixed costs by major category for all fry production strategies in the Central Thailand model	292
6.9	Initial fixed costs for (1) spawning category by item for tank strategies (T31-T34) and (2) incubation category by item for hapa strategies (H31-H35) in Central Thailand	293
6.10	Annual variable cost by category for all fry production strategies in the Central Thailand model	294
6.11	Discounted total costs (investment period 10 years) and initial investment costs for 43 swim-up fry production strategies in Northeast Thailand	299
6.12	Annual variable costs by category for selected swim-up fry production strategies (P23, H12, T11, H 34a, H34b, HUPF) in five hatcheries in Northeast Thailand	301
6.13	Cost of hormone treatment for one million fry in (i) shallow concrete tanks (Area = 4 m^2) and (ii) nylon hapas (Area = 5.4 m^2)	303
6.14	Annual swim-up fry production for strategies in Northeast and Central regions of Thailand	305
6.15	Mean harvested yields by size class of MT-treated and untreated <u>O. niloticus</u> stocked in 200 m² fertilised (septage) earthen ponds within the AIT experimental system for 5 months and fed fine rice bran	308
6.16	Marginal analysis of substitution of MT-treated fry for untreated fry and later marketing in an urban market in the Northeast of Thailand	309
6.17	Marginal analysis of the use of MT-treated fry for three different market scenarios	310
6.18	Total annual value of revenue components for 29 swim-up fry production strategies in Central Thailand	313

<u>Table</u>		<u>Page</u>
6.19	Marginal analysis for investment in undominated strategies in Central Thailand of different discounted total cost (fry price Bt 0.1) after sex-reversal in tanks and hapas	317
6.20	Undominated strategies for producing swim-up fry over a range of discounted total cost in five hatcheries in Northeast Thailand	321

LIST OF FIGURES

<u>Figure</u>		<u>Page</u>				
1.1	Natural reproductive cycle of <u>Oreochromis</u> indicating approximate duration and location of spawning and conditioning	13				
2.1	Fry production in large static water concrete tanks (Area = $100~\text{m}^2$) with production period in the Philippines	37				
2.2	Plan of the EEC Tilapia Hatchery, Asian Institute of Technology, Bangkok, Thailand. Dimensions in metres.	44				
2.3	Section of the main components of the EEC Tilapia Hatchery showing water supply and filtration, spawning and conditioning tanks	45				
2.4	Section of horizontal filter used in spawning and conditioning systems of the EEC Tilapia Hatchery	47				
2.5	Plan of earthen pond and hapa-in-pond experimental facility at AIT	50				
2.6	Section of design of hapa-in-pond experimental facility at AIT. Dimensions in metres.					
2.7	Actual production of seed per harvest from concrete spawning tanks (Area = 12.57 m²) over experimental period (a) T11; no exchange of females at harvest (b) T12; all females exchanged at harvest (c) T13; swim-up fry removed daily, no disturbance of broodfish (d) T14; spawned females exchanged at harvest	62				
2.8	Estimated production of seed per 5 day period from concrete spawning tanks (Area = 12.57 m²) over experimental period (a) T11; no exchange of females at harvest (b) T12; all females exchanged at harvest (c) T14; spawned females exchanged at harvest	63				
2.9	Relationship between time of spawning and stage of seed harvasted for broodfish stocked in spawning tanks and hapas for a 5 day period	65				
2.10	The relationship between percentage change in total body weight during spawning (Interharvest period 10 days) of individual mouthbrooding females with stage of seed harvested (a) T11 all					

	•	
	females replaced in spawning tank; (b) T12 all females exchanged; (c) T14 spawned females exchanged	73
2.11	The relationship between percentage change in total body weight of individual female broodfish after seed harvest during conditioning period with stage of seed (a) T11 all females replaced in spawning tank, (b) T12 all females exchanged (c) T14 spawned females exchanged	74
2.12	Total seed production per harvest per tank (Area = 12.57 m^2) over the whole experimental period for broodfish under different exchange and management regimes (T21 - T24)	93
2.13	Total seed production per harvest per hapa (Area = 40 m^2) over the whole experimental period for broodfish under different exchange and management regimes (H11 - H14)	94
2.14	Frequency of seed harvest from tagged individual female broofish spawning in (a) hapas and (b) tanks over the experimental period of 116 days	101
2.15	Seed production as a percentage of total production (from the harvest of the first seed clutch to end of experiment; 116 days) of individual female broodfish over a range of treatments in (a) tanks (T21 - Y24) and (b) hapas (H11 - H14)	105
2.16	Relationship between total weight of seed produced and individual body weight gain (from the harvest of the first seed clutch to end of experiment; 116 days) of female fish spawned over a range of treatment in (a) tanks (T21 - Y24) and (b) hapas (H11 - H14)	107
2.17	Mean daily production of seed per hapa (Area 40 m²) for treatments exchanging female broodfish completely after 10 days conditioning (H31) and (a) selectively exchanged females (H34) after a minimum of 10 days conditioning (b) completely exchanged females conditioned for 20 days (c) completely exchanged females conditioned for 15 days and (d) completely exchanged males and females conditioned for 10 days. Interharvest Interval of 5 days for all treatments	126
2.18	Variation in (a) mean daily production of seed per g of female broodfish and (b) mean dissolved oxygen level at dawn in spawning and conditioning hapas under different broodstock conditioning and exchange strategies over the experimental period	

	(all treatments combined)	127
3.1	Effect of stocking density on mean daily output of seed from (a) tanks (Area = 12.57 m²; Interharvest Interval 5 days; sex ratio = 1:1 and (b) hapas (Area = 40 m²; Interharvest Interval 5 days; sex ratio = 1:1)	166
3.2	Density of all broodfish (g/m^2) in (a) tanks (Area = 12.57 m²) and (b) hapas (Area = 40 m²) mainatined at four different densities (by number, sex ratio = 1:1)	170
3.3	Dissolved oxygen level at dawn (a), mean harvest clutch size of seed (b) and frequency of female broodfish harvested with seed in the mouth (c) from female broodfish maintained at variable density (density of all broodfish < 800 g/m²; sex ratio 1:1) in nylon hapas (Area 40 m²) suspended in an earthen pond	175
3.4	Mean dissolved oxygen in (a) spawning tanks (Area = 12.57 m²) and (b) spawning hapas (Area = 40 m²) stocked at variable densities (tanks \langle 3 kg/m²; hapas \langle 0.8 kg/m²) measured weekly at dawn and at 09.00 a.m. respectively (mean of all treatments)	180
4.1	Production of two size classes of swim-up fry removed by dip-net from the margin of large earthen ponds (Area = 1740 m^2) (a) at high harvest intensity (6 times per day) and (b) low harvset intensity (3 times per day)	207
4.2	Mean maximum and minimum temperatures measured daily at 20 cm below the water surface during swim-up fry production in large earthen ponds (a) at different levels of harvest intensity (P1) and (b) using two size classes of broodfish (P2)	210
4.3	Main categories of harvested net production (wet weight) from partial and final draining of large earthen ponds (Area = 1740 m^2) after experiments using (a) two levels of harvest intensity (P1) and (b) two size classes of broodfish (P2)	213
4.4	Production of swim-up fry suitable for hormone treatment (mean weight < 0.015 g) in large earthen ponds (Area = 1740 m²) (a) at different levels of harvest intensity and (b) using two size classes of broodfish	214
5.1	Incubation system used for experimental trials in the AIT hatchery suitable for incubating > 100,000 seed at a time (a) cross section (b) plan	231

5.2	Design and dimensions of incubation bottle (Volume = 20 l) used in experimental trial in the AIT hatchery	232
5.3	A flow diagram of major system processes for Oreochromis seed production in recirculated water supplied tanks, static hapa-in-pond and earthen pond systems	257
6.1	Strategies used for the production of <u>Oreochromis</u> fry	264
6.2	The location of hatcheries modelled for production of MT-treated fry in Central and Northeast Thailand	266
6.3	Schema showing the strategies for producing MT-treated tilapia fry in Central Thailand	273
6.4	Dominance analysis using a Net Present Value curve	282
6.5	Summary of methodology for the estimation of Net Present Value (NPV) for MT-treated fry production in Central Thailand	286
6.6	Discounted fixed and variable costs as a proportion of total discounted costs of undominated swim-up fry production strategies in Central Thailand	298
6.7	The relationship between the marginal rate of return (MRR) and fry price after substitution of MT-treated fry for untreated fry by a grow-out farmer	311
6.8	Discounted cash flow of 29 swim-up fry production strategies in Central Thailand after (a) sex-reversal in tanks.	316
6.9	Sensitivity of IRR (16% and 20%) of fry price for the production of MT-treated tilapia after treatment in (a) hapas and (b) tanks after swim-up fry production in a range of pond, hapa and tank methods (undominated strategies)	319
6.10	Discounted cash flow of 56 swim-up fry production strategies in hatcheries in Northeast Thailand after sex-reversal in hapas strategies (a) mean for Nongkhai Surin and Udorn (b) Ubon	322

LIST OF ABBREVIATIONS

Asian Institute of Technology AIT

BL body length

Bt Baht

body weight BW C.P. crude protein

COD chemical oxygen demand

CS clutch size

coefficient of variance CV

.DO dissolved oxygen

European Economic Community EEC

ETN egg total number ETW egg total weight

F value one-way Anova F FCR Food Conversion Ratio GSI gonadosomatic index

human chorionic gonadotropin HCG

IEW individual egg weight IHI Interharvest Interval Internal Rate of Return IRR ISI Interspawning Interval Fulton's Condition Factor Κ

Marginal Rate of Return MRR

11-a methyltestosterone treated fry MT-fry

ΜT 11-a methyltestosterone

number n

compounded fertilizer (nitrogen & phosphorous) NP

NPV Net Present Value degree of association r,

coefficient of determination

SL standard length sts septic tank slurry

TL total length

ABSTRACT

Intensive methods for the mass production of <u>Oreochromis</u> <u>niloticus</u> (Chitralada strain) seed using concrete tanks, hapas within earthen ponds and earthen ponds were investigated. On the basis of these trials, the productivity and economic performance of various production strategies for hormonally sex-reversed <u>Oreochromis</u> fry (MT fry) were then compared and modelled for adoption in Central and Northeast Thailand.

Regular disturbance and harvesting of seed after a short period of spawning opportunity (5-10 days) was found to increase seed production in concrete spawning tanks. Exchange of female broodfish increased synchrony of breeding.

A change in conditioning and spawning environment had no effect on seed yield from spawning tanks and hapas (area =12.57 m² and 40m² respectively). Seed wet weight, seed clutch size and weight was greater in female fish spawned in tanks than hapas. Females conditioned in hapas however produced heavier seed clutches of larger absolute and relative size than tank conditioned fish.

Records of tagged females indicated considerable differences in the frequency of spawning; in hapas the distribution was normal wheras in tanks it was skewed. The evidence suggests that hierarchy is important in the control of reproduction and exerts it's strongest effect in clear water, densely stocked tanks.

Selective female broodfish exchange optimised seed yield per unit weight of broodfish and seed production was not improved by conditioning females for periods longer than 10 days. Male broodfish exchange did not significantly improve (P > 0.05) seed yields.

Early nutrition of broodfish raised under different supplemental feeding regimes in fertilised earthen ponds had a significant effect on later spawning frequency in concrete tanks. However, this effect was confined to broodfish maintained at densities lower or higher than optimal for seed production.

Broodfish stocked over a range of densities for extended periods (201 days) showed greater variability of seed production in hapa than tank production systems. This was mainly due to periods of poor water quality in hapas; when water quality was high seed production was significantly higher in hapas than tanks over a range of broodfish densities.

The optimal density of broodfish for seed production was exceeded in tanks but not hapas. The relationship between seed production and broodfish density over time suggested that both stocking biomass and number have an effect on fry output. Density of broodfish showed an inverse relationship to clutch

size in both tanks and hapas and synchrony of spawning in tanks.

Production of swim-up fry in large earthen ponds (area=1740m²) was not significantly different (P>0.05) at 2 levels of harvest intensity.

The use of small broodfish however produced double the yield of hormone treatable fry than a similar biomass of larger broodfish of the same cohort.

A commercial scale incubation system was devised and evaluated in order to allow tank and hapa systems harvesting unhatched seed to be compared with the production of swim-up fry obtained from earthen ponds. Seed removed from mouthbrooding females was roughly staged and incubated in batches of similar development to give information on survival to swim-up fry. A simple incubation system was designed with a capacity for hatching >100,000 eggs/set. A mean survival of 75% of all harvested seed to swim-up fry was obtained over several trials.

A trend to intensification (fry/m²/day) from ponds to hapas to tanks is evident when yields of swim-up fry are compared. Productivity exceeded any in the published literature for comparable systems, largely because of the intensity of broodstock management and early and efficient harvest of seed. Broodfish productivities (fry/kg female/month) were also higher across the range of systems tested often by a factor of 1.5-3.

The best strategies were selected over a range of total investment cost using dominance analysis. Economic analysis suggested that for a start-up operation in Central Thailand fry production in earthen ponds can give the best return on levels of investment of less than Baht 0.8 million. Substitution of techniques into current carp fry production operations in Northeast Thailand indicated that more intensive methods (production in tanks and hapas) are more attractive over a range of investment levels. The break-even price of MT fry after hormone treatment in nylon hapas was approximately half the cost of treatment in a recirculated water concrete tank system. The break-even price in Central Thailand was lower than the Northeast by a factor of around 1.5 but the break-even price for both areas was lower than the current price of untreated Oreochromis fry.

CHAPTER 1

GENERAL INTRODUCTION

1.1 Introduction

The lack of a reliable supply of fry has been an important constraint to the promotion of many species of fish in aquaculture and can still be a limiting factor where infrastructure is poorly established or finances limited. Thus, self sufficiency of fry production has been found to be essential for the success of fish culture amongst small-scale farmers in remote areas. Tilapias are one of the few cultured species that can make this goal achievable (Lovshin and Pretto, 1983).

Recently, the many desirable culture characteristics of the tilapias have also stimulated their use in a range of more intensive grow-out systems (Balarin and Haller, 1982).

The Nile tilapia (Oreochromis niloticus), perhaps the most suitable tilapia species for culture, is believed to have evolved in unstable African riverine environments (Fryer and Iles, 1972; Noakes and Balon, 1982) but is now bred and cultured in a large range of natural and artificial aquatic systems. Current methods for production of Oreochromis niloticus fry vary greatly in intensity. Simple systems have existed for many years which combine breeding and grow-out of tilapias in the same earthen pond. The free-breeding habit of most cultured species of tilapia, frequently reported since the earliest descriptions by fishery biologists (Aronson, 1949; Lowe McConnell, 1955; Fryer, 1961), must have been important for their early success as farmed fishes.

The high densities of tilapia that result from free-breeding

may produce considerable yields of fish from simple pond systems; if acceptability of small fish is not a problem this may represent the most viable system. However, uncontrolled spawning and recruitment makes management of the stocked fish to produce large, even-sized individuals difficult. Under most pond conditions as the density of fish increases feed and oxygen become limited and 'stunting' of the population results (Noakes and Balon, 1982). Therefore, the separation of fry production and grow-out to improve the management and predictability of the harvest has been an important part of intensifying tilapia production.

Various methods to control reproduction in the grow-out system are reviewed by Guerrero (1982). Use of predators and/or stock selection can improve results under some circumstances, but monosex fry has greater applicability. Monosex production by handsexing and stocking only the faster growing males have been used with some success under simple conditions for many years and continues to be promoted in areas in which more sophisticated methods are unavailable (Someren and Whitehead, 1961; Loyshin and Pretto, 1983).

Hybrid fry production has been promoted for both reasons of reproductive control and improved culture characteristics but has been discarded on the first count largely because of the management difficulties intrinsic in the method (Lovshin, 1982). Fry yields from interspecific spawning are also lower (Lee, 1979; Mires, 1982). The basis of sex determination in tilapias is now believed to be polygenic and/or multiallelic

(McAndrew and Majumdar, 1983) and earlier hopes of achieving a completely homozygous parent stock by steroid treatment have not proved possible (Jensen and Shelton, 1979; Hopkins, Shelton and Engle, 1979; Obi and Shelton, 1983; Shelton et al., 1983). Direct hormonal treatment of undifferentiated fry steroids in the diet (Clemens and Inslee, 1968; Guerrero, 1975) has, meanwhile, become the main method in use for the production of all-male tilapia. The availability and cost of the purified hormone, plus alcohol to mix the hormone into a high quality feed, have constrained adoption of this technique in some places. The use of natural sources of testosterone such as ram's testes has, apparently been successful (Haylor, 1989). Scientific trials and practical experience have continued to improve the understanding of the critical parameters required hormone dosage, frequency of feeding, level of natural feed and age and size of fry under commercial conditions. However, poor results with sex reversal using techniques are still common even under experimental conditions (Guerrero et al., 1985; Nacario, 1987).

Earlier doubts as to the safety of the method (Shelton, Hopkins and Jensen, 1973) have now been largely dispelled (Fagerlund and Macbride, 1978; Johnstone, Macintosh and Wright, 1983; Rothbard, Solnik, Shabbath, Amado and Grabie, 1983; Goudie, Shelton and Parker, 1986).

The chief obstacle to its wider adoption now appears to be the difficulty of mass producing fry suitable for sex reversal on a commercial scale. The reproductive biology and behaviour of

mouthbrooding tilapias have been a major constraint but there has also been a lack of effort on the part of aquaculturists. Fish farmers and scientists alike have, until recently, approached tilapia fry production as relaxed observers, an attitude arising as much from the fishes fascinating sexual habits as from the ambiguity of a fish that spawns easily enough to be a problem during grow-out and yet refuses to produce fry freely in the manner of the carps.

The present methods of mass production of fry suitable for sexreversal, as used in Taiwan and Israel, are based on open-pond
spawning of large numbers of breeders; and they may not be
suitable in other places for management or economic reasons.
Pond production can be expensive in terms of land, water and
energy consumption; and fry production from smaller, less well
managed systems may be too unpredictable. More intensive
methods of fry production using egg removal and artificial
incubation, which have been successful on a experimental level
(Lee, 1979; Rana, 1986) have yet to be tried on a commercial
scale. Broodstock held in net hapas and tanks have been used
for fry production commercially in some parts of the world;
yields of seed per unit area may be much higher than pond
systems but broodstock productivity is usually far below the
levels obtained in the laboratory (Rana, 1986).

Predictable yields and improved control of seed production are of key importance if tilapias are to fulfil their role as the 'aquatic chicken' of tropical aquaculture (Maclean, 1984). Selection for desirable characteristics requires separation of

breeding from possible contamination, both genetic and disease. Furthermore the introduction of methods that produce large numbers of seed under controlled conditions is crucial.

Table 1.1 Status of Fry Production of Major Commercially Farmed Fish in Asia

			Grow-out	FRY PRODUCED BY Farmer Specialist		
Species	Importance of Broodfish wild seed ⁵		No mani- pulation required	Induced Spawning		
		of Broodi Ish		Environmental control	+Hormonal control	+High cost ⁴ nursing
Milkfish (<u>Chanos chanos</u>)	xxxx	wild, culture1	-	+	+	-
Penaeids	xx	wild, culture!	-	-	+	+
Sea Bass (<u>Lates</u> <u>calcifer</u>)	х·	culture	-	-	+	+
Grouper(Epinephelus tauvina)	xxxx	wild	-	-	+	+
Chinese and Indian Major Carp	x	culture ³	-	-	+	-
Snakehead(<u>Channa striata</u>)	xxxx	wild ¹ , culture ¹	-	-	+	-
Walking Catfish (<u>Clarias macrocephalus</u>) (<u>Clarias batrachus</u>)	x x	culture ² culture ^{2,3}	- -	-	+ +	+
Giant Freshwater Prawn (<u>Macrobrachium rosenbergii</u>)		culture ²		+	-	+
Silver Barb (<u>Puntius gonionotus</u>)		culture ^{2,3}	-	+	. +	-
Common Carp(Cyprinus carpio)		culture ^{2,3}	-	+	+	+
Snake-Skin Gourami (<u>Trichogaster pectoralis</u>)	x	culture ²	-	+	-	-
Nile Tilapia (Oreochromis niloticus)	x	culture ^{2,3}	+	+ .	-	+

Legand: ¹: experimental only, ⋅2: market ready fish, 3: specifically raised as broodfish 4: require expensive live feeds and/or prolonged labour-intensive nursing period. 5: x → xxxx (unimportant → very important)

1.2 Intensive hatchery production of commercial fish speciesthe tilapia as a special problem

The continuous availability of seed homogenous in quality and size is a prerequisite for intensification of aquaculture both for the improved management of traditional and more intensive fish production systems. However, many species of commercial fish and crustacea fail to spawn, or do not even achieve maturity under normal culture conditions (Table 1.1). Cultured marine species such as the milkfish, Chanos chanos and penaeid shrimps are examples of the former and their production until recently has been totally dependent on capturing fry and juveniles from the wild. In contrast, the problems that can have subsequent grow-out on uncontrolled breeding performance are well known for the freshwater tilapias because of their unique ability among farmed species to breed freely under normal pond culture conditions.

Other cultured species such as the common carp (<u>Cyprinus carpio</u>) or snake-skin gourami (<u>Trichogaster pectoralis</u>) do become mature under culture conditions and spawning can be stimulated by simple environmental manipulation. Some fish, such as the smaller carps and catfish (e.g. <u>Clarias macrocephalus</u>) are often easily obtained as ripe fish from the wild or grow-out farms. Other carps are more difficult to spawn; Chinese and Indian major carps mature at a large size, necessitating the keeping of broodfish which normally spawn only after hormone injection (hypophysation). These fish have been cultured for centuries but mainly in the proximity of the

major rivers in Asia which were the traditional source of wild fry; hypophysation was a necessary technical development before their widespread production became possible.

The controlled reproduction of commercial fish covers a range of complexity but each is divisible into four phases (a) broodstock maturation (b) production of fertilised eggs or spawning, (c) egg incubation and (d) early nursing of fry or juveniles. Although these events may occur naturally in the same environment, better results can be obtained if conditions are optimised at each stage.

The optimal environments for conditioning and spawning fish are often dissimilar. Thus, Dubisch ponds which are small earthen spawning ponds (Huet, 1972) traditionally used to spawn common carp in Europe are used to induce natural spawning and enhance egg and fry survival rather than condition broodfish. Further improvements in production can be achieved by intensifying the natural spawning of broodstock in concrete tanks. Isolation from disease and predators, plus greater control over water quality, can improve egg and early fry survival over that possible in earthen ponds (Jhingram and Pullin, 1988). Careful attention and feeding of selected broodstock become important if high productivity is required. The use of anaesthetics, hypophysation and closure of the female genital opening to improve synchrony of female ovulation, and dry fertilisation and the chemical removal of egg stickiness to permit up-welling incubation have made common carp production predictable and efficient in eastern Europe (Horvath, Tamas and Tolg, 1984).

Thus, separation of the reproductive phases, a technique now well advanced for some fish species, is following trends already established in intensive livestock production. The moves to intensify fish seed production have important economic consequences; in general, industry tends to follow economic rather than technical optima. Consequently, the motivation for technical development must have an economic basis. development and adoption of hypophysation techniques for fry production of <u>Clarias</u> <u>macrocephalus</u> in Thailand was almost certainly stimulated by high demand (= price) for the product and the inability to reproduce this fish by the simple methods used for the other cultured species of walking catfish, Clarias batrachus. Conversely, techniques for hatchery produced milkfish fry have developed slowly whilst the availability and economics of wild fry remain favourable. The rapid progress of the shrimp industry is stimulating the commercial maturation of brooders as prices of wild parent stock continue to rise and their availability decreases. The extra costs inherent in the development of live feeds for nursing juvenile shrimp and fish have been necessary because there is no viable or simple technical alternative.

The high costs inherent in more intensive fry production systems need to be counteracted by greater productivity and improved certainty or quality if the methods are to be adopted by fry producers. This is particularly true for fish such as tilapias that are already simply and easily produced. To use an

analogy, intensive production of broiler chicks evolved because of the improved profitability of hybrid varieties over traditional multipurpose poultry. Sex-reversed fry have the potential to play a similar role in tilapia culture.

A comparison of a range of hatchery systems for tilapia in one location under controlled conditions for both technical and economic performance was made in the present study. The theme of greater separation of the breeding process to improve efficiency of production was followed. A strain of Oreochromis niloticus was used that is known from electrophoretic analysis to be of high purity (Chitralada strain; Tangtrongpiros, 1988). The research involved long term trials to collect data suitable for direct commercial application. Productivity and spawning characteristics of individually identifiable fish monitored to give a better understanding of the dynamics of breeding and seed production in commercial-scale tank and hapa study involved the scaling-up of breeding systems. The intensive techniques known to work at the laboratory scale and their evaluation alongside conventional commercial methods. Intensive strategies for fry production were compared in terms of both technical and economic performance parameters; the experimental treatments compared were based on different levels of production cost and the economic aspects of intensive and 'improved' technologies for tilapia fry production formed a major consideration of this study.

Early harvest of seed from the mouth of female broodfish held in commercial size tanks and more intensive broodstock management techniques needed to complement this technique were investigated. Large nylon hapas were compared as an alternative method of fry production; small hapas have been used in research and commercially in the Philippines but a larger net size, commonly used by fry dealers in Central Thailand for holding fish, was evaluated. Hapas were compared on a level of management similar to that investigated for tanks using early seed removal and broodstock conditioning and exchange.

Earthen ponds representing a more extensive system were managed to produce swim-up, or yolk-sac absorbed, fry. The incubation efficiency for seed removed from different tank and hapa strategies was established to allow a valid comparison between systems in terms of swim-up fry production. Experiments with tanks, hapas and ponds sought to increase yield of seed whilst reducing costs. A 'commercial-scale' was sought at all times to eliminate errors of extrapolation and to allow a more realistic economic analysis.

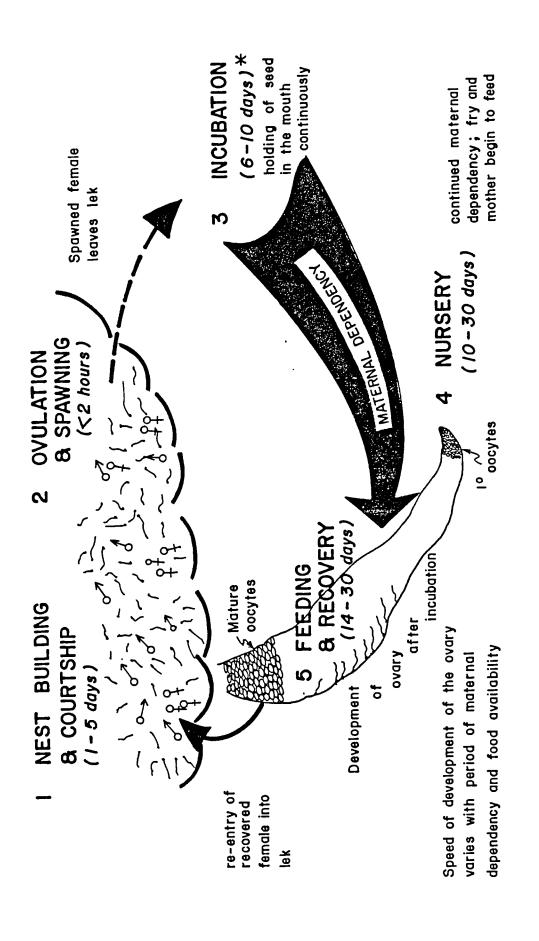
Finally, the technical performance of the various fry production strategies studied was analyzed economically based on a start-up operation in Central Thailand where tilapia is the most important cultured species. The addition and substitution of the range of 'improved' technologies was also modelled for several small-scale carp hatcheries in Northeast Thailand in which tilapia, despite high demand by grow-out farmers and consumers, is presently neglected by fry producers

in favour of native and exotic carps. The adoptability of the 'new' methods to produce a 'new' product, i.e. <u>Oreochromis</u> fry treated with the hormone 11-a methyltestosterone (MT), in rural areas was a central theme and the basis for final comparison of the different strategies for fry production.

The methodological approach used has relevance for the introduction of intensive tilapia fry production and MT-treatment in other parts of Asia.

Fig. 1.1

Natural reproductive cycle of <u>Oreochromis</u> indicating approximate duration and location of spawning and conditioning



Natural reproductive cycle of Oreochromis spp

* incubation period is temperature dependent

boundary of lek

1.3 A review of existing fry production systems

1.3.1 Reproduction under natural conditions

The reproductive behaviour of the <u>Oreochromis</u> species in natural fish communities, especially those in various African lakes, has been described by several authors (Lowe McConnell, 1959; Welcomme, 1969; Iles and Holden, 1969; Fryer and Iles, 1972; Fishelson, 1983). <u>Oreochromis</u> are maternal mouth brooders and are now classified by Trewavas (1983) as a separate genus within the tribe Tilapiini. The substrate spawners in the genus <u>Tilapia</u> and biparental mouthbrooders in the genus <u>Sarotherodon</u> are members of the same tribe and all three genera are described by the common name 'tilapia'.

Reproductive activity in <u>Oreochromis</u> is based around the lek (Fig. 1.1), an assembly of adult males which females visit solely for the purpose of copulation (Bradbury and Gibson, 1983). The lek has also been used to describe the natural 'arena' (Fryer and Iles, 1972) used for courtship by <u>Oreochromis</u>. It is akin to that used by many bird species, and is generally formed in a shallow area where individual 'nests' are excavated and defended by the males. Under natural conditions the margins of lakes are common sites for lek establishment. The size of the nest depends on each male's ability to defend it during the early stages of occupation (Fishelson, 1983), but may also be influenced by depth; De Silva and Chandrasoma (1979) reported that the size of nests of <u>O. mossambicus</u> increased with depth in a man-made lake in Sri

Table 1.2 Strategies used for the production of tilepis fry

Strategy	Xethod	Adventages	Disadvantages	Spawning system	Status
(a) Sy-product of culture	Fry and/or fingerlings removed from culture pands during and/or after production period	- Fry and/or fingerlings obtained as a bonus to grow-out fish; can be used to restock grow-out operation - No special facilities required	spawning and recruitment reduces	Earthen ponds	Yidespread
(b) Mixed age/size fr groduction	Tilapia broodfish are stocked in earth ponds fry by other fish and adult and fingerlings results in non-even and fryfingerlings tilapia cf. (a) sized fry; high rates of cannibal removed as required. Optimum broodstock — Improved broodstock productivity for best results (feeding, stocking density and ratio)		Earthem ponds	Yidespread	
(c) Even-sized fry produc- tion	Broodfish are stocked in spawning tank or sond. After short breeding period (14- 21 days) Broodstock and/or fry are removed for separate grow-om	- Reduced cannibalism between fry reduced necessity for grading - Reduced predation on tilapia fry by adult tilapia cf.(a) and (b)	 In earther ponds, broodstock removal is difficult, usually necessitates draining of pond Large concrete tanks facilitate removal of brooders 	Earthem ponds Static concrete tanks	Theiland (N.[.F.[.)
	Int. sekerace 31 ns. os.	- Further improvements in brood stock productivity	- Loss of eggs and young fry if me incubation facilities		
(d) Swim-up fry production	(i) Fry removed by fine seine netting. Broad	Fry produced suitable for sex reversal (i) Large numbers of fry are . cheaply produced suitable	(i) Loss of eggs and young fry if no incubation facilities available;	Earthen ponds Magas	Isrzel Philippines
,	fish are them restocked	for sex-reversal some resynchromisation of brood stock.	high sortalities during harvest in earthen ponds — interruption factor — high labour requirement during harvest.	·	
	(ii) Broodstock retained im pond or tank conti- nually. Fry removed by	(ii)Only cheap harvest equipment required - low labour requirment	<pre>(ii)Incomplete harvest of swim-up fry - cannibalism by uncaught fry</pre>	Earthem ponds Concrete tanks	Talvan Philippines
	dip metting or entrep- ment	 no interruption of spawners no incubation facilities required 	requires intermittent draiming and restocking - harvest of some older fry - grading required	Ārenas	Kenya
(e) Removal of- eggs and pre-released	- Check broodstock every 10-14 days and remove eggs & fry	- Fry and eggs recovered completely	- High capital cost	Tanks Hapas	Experimental
fry	- Restock brooders - Incubate eggs artificially	 Improved synchronisation of adults Improved productivity of females 	- Problems of incubation		-
(f) Removal of just-spawned	- Check broadstack daily for eggs	- Eggs recovered completely	- Yery high labour requirement	Small tanks Aquaria	
1998	- Incubate eggs artificial	y - Yery high female productivity	- High capital cost		Experimental
		possible			

Lanka. The period spent by the female in the vicinity of the male's nest may be brief. An ovulated female responds to the courtship display of the males and enters the nest; a transient pair bond is formed during which time spawning and fertilisation occur. The female lays eggs in up to twenty small batches (Trewavas, 1983) and may visit several different males during this period, which lasts 45 minutes to two hours. Ruwet (1963) reported this successive polyandry to be common behaviour although Trewavas (1983) described it as a rare phenomenon. Fishelson (1983) observed colonies of <u>O. aureus</u> males to frequently number over ten individuals in an area of around 30 m² and females to number up to 32 per lek-site.

After completing spawning, females leave the lek sometimes moving away a considerable distance to 'nursery grounds'; incubation of the eggs and fry may then continue for between 10-21 day (Baerends and Baerends van Roon, 1950).

Lowe McConnell (1983) discussed factors controlling tilapia reproduction under natural conditions and suggested that in stable environments such as large lakes, biotic pressures from predation, competition for suitable spawning grounds, nursery sites or food among juvenile fishes, may be particularly important.

1.3.2 Reproduction in culture systems

Methods

Oreochromis fry produced as a by-product from the main harvest of marketable table fish are often an important source of new

pond stock (Table 1.2; a). Yields of large fingerlings can be a particularly high proportion of the total production in pond systems in which predatory and competitive fish have been eradicated and food is abundant for the stocked fish. Edwards et al. (1986) recovered an average yield of 7 small fingerlings (5-10 g) and 4 large fingerlings (20-30 g) per m² in addition to an extrapolated gross marketable yield of 9t/ha/yr from small earthen ponds enriched only with duck manure and originally stocked at 4 fish per m².

This type of system can therefore generate surplus seed, but in places where predators or competitors are present fry production can be far more variable. For example, small numbers of predatory fish such as the snakehead (Channa striata) and eye-spot barb (Hampala dispar) were effective in reducing fingerling production to almost zero in some farmers' ponds stocked with Nile tilapia in Northeast Thailand (Edwards et al., 1986).

Fry production methods may be divided into those that produce fry of a suitable size and age for hormonal sex reversal and those in which the fry are harvested at a later stage (Table 1.2; a-c). Sex differentiation is believed to occur between 16 and 20 days post hatching in the mouth brooding tilapias (Nakumura and Takahashi, 1973) and treatment of fry with a hormone supplemented feed is therefore required during this period. Other advantages are also achieved by the use of more intensive techniques (Table 1.2; d-f).

Culture systems that produce fry suitable for MT-treatment

Table 1.3 A comparison of swim-up fry from the three major types of system (Pond, Hapa and Tank) using intensively managed broodstock in terms of different productivity measures

Types of system	Species	System of broodstock management	Production				
			average kg female/m²	fry/m²/ day	fry/kg female°/month	Source	
Earthen ponds Israe) (production)	F ₁ hybrids of <u>O.niloticus</u> and <u>O.aureus</u>	Broodfish stocked at 4000/ha. Remove swim-up and broodstock after 17-19 days (3 males: 4 females ratio)	0.69	3.98	1738	Rothoard <u>et al.,</u> 1984 80-100 day study	
Hapas USA (experimental)	O.niloticus O.niloticus	Maximum production in small hapas uses mixture of weight classes at 5/m². Remove mixture of eggs and yolk-sac and swim-up fry every 10-18 da (10 males : 2 females ratio) In hapas 21 day harvest	0.49 ays	29*	1778	Hughes and Behrends. 1984 70 day study	
	0,1111001000	- no exchange	0.23	41.74	5481*	Lovenin and	
		 female exchange male and female exchange 	0.25 0.25	45.0ª 51.9ª	2673* 3177*	Ibrahim, 1987 105 day study	
Static tanks Philippines	O.niloticus	Large static concrete tanks (100 m²) swim-up fry removed at edge 5 or 6 times/day Broodfish stocked at 3/m²	0.22	9.0	1277	60 day study (Guerrero and Guerrero, 1985)	
		(1 male:2 females ratio)	0.22	16.0	2182	20 day study	
Arena Kenya	O.aureus x O.niloticus hyorid	Large concrete arenas swim- up fry automatically removed into raceway. Broodfish stocked at 1.8/m². [1 maie: 6 females ratio.]	0.22	6.3	859	Baların and Haller, 1982	
Recirculating tanks Scotland	O.niloticus	Recirculating 2.25 m ² tanks. Broodfish stocked at 8.4/m ² . Remove fry and eggs every 14 d (1 maie:3.75 femaies ratio)	2.0 ays	78.3	1253	Baların and Haller, 1982	

refers to all seed (1.e. eggs, yolksac larvae and fry) harvested.

^{73.4} seed/m²/day harvested.

fry of all females used in the trial.

involve the harvest of swim-up fry (Table 1.2; d) or eggs and pre-released fry (Table 1.2; e-f); at present most commercial operations use the former system. Many studies however, shown the potential of egg removal strategies in intensive systems (Lee, 1979; Berrios-Hernandez and Snow, 1983). Removal of eggs and fry from broodstock managed intensively permits the condition of broodstock to maintained and reduces fry losses from cannibalism and predation. In addition, methods suitable for the production of swim-up fry for sex-reversal tend to be more efficient in terms of the usual parameters of system productivity (fry/m²/day and fry/kg female/month). There is, however, large variation in fry productivity between the systems producing swim-up fry (Table 1.3).

Systems

Tilapia have been bred successfully in spawning units of variable size and construction. Small glass aquaria have proven successful for behaviourial and genetic research (Neil, 1964, 1966; Rothbard and Pruginin, 1975; Silverman, 1978abc; Mires, 1982) whereas the main systems used commercially for producing fry are tanks, hapas and ponds.

Many tank designs have been used for tilapia fry production. The main distinction between tanks and ponds in this thesis is that the former are discrete from their immediate environment whereas ponds, even when lined with various materials, remain intimately associated. Tanks used for seed production range from converted plywood assault boats (Uchida and King, 1962)

and small plastic swimming pools (Shell, 1966), to purposebuilt fibreglass or concrete units.

The ideal characteristics of tanks as spawning units are generally the same as those required for any intensive culture unit (Balarin and Haller, 1982) but they may be adapted to allow self removal of fry (Haller and Parker, 1981) or have added substrate material to encourage nest building (Uchida and King, 1962). Tanks tend to have advantages in terms of space utilisation over earthen ponds and can, if water is recirculated or if the tanks are constructed as part of a larger agricultural system, be efficient in terms of water and energy use, whilst also being highly productive. Most tank systems to date however, especially those using recirculated water, have been either experimental and/or constructed in areas in which freshwater is limiting e.g. Hawaii (Hida, Harada and King, 1962), Kuwait (Hopkins, 1985) and coastal Kenya (Haller and Parker, 1981).

In general, tanks can be managed more intensively than earthen ponds with respect to water quality and broodstock. They have been advocated for production of hybrid fry which is problematic under pond conditions (Lovshin, 1982). However, there must be significant economic advantages associated with their use to cover the expected increased costs. Initial costs per unit area are inevitably higher than those for simple earthen ponds, as are the variable costs inherent in the greater water exchange and feeding required to maintain

breeding fish at higher densities.

A facility for water exchange, allowing maintenance of water quality and/or more efficient recovery of seed, is an important feature of most tank systems. When systems are small seed may be removed efficiently without draining, water quality being maintained with aeration and partial exchange of water. The ease with which the tank can be drained can allow tank fry production to be more manageable but in practice many systems fall short in this respect because of weaknesses in both design and management.

Cage culture of tilapia originated in experimental work carried out at Auburn University around 1970 (Coche, 1982). Since then it has become of major importance in the Philippines for growout and fry production in lakes and reservoirs. The lack of fry production in grow-out cages, due to inhibition of spawning and/or loss of eggs and fry through the mesh was seen as a positive feature for grow-out and fine mesh hapas became popular for fry production (Guerrero, 1977; Coche, 1982). Cages have also been used for nursing large fry after spawning and early nursing in earthen ponds (Campbell quoted in Coche 1982) and rectangular tanks (Coche, 1983). More recently net hapas have been used for the hormone treatment of swim-up fry (Buddle, 1984; Guerrero et al. 1985; Nacario, 1987; Berger and Rothbard, 1987).

Major advantages of hapas compared to ponds are (a) higher productivity per unit area (Table 1.3), (b) the possibility for landless sections of the community to participate in

aquaculture by operating hapas in communal waters (Beveridge, 1984) and (c) start-up costs can be relatively modest (Yater and Smith, 1985). However, to date most small-scale operators in the Philippines use ponds for fry production rather than hapa in ponds or in lakes (Guerrero, 1985). The risk of losses through pollution, decrease in natural productivity, theft, vandalism and typhoons have been quoted as possible reasons for this conservatism (Beveridge, 1984). The relative lack of interest by pond operators in intensification of hapa production is of interest as most small and medium operators use hapas for nursing swim-up fry (Guerrero, 1985).

Collection of swim-up fry from simple earthen ponds stocked with broodfish is the most common commercial method of production. Ponds may be managed for continual or batch production of fry (Table 1.2). Cost per unit size is low but so is the stocking density and output of fry (Table 1.3). The average pond size for producing swim-up fry varies from 200-400 m² in the Philippines (Guerrero, 1985), to 500-1000 m² in Taiwan (Liao and Chen, 1983) and up to 1 ha in Israel. The large units used in Israel allow considerable quantities of fry to be produced simultaneously for subsequent sex-reversal.

1.4 Major factors affecting <u>Oreochromis</u> fry production under culture conditions

1.4.1 Breeding intensity

The breeding intensity, i.e. the total yield of eggs spawned in a production system per unit area per unit time is dependent on both biological and environmental factors. Aronson (1949) claimed that for a fish with as complex a breeding behaviour as tilapia many important factors would have to be satisfied to induce even a ripe fish to breed. Moreover, the potential for stripping tilapia in a similar manner to salmonids or carps has not been demonstrated on any scale, largely because of this behaviour. As spawning is only possible if females show three 'on heat' signs simultaneously (Rothbard and Pruginin, 1975), it is likely to be very difficult to stimulate this condition in more than a few fish at a time. The use of hypophysation has also proved disappointing (Dadzie, 1970; Srisakultiew and Wee, 1988).

Density and sex ratio of broodstock during spawning have been found to have a particularly important effect on natural breeding intensity (Uchida and King, 1962; Hughes and Behrends, 1983). Ideally, the sex ratio should enable any female in the right physiological condition to be able to find a male and spawn. The stocking of a high ratio of females to males is common since males are polygynous and can spawn with several females on the same day (Lowe McConnell 1955, 1959; Fryer and Iles, 1972). However, several authors using different systems

have shown that increasing female to male ratio decreases total fry output (Mires, 1982; Hughes and Behrends, 1983). Mires suggested that 'male pressure' might be an important factor in increasing the intensity of breeding. A ratio of 1:1 (female:male) gave improved production during interspecific breeding to produce hybrids (O. niloticus x O. aureus), whereas Lovshin (1982) reported that a ratio of 1:2 was better than 1:1 or 2:1 in O. niloticus x O. hornorum hybrids.

Hughes and Behrends (1983) also indicated that an increase in stocking density in small hapas decreased seed production. Interruption of spawning during the period of intermittent egg laying by other broodfish is likely to be more frequent, and inhibition of spawning (Coche, 1982; Rana, 1986) is also known to occur, at higher densities. However, stocking density is highly system specific. High densities have been used successfully in tank and hapa systems in which environmental conditions can be maintained through adequate water exchange (Table 1.3).

Breeding intensity is also likely to be highly affected by broodstock condition and in particular the readiness of the females to spawn. Clearly, the condition of broodstock in any spawning unit is likely to be a function of a complex of factors. Age and size specific fecundities suggest that smaller fish produce a larger clutch size relative to body weight than larger fish and that the interspawning interval (ISI) can be slightly shorter (Lee, 1979; Siraj, Smitherman and Castillo-Gallusser, 1983).

Hughes and Behrends (1983) found that broodfish of mixed weight class produced more seed than large and small weight class broodstock raised separately in small hapas. The ISI of individual females can also be considerably reduced by loss of their egg brood (Fishelson quoted in Peters, 1983; Dadzie, 1970; Lee, 1979; Rana, 1986).

The spawning frequency and output per female may be increased by egg removal in intensive systems. Presumably egg removal limits the loss of body condition that normally occurs over the natural period of incubation and nursing when a female's feeding is restricted.

Environmental factors are important in the control of spawning activity under both natural and culture conditions. Cichlids can have predictable restricted spawning periods even in equatorial regions (Bye, 1984). Trewavas, 1983 (quoting El Zarka et al., 1970), reported that during the peak spawning season in the Nile Delta, over 33.5% of the catch were ripe fishes. Hyder (1970) observed that sunlight and temperature were the most important factors influencing development of the gonads and subsequent spawning of T. leucosista in Lake Naivasha. Rainfall was reported to stimulate breeding activity in different species of tilapia (Lowe McConnell, 1959), although both Aronson (1957) and Hyder (1970) indicated that heavy rains, together with reduced light, checked breeding and caused retrogression of the gonads. More subtle changes in the environment may also be important where seasonal differences are minimal. Lunar periodicity of spawning has been reported in

both riverine (Schwanck, 1987) and lacustrine environments (Okorie, 1973), with observations showing an increased proportion of fish breeding during the full moon.

The introduction of Oreochromis spp. to areas outside their natural geographical range has increased the importance of environmental factors in controlling the production of fry. Hatchery output in Taiwan and Israel is limited to the warmer months after which broodstock are overwintered until the following year (Liao and Chen, 1983; Rothbard et al., 1983). The spawning period is restricted to only four months in Israel (May-August) because fry must be large enough to survive overwintering (Mires, 1982). Behrends and Smitherman (1983) found that gonadal development and spawning frequency of four species of Oreochromis increased as average daily water temperatures increased from 20-28°C. Broodstock condition, as indicated by gonadosomatic index, became less variable between fish as environmental conditions approached the optimum. Bautista, Carlos and San Antonio (1988) reported that seasonal fluctuations in temperature, rainfall and water quality affected fry production in hapas and tanks in the Philippines. Low dissolved oxygen associated with algal blooms and die-offs corresponded with poor fry production in hapa nets.

1.4.2 Incubation success

Intensive breeding must be followed by successful incubation if large numbers of fry are to be produced. Both natural and artificial incubation suffer losses such that total fecundity, or the number of eggs per fresh clutch (Rana, 1988) is an unrealistic measure of the level of commercial fry production possible.

The type of losses that are likely to increase with density (both fertilised dependent) include eggs (density unfertilised) stolen by other broodfish during spawning, poorly fertilised clutches and cannibalised eggs. Peters (1983) that small numbers of eggs in both male and female stomachs were a common occurrence, although he did not document the breeding condition of the fish. The natural behaviour of female fish to seek isolation from the breeding lek perhaps also indicates that egg cannibalism is commonplace. Breeding systems using lower stocking densities, and those such as the Baobab spawning arena in Kenya (Balarin and Haller, 1982) which allow separation of spawned females, may improve the success of natural incubation. Poor fertilisation of eggs has been related to successive spawning by individual males (Rana, 1986) and such negative effects of polygyny are likely to be exacerbated at high female: male ratios. Clutches of eggs of low fertility would tend to yield poor results whether incubated naturally or artificially.

Hatchability of eggs may also vary between fish of different types. Siraj et al. (1983) observed differences in hatchability between eggs of different age classes of female fish with older fish having significantly better yields than younger individuals. Rana (1986) observed that fertilisation rates varied from 4-100% and young broodstock (6-9 months) had

particularly poor rates of natural fertilisation (<60%). This contrasted with far less variable rates for manually stripped fish (range 83-100%, mean 93.7%), suggesting that younger fish suffer lower hatchabilities primarily because of less efficient fertilisation during natural spawning. This is possibly compounded by inexperienced mouth brooding by young fish. The phenomenon of egg reabsorption in <u>Oreochromis</u> as observed by Peters (1983) is common in both wild and captive fish. Females in which reabsorption of eggs has already begun may still spawn but the mortality of embryos is very high. Some types of broodstock management may increase the occurrence of reabsorption of eggs, by preventing females with ripe ova from spawning and this is likely to reduce incubation success.

Artificial incubation of <u>Oreochromis</u> eggs has yet to be realised as a viable commercial method. Low and/or variable survival, especially of eggs less than 48 hours post fertilisation (Lee, 1979) has reduced many of the potential productivity benefits from frequent harvest of eggs.

Mechanical trauma of eggs in artificial incubators associated with poor system design has been identified as the major problem causing poor hatchability (Hempel, 1979; Rana, 1986) but this can be compounded by bacterial and fungal infection of damaged eggs (Subasinghe and Sommerville, 1985; Suliman, 1987).

A variety of systems has been used to agitate the heavy, yolky eggs of Oreochromis to mimic natural incubation. The best

results to date have been achieved in down-welling incubators with recirculated, ultra-violet (UV) light sterilised water in which hatchabilities in excess of 80% of just-fertilised eggs have been achieved (Rana, 1986; Suliman, 1987).

Other types of incubation system have yielded poor results for eggs taken from the mouth just after fertilisation (Shaw and Aronson, 1954) but the poor performance of these types of incubators is difficult to interpret as details of fertilisation rates and bacteriological quality of the water are rarely given (Rana, 1986).

1.4.3 Harvest efficiency

Swim-up fry suitable for MT-treatment are physically fragile and under natural conditions may still show dependency on the mother fish. The buccal cavity is known to be an attractant to young fry for another 12-16 days after first release from the mouth (Baerends and Baerends Van Roon, 1950; Neil, 1964; Russock and Schein, 1977). This extends well into the period required for MT-treatment. In practice, swim-up fry have been harvested from 10 days after stocking breeding fish in tanks (Berrios-Hernandez and Snow, 1983) or after 12-14 days in ponds (Liao and Chen, 1983; Guerrero, 1985). After successful spawning and incubation, losses of eggs and fry during harvest from the system can be high in seed production systems. Seining and complete drainage to remove all swim-up fry after a spawning interval of around 20 days in large earthen ponds led to losses estimated at 30% of the total harvest (Rothbard et

al., 1983). Partial removal of fry may be satisfactory in more extensive systems but the presence of large numbers of unharvested fry in any system has a negative effect on subsequent production as they cannibalise succeeding groups of fry (Liao and Chen, 1983).

Recruits will also subsequently compete with broodstock for space and food, further reducing broodstock condition and spawning activity. Therefore, a complete harvest of seed has advantages but it is dependent on the breeding system being drainable.

Table 1.4 Summary of the scope of research

Main Experiments		System			Duration
		Tank	Нара	Pond	(days)
-	The effect of intensive broodstock management on egg and yolk-sac fry production	Т1	-	_	155
_	The effect of conditioning environment & female broodfish exchange strategy on egg and yolk-sac fry production	Т2	H1	-	116
-	The effect of duration of female conditioning, selective female exchange & male exchange on egg and yolk-sac fry production	-	Н3	-	106
-	The effect of broodfish density on egg and yolk-sac fry production	Т3	Н2	-	201
-	The effect of harvest intensity on swim-up fry production	-	-	P1	116
_	The effect of broodfish size on swim-up fry production	-	-	P2	105

1.5 Scope of research and nomenclature

A series of experiments was carried out in three types of system:

- (a) concrete tanks supplied with recirculated water (T). Tank area = 12.57 m^2 .
- (b) nylon hapas suspended in an earthen pond (H). Hapa area = 40 m^2 , earthen pond area = 1740 m^2 .
- (c) in earthen ponds (P). Earthen pond area = 1740 m^2

The initial experiment (T1) in concrete tanks only, investigated the effect of broodfish disturbance and exchange of female broodfish on seed production. The effect of increasing frequency of harvest was also observed.

A further experiment compared the production of seed from tanks (T2) and hapas (H1) after exchange of females conditioned in either tanks or hapas for a period of 10 days.

A third experiment evaluated the duration of female conditioning before use in spawning hapas, the use of selective female exchange based on optimising the breeding condition of females in the spawning hapa and male broodfish exchange (H3). Seed production in tanks and hapas at different broodfish stocking densities was investigated in a further experiment (T3 and H2 respectively).

Pre-maturation production of broodstock under different quality supplementary feeding regimes was analyzed (Section 3.1) after observing subsequent spawning frequency of broodfish in

concrete tanks (T3).

Two sequential experiments in earthen ponds compared swim-up fry production at two harvest intensities (P1) and using broodfish of two size-classes (P2).

A series of incubation trials (Chapter 5) then allowed a comparison of seed production from tank and hapas with earthen ponds on a systems basis (Section 5.5). An economic analysis of the various treatments within experiments was then modelled to develop production strategies for MT-fry in Central and Northeast Thailand (Chapter 6).

The main experiments are summarised in Table 1.4.

Experiments are described using a three digit code. The first (T,H,P) refers to the system i.e. tank, hapa and pond respectively. The second digit describes the experiment i.e T2 is tank experiment 2. The third digit describes the treatment within the experiment i.e. T23 is treatment 3 of tank experiment 2.

CHAPTER 2

BROODSTOCK EXCHANGE STRATEGIES IN TANKS AND HAPAS

2.1 Background

Intensified broodstock management is a way of improving the productivity of tilapia seed production in tanks and hapas. The major objective of broodstock management is to maintain breeding stock at their most productive level. Ideally the stocking of breeders into any seed production system should coincide with the beginning of this productive period and their removal at the end. The concept of commercial lifetime is an important production parameter well recognised for livestock but it has been rarely considered or derived empirically for most species of fish.

In contrast to warm-blooded livestock, fish continue to grow throughout their lifetime (Bagenal and Tesch, 1978). Since the number of eggs obtained from a fish is related to body size and/or age (Rana, 1986), the unit output of a breeding female continues to increase with time which is a major disincentive for replacement of broodfish. The advanced reproductive strategy of tilapias distinguishes them from most other cultured fish; studies using small numbers of fish under controlled conditions have shown considerable improvements in productivity of breeding fish with intensification broodstock management (Macintosh and Sampson, 1985). This is undoubtedly related to their mouthbrooding habit and, by classical definition, their low fecundity. Since much larger numbers of broodstock are required for an equivalent output of fry than, for example carps, and spawning is asynchronous and virtually non-seasonal under tropical conditions, there is

great incentive to manage broodstock to optimise individual productivity and breeding synchrony. Furthermore, although the number of eggs produced increases with fish size (Lowe McConnell, 1955; Peters, 1959) it does so at a slower rate than for non-mouthbrooding fish and consequently the advantages of using very large broodfish are reduced.

Broodstock management in most tilapia hatchery systems is simple and generally relies on the continuous use of the same fish throughout the period of production. Broodstock are replaced at various intervals of time and presumably at various ages, usually after a noticeable decline in fry production Yater and Smith (1985) reported that broodstock occurs. replacement in hatcheries in the Philippines is more frequent on larger farms with more intensive management (every 15 months) than smaller farms with poorer productivity (every 21 months). In Taiwan broodfish are selected at 2 months of age from the time the fish first spawned in the season; they are raised for the rest of the year and used as broodfish for a further two years. During this production period the fish breed a total of about 12-14 times (Liao and Chen, 1983). In Brazil, broodfish are replaced annually as fry production declines by around 50% over the same time (Lovshin, 1982).

The gradual decline in fry production in commercial systems seems in contradiction with the increase in clutch size with female age and size observed in aquaria. The reasons are inevitably interactive and systems specific. Broussard et al. (1983) explained the decline in fry production with time in

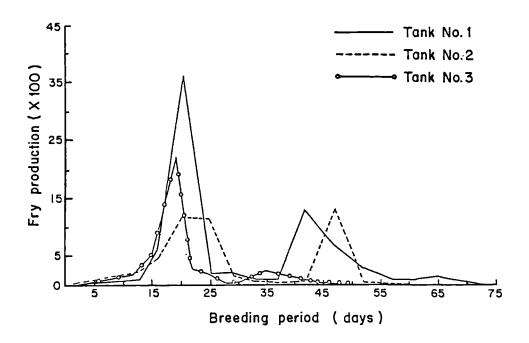
earthen ponds as cannibalism-related mortality rather than a consequence of poor broodstock condition, as stocking density was low and breeders continued to be found incubating eggs and fry.

Over extended periods of time fry production may also be affected by a decline in frequency of spawning. It appears that breeding by larger fish, especially in extensive systems, may become less frequent. Even when broodfish are managed more intensively, Lee (1979) recorded some increase in interspawning interval (ISI) with time, although Rana (1986) found little change with successive spawns.

Decline in fry production with time is often paralleled by a deterioration in water quality. Poor water quality could affect fry production at any stage i.e. broodstock condition, spawning activity, egg survival or fry survival. The reasons for a drop in fry production are thus complex and probably interactive. However, the decline in broodstock condition following spawning and extended mouthbrooding, is probably the single important reason for depression in fry yields in most systems. Reproduction exacts a high demand on the body resources of any animal. In times of poor food supply female mammals can still produce and raise their young by depleting body reserves, but under intensive farming conditions feeding standards must be high to sustain reproductive output (McDonald et al., 1981). Naturally, oral incubation imposes an even higher burden on breeding Oreochromis as feeding is inevitably disrupted during this period of 8-20 days (Fig 1.1).

Fig. 2.1

Fry production in large static water concrete tanks (Area = 100 $\rm m^{4}$) with production period in the Philippines



Relationship of Oreochromis niloticus fry production and breeding period in concrete tanks (after Guerrero & Guerrero 1985)

Poor broodstock condition as a major cause of declining fry yields is suggested by the breeding performance of fish in small intensive systems in which eggs are removed shortly after spawning. It is known that individual fish will continue to spawn frequently for periods of more than 6 months if egg removal is practised (Lee, 1979; Rana, 1986). In terms of Interspawning Interval (ISI), females have spawned every 7-24 days in small tanks and aquaria compared to 30-60 days in ponds (Rana, 1988) (Fig 1.1).

Decline in fry production can be observed over a much shorter period and this supports the view that broodstock exchange could be used as a management technique to maintain active breeding in any intensive spawning unit.

Guerrero and Guerrero (1985) reported an early peak in fry production within a 70 day production cycle. Their interpretation of the results indicated that productivity over the early part of the cycle (20 days) was nearly twice that over the whole cycle. The data suggest that the system's productivity could be maintained at the initial high level if the broodstock were exchanged with a rested group of fish (Fig 2.1).

Oogenesis is known to be very flexible in <u>Oreochromis</u> and to take place over a matter of days or weeks under suitable conditions (Iles, 1973); the exchange of spawned fish need therefore be only temporary.

The large peak in fry production observed by Guerrero and Guerrero (1985) also indicated that a high proportion of the fish spawned around the same time i.e. that breeding was synchronised.

The possibility of improved synchrony of spawning in Oreochromis indicates that some of the benefits of intensified broodstock management illustrated on a small-scale may have potential with larger numbers of broodstock on a commercial-scale. If stocked fish spawned synchronously, in theory the harvest interval could be reduced further and the seed harvested directly from the mouths of the females as eggs or larvae.

Induced spawning in many fish species has allowed control of ovulation and seed production to a degree not practically possible for most livestock (Sorensen, 1979). However, the use of hypophysation, routine in the production of carp seed (Pullin and Jhingran, 1985) have not proved successful with tilapias. Dadzie (1970b) reported induced spawning of <u>O. aureus</u> with human chorionic gonadotropin (HCG) but female <u>O. niloticus</u> were not responsive to Chinese carp and Indian major carp pituitary gland extracts or HCG over a large range of doses; and spawning levels were lower than controls (Srisakultiew and Wee, 1987).

Reproduction in individual <u>Oreochromis</u> under hatchery conditions is frequently asynchronous (Jalabert and Zohar, 1982) but there is evidence to suggest that breeding is synchronised under some conditions. Lee (1979) noted that

females in the same aquarium typically spawned on the same day and attributed this to mutual stimulation and inducement. The descriptions of lek behaviour in nature by Fryer and Iles (1972) and Fishelson (1983) suggest that breeding activity can be intense with large numbers of fish spawning per unit time. Thus, females will readily spawn with males provided that the right environmental and social conditions exist and the female is in breeding condition.

Synchrony of readiness to spawn within a group of females necessitates a homogeneous stage of oocyte/egg maturation. Srisakultiew and Wee (1988) reported improved synchrony of breeding when ripe broodstock were cold shocked for six hours prior to release in spawning hapas; the broodstock had previously been conditioned in tanks in single sex groups. Guerrero and Guerrero (1985) similarly separated sexes and raised the fish with intensive feeding (3% BW/day) prior to their use in spawning tanks. Pre-conditioning in this manner may be a necessity in places where fry production is seasonal. In Taiwan where low temperatures restrict fry production for part of the year, broodstock are overwintered in single sex groups before stocking in earthen ponds.

The separation of broodstock by sex as a method for bringing Oreochromis into spawning condition in present practice has a sound basis, but whether ovulation and resorption of eggs can be prevented is unknown. The resorption of ripe eggs is related to the lack of opportunity to spawn and it is believed that Oreochromis can only maintain such eggs for approximately one

week (Peters, 1983). Therefore, the duration of the conditioning period and selection for replacement are likely to be critical parameters in any successful replacement strategy. Female <u>Oreochromis</u> may attain and maintain breeding condition in the absence of males (Silverman, 1978a), to the degree that ovulation and spawning may occur between two conspecific females. Effective conditioning of females would therefore require stocking densities that would prevent the territory establishment that initiates the final stages of courtship and ovulation (Coche, 1982; Balarin and Haller, 1982).

The separation of spawning and non-spawning phases is a natural phenomenon amongst many species of fish, especially those producing large batches of eggs annually or seasonally. Most important commercial species of fish spawn easily only once a year (Burt et al., 1988) and the emphasis of hatchery development is towards extending the breeding season with the economic incentive of fry for sale earlier in the season. In monsoonal Asia, the onset of rains is synchronised with peak demand for freshwater fry for stocking in rain-fed culture systems. Fry producers who can spawn fish early in the season have a large competitive advantage. Early and/or extended production of fry is also an advantage for fish species unconstrained by season for the grow-out phase such as the white sea bass (Lates calcarifer) in Thailand.

Although tilapia spawns aseasonally in the Tropics an improvement in control of fry production by separation of broodfish conditioning and breeding could have a major effect

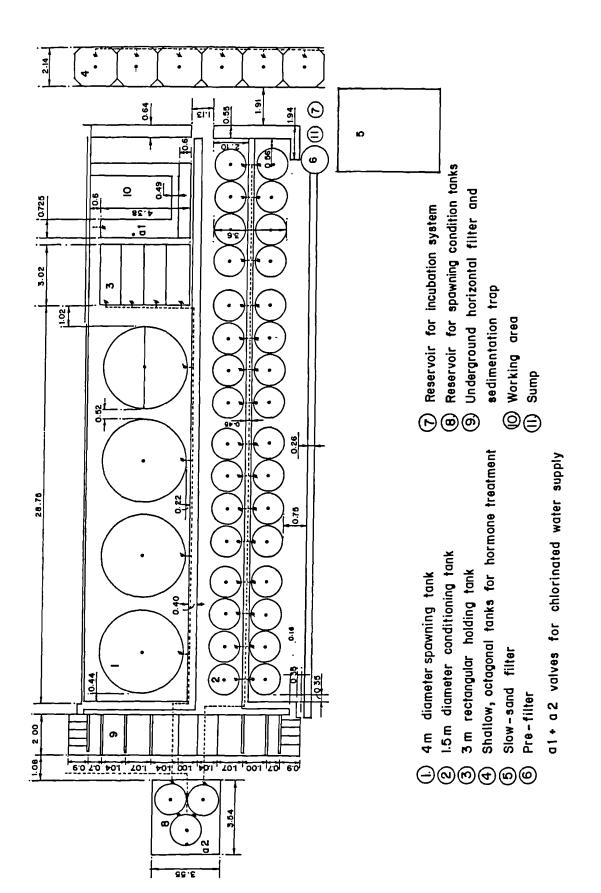
on the economic viability of hatchery operations. Year round propagation of fish and sales of fry could yield a better return on capital investment, as in salmonid culture (Bromage and Cumaranatunga, 1988). The demands on equipment and labour that can be limiting on the production and marketing of fish during a seasonal glut can also be reduced.

The holding of large numbers of <u>Oreochromis</u> breeders as single sex cohorts has important economic consequences. The optimum stocking density is much higher for most carps or other species that naturally spawn as a shoal, with free release of gametes into the water, than in the tilapias in which reproductive behaviour may be inhibited at a relatively low density (Balarin and Haller, 1982). However, such low densities are not required for conditioning fish as mature females can be obtained from tanks and aquaria systems stocked at very high densities. Thus, the total capacity of breeding fish within a hatchery of a given area or volume of water could be much higher in systems using replacement strategies.

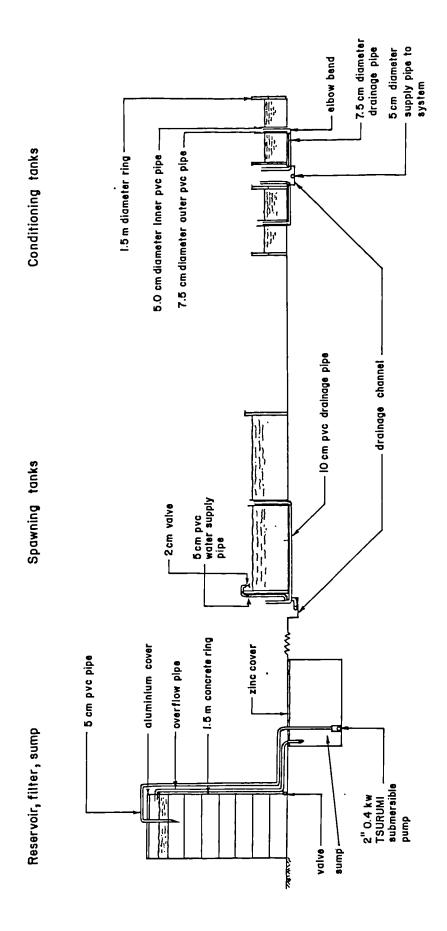
The effect of broodstock exchange on both individual and system seed production and synchrony of spawning were investigated in the present study. Body condition and growth of broodfish held under intensive management regimes in tanks and cages were also monitored to enable a better understanding of the interaction between growth and reproduction. The interval between harvests (Interharvest Interval, IHI) chosen was much shorter (5-10 days) than that of previous studies (Table 1.3) to prevent prolonged natural incubation. Larger numbers of fish were used

in exchange treatments to allow conditioning and spawning to occur simultaneously.

Plan of the EEC Tilapia Hatchery, Asian Institute of Technology, Bangkok, Thailand. Dimensions in metres.



Cross-section of the main components of the EEC Tilapia Hatchery showing water supply and filtration, spawning and conditioning tanks



2.2 General methodology

The experimental systems in which <u>Oreochromis</u> fry production were investigated i.e. concrete tanks, nylon hapas and earthen ponds are described in this section (2.2.1) together with the experimental methodology used (2.2.2) for tanks and hapas. Experimental methodology for earthen pond production is given in 4.2.

2.2.1 Experimental system design

2.2.1.1 Concrete tanks within a recirculation system

The experimental system used for spawning and conditioning broodfish was located within the EEC Tilapia Hatchery on the campus of the Asian Institute of Technology (AIT) (Fig 2.2). The concrete tank system was located along a North-South axis and sheltered by a galvanised iron roof approximately 3 m above the tanks. The tank environment was thus protected from direct sunlight, rainfall and wind except under the most extreme conditions. The spawning and conditioning tanks were located in separate but similar recirculation systems. The four large circular concrete tanks of 4 m diameter received clean water from a concrete ring header tank (Fig 2.3) via a 5 cm diameter PVC supply pipe. Wastewater flowed from the concrete tanks by way of a double central standpipe to an open channel which finally drained to a horizontal settling tank, biological filter and sump. Water was continuously pumped from the sump into the header tank; a constant head of water pressure and

Cross-section of horizontal filter used in spawning and conditioning systems of the EEC Tilapia Hatchery

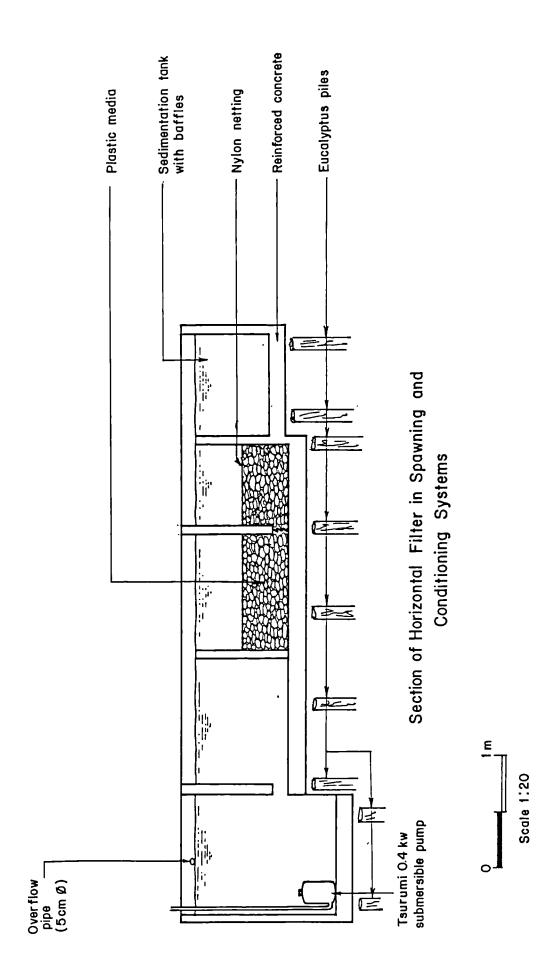


Table 2.1 Physical parameters of the recirculated water tank system in the EEC hatchery at AIT used for tank experiments (T1, T2, T3, H1)

1	al water Lume (%)	Total water contact (%) surface area		Residence time (min)
Spawning tanks Discharge channel Settling tank Biofilter Pumping chamber Header tank Pipework	77.9 4.3 1.5 4.6 1.1 7.7 2.9	7.8 1.1 0.5 88.7 0.4 1.0	115 115 115 115 220 220	438 24 9 26 <1 23
Total (1) (m ²)	64698	1365		
Conditioning tanks Discharge channel Settling tank Biofilter Pumping chamber Header tank Pipework		1.3 1.2 0.5 96.2 0.4 1.0 0.4	95 95 95 95 220 220	89 29 10 31 <1 23
Total (1) (m ²)	22788	1259		

aeration were provided by overflow back into the sump.

Spawning Tanks

Four circular spawning tanks were constructed with small, traditional Thai bricks covered with plaster reinforced with stretched construction wire (Fig 2.3). Standard concrete blocks were placed perpendicular to the wall at intervals of 16 cm to act as nest sites for the males. A circular flow of water was maintained at 29 l/min giving a complete change of water every 7.3 hours (Table 2.1). A small glass window in the side of the tank allowed lateral observation of fish behaviour.

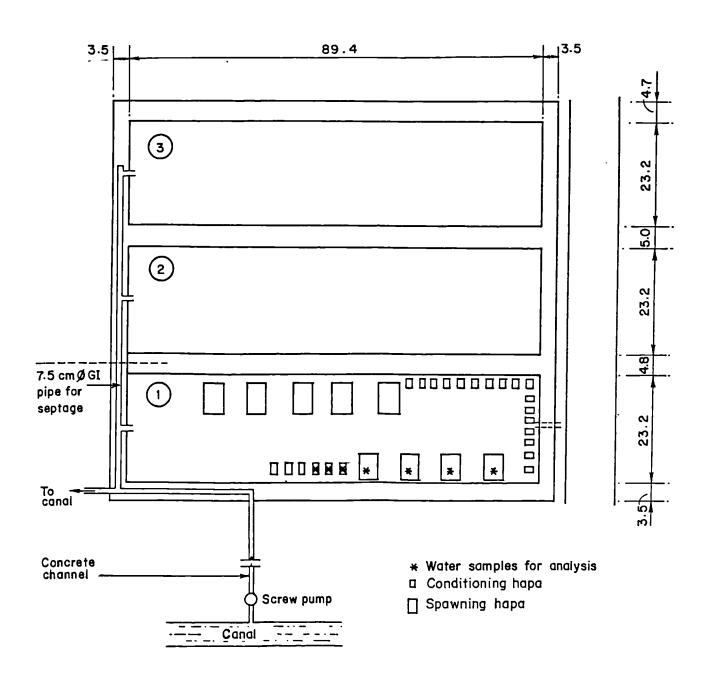
Conditioning Tanks

Tanks used for conditioning fish were made from two concrete reinforced rings (diameter 1.5 m) plastered together on top of a reinforced concrete base. A total of 32 tanks were located in the recirculation system but only 8 were used at any time for this series of experiments. Water flow was maintained at 12 l/min giving a complete change of water every 89 minutes (Table 2.1).

Filter and Water Supply

Water was recirculated in both systems without additional topup water for a complete interharvest period (5 or 10 days). At harvest, most of the water in the spawning and conditioning tanks was drained to waste and freshwater was added from a chlorinated deep groundwater supply to the head of the rear

Plan of earthen pond and hapa-in-pond experimental facility at AIT. Dimensions in metres.



water supply tank (a2, Fig. 2.2) and the top of the drainage channel (a1, Fig. 2.2). The horizontal filters were identical for both systems (Fig.2.4).

Primary sedimentation was achieved in the first compartment fitted with sloping asbestos plates as baffles (Fig. 2.2) before water flowed through a biological filter into a deeper sump. Washed plastic bottle tops and other assorted media were held in place with nylon netting/bamboo frame; this material had a void space of 0.62 l per 1 l of filter material and a total filter area of 1 m^2/l (du Feu, 1987). The residence time of the water in the biological filter was approximately 26 minutes. Water was pumped to a concrete header tank from the sump by a 0.4 kw submersible pump fitted with a PVC pipe (5 cm diameter). Hydraulic parameters for the operation of the spawning and conditioning tank systems are given in Table 2.1.

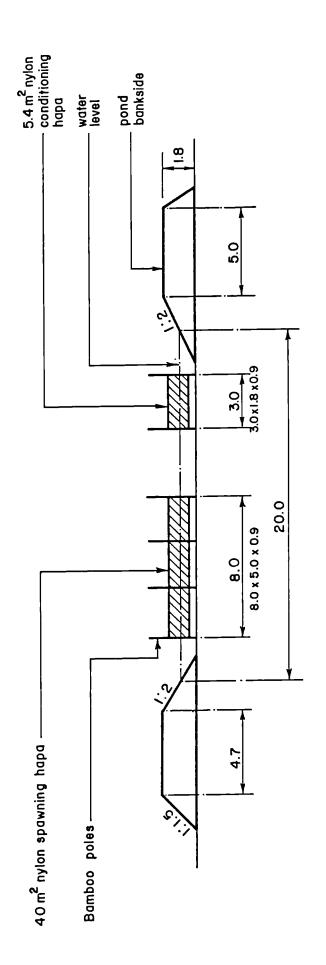
2.2.1.2 Nylon hapas in earthen pond

Earthen Pond

Hapas for spawning and conditioning were positioned in a single rectangular 1740 m^2 earthen pond (Fig. 2.5). A water level of 1 m was maintained by topping up from the AIT canal and overflow via a concrete pipe.

The pond was fertilised with either septage (sts) and/or compound chemical fertiliser (NP) on a twice weekly basis to encourage eutrophic conditions.

Section of design of hapa-in-pond experimental facility at AIT. Dimensions in metres.



Nylon Hapas

Standard, commercially available hapas locally fabricated from knotless nylon (mesh 1 mm:20 grade) were used. Hapas of 8.0 m \times 5.0 m \times 0.9 m, a size used by commercial fry dealers for holding fry, were found suitable for spawning hapas; smaller hapas (3 m \times 1.8 m \times 0.9 m) were used for conditioning broodstock. The net hapas were suspended in earthen ponds by attachment with nylon twine to bamboo poles (Fig 2.6).

The bottom of the net was anchored at each corner and half-bricks were used to prevent the centre of the net from floating to the surface. Nets were positioned to allow a maximum free-board of 40 cm above the water level. Spawning hapas were placed equidistant along the longitudinal axis of the pond such that replacement nets could be inserted between them (Fig 2.5).

2.2.2 Experimental methods

2.2.2.1 Concrete tanks

Water level was maintained at 1 m for the duration of each experiment except during total harvest of eggs and fry when the water level was dropped to 20-30 cm to facilitate capture of broodfish. The total harvest of seed required that each fish be caught in a double net, held gently in a gloved hand and mouth opened and inspected for eggs and fry. If present these were washed out by moving the head of the fish backwards and forwards through the water. Free swimming fry, if present, were

caught in a single fine-mesh net whilst draining water to waste. Surface algae and detritus were removed from the base of spawning and conditioning tanks after removal of broodstock and seed.

Broodstock biomass, growth and condition were estimated by measuring fish before and after spawning and conditioning. All fish were first anaesthetized in aerated water using benzocaine (Ross and Geddes, 1979).

Spawning and conditioning tanks and nets of different treatments were changed spatially throughout the experiment to randomise variation due to environmental factors.

Broodfish in conditioning and spawning tanks were fed to appetite with a floating catfish pelleted feed (Chareon Pokaphand, Bangkok; crude protein content 30%) three times daily; each meal comprised an initial amount which was approximately 50% of the previous amount fed, followed 10 minutes later by a second portion. The feeding of the second portion was dependent on the first being completely consumed, indicating an active feeding response.

Water quality conditions were monitored by weekly measurements of dissolved oxygen, pH, total ammonia and nitrite using standard methods (see Appendix 1). Temperature was recorded daily using a maximum-minimum thermometer.

2.2.2.2 Nylon hapas

Eggs and fry were harvested every 5 days and broodstock managed according to the treatment and experiment protocol.

The bottom strings of the spawning nets were untied before broodstock were concentrated at the pond side end of the net using a large bamboo pole that was slid under the net. Broodstock were then removed one at a time and the females checked for eggs and fry. Eggs and fry were initially washed into plastic bowls as individual clutches. Harvested fish were placed in floating net hapas before being lifted onto the pond side for weight and length measurement under anaesthesia. The separation of males, females with seed, and females without seed at harvest allowed growth and condition to be analyzed independently for each group.

All nets were replaced every 10 days to avoid water quality deterioration inside the net due to fouling. The nets were removed from the water and cleaned well with a brush and hose before reuse. The spatial arrangement of different treatments relative to one another was altered at each net change to avoid the effects of any local differences in water quality within the pond.

Water quality parameters, pH, total ammonia, chlorophyll a and dissolved oxygen at dawn were measured weekly both inside and outside the nets using standard methods (Appendix 1). Temperature was measured daily using a maximum-minimum thermometer suspended 30 cm below the water surface.

2.2.2.3 Seed storage and analysis

Eggs and fry were preserved and analy as discrete clutches for studies on individual tagged fish; otherwise clutches from the same treatment were combined by stage and analy collectively. Seed were staged at harvest according to development into (1) uneyed eggs, (2) eyed eggs, (2-3) imminent hatching, (3) yolksac fry and (4) swim-up or yolk-sac absorbed fry. Eggs and fry were stored in 4% formaldehyde (Peters, 1983) in sealed glass or plastic bottles.

The numbers and average weight of eggs and fry were estimated by weighing a known sample number of eggs and fry of the same batch and then bulk weighing the total batch on an electronic balance (Mettler Pe3600) to two decimal places. Tuan (1985) demonstrated the greater accuracy of this method compared to a volumetric method for enumerating large numbers of Oreochromis niloticus eggs and fry. The method was used for both single and combined batches of eggs and fry although no more than 25 g of eggs or fry were weighed in any one batch.

Three samples of 100 eggs/fry were counted manually and weighed separately after dabbing to remove moisture on a piece of dry absorbent tissue for 10 seconds whilst held in a fine-mesh nylon net. Batch samples were treated similarly after allowing 15 seconds for moisture removal.

Table 2.2 The experimental design for investigating the effect of exchange of female broodstock on spawning in concrete tanks (T1)

	1	Broodfis	sh		-	Brok			
System/ Experiment/	Number of fish		Category	Restocked to spawning		Exchanged from conditioning		Interval (days)	
Treatment	per spawning	Total		Tank	Нара	Tank	Hapa	Spawning	Cond.
T11	40	40	males	х					
	40	40	spawned females	x				10	
	40	40	unspawned females	x					-
T12	40	40	males	x					
	40	40	spawned females			x		10	10
	40	40	unspawned females			х			
T13	40	40	males						
	40	40	spawned females		UNDIST	TURBED		125	
	40	40	unspawned females						
T14	40	40	males.	х			ļ		
	40	60	spawned females			x		10	10
	40	00	unspawned females	x					

Cond. = Conditioning

x denotes restocking and conditioning strategy

2.3 The use of intensive broodstock management strategies for seed production in concrete tanks

2.3.1 Experimental methodology

(a) Origin and pre-treatment of broodstock

Broodstock were obtained from a large earthen pond (1740m²), in which a freely breeding population of fish, stocked originally as fingerlings (2 g) from a local fry dealer, were harvested regularly by seining over an 18 month period. The fish used for this experiment came from the final pond harvest and were of mixed ages (indeterminable) and size classes (70-300 g). Septage was the only feed/fertiliser input into the system and was used at a rate of 150 kg chemical oxygen demand/ha/day loaded twice weekly.

Broodfish were placed in small concrete tanks connected to the hatchery recirculated water system to recover before individual female broodfish were tagged. Small plastic tags were attached behind and under the dorsal fin with nylon line after the individual fish were anaesthetized. A damp cloth was employed to hold each fish while a tag was attached using a hypodermic syringe. Each fish was then weighed (to 1 g) on a battery operated Soehnle balance. Total length was also measured to the nearest 0.1 cm. Male broodfish were also individually weighed and measured.

(b) Stocking and harvest

Broodfish were originally stocked in each of the four treatments (Table 2.2) at a ratio of 2:1 (female:male) although

this was subsequently reduced to 1:1 as the experiment proceeded and stocking density was adjusted for growth. The total number of broodfish required varied with treatment and female broodfish were exchanged in only two treatments (T12, T14). Broodfish in these treatments were exchanged for females maintained in conditioning tanks.

In one treatment (T13) swim-up fry were removed with a finemesh dipnet 5-6 times dai.ly but broodfish otherwise remained undisturbed for the duration of the experiment. Every 10 days, eggs and fry were harvested from the other treatments (T11, T12 and T14) after draining down the water and catching all the broodfish. Each clutch of eggs or fry was labelled separately with the fish number, date and treatment and stored for later analysis. Free swimming fry were removed with a fine mesh dip net and preserved in the same way. All female fish were subsequently individually weighed and measured. Males were counted and bulk weighed at each harvest. In treatments in which all or only spawned female broodfish were exchanged at harvest (T12 and T14 respectively) replacement female fish removed from conditioning tanks were similarly treated before being placed in the spawning tanks. Replacement fish for spawned females in Treatment 4 were selected randomly from the conditioning tank. Mortalities for every treatment were replaced using fish of the same origin maintained in similar conditions in the hatchery.

Post-trial observation

After the 60 day trial was completed, the experimental fish were restocked in the same treatments. Management continued in the same way except that the Interharvest Interval was reduced to five days.

. (c) Biometrics

- 1. Egg/fry production with time showed non-homogeneous variance between treatments and non-normal distribution. The square-root transformation used is suitable for data in which some of the values are very small or equal to zero (Steel and Torrie, 1960). Analysis of variance was then performed on the transformed data to detect differences between the means at the 0.05 level of probability.
- 2. Differences in mean egg/fry production during harvests at five and ten day intervals were analyzed using a Wilcoxon test for paired observations (Walpole and Myers, 1978).
- Change in body weight was calculated on the basis of

$$(Wo-W1)$$
 $(W1-W2)$ dtWs = ----- x 100 and dtWc = ---- x 100 W1

where, dtWs = change in weight during spawning
 dtWc = change in weight during conditioning
 Wo = body weight before spawning (t)
 W1 = body weight after spawning (t+10)
 W2 =body weight after conditioning (t+20)

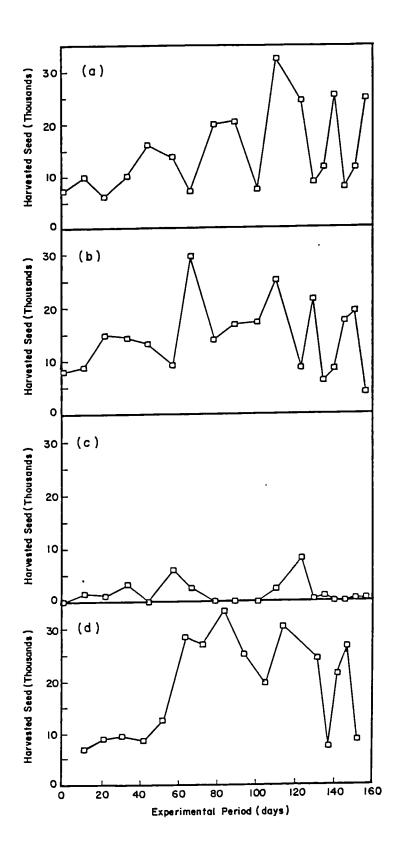
4. Mean percentage change in body weight were compared using

Analysis of Variance and Duncan's Multiple Range test for

significance at the 0.05 level of probability.

Actual production of seed per harvest from concrete spawning tanks (Area = 12.57 m^2) over experimental period

- (a) T11; no exchange of females at harvest
- (b) T12; all females exchanged at harvest
- (c) T13; swim-up fry removed daily, no disturbance of broodfish
- (d) T14; spawned females exchanged at harvest (partial exchange)



Estimated production of seed per 5 day period from concrete spawning tanks (Area = 12.57 $\rm m^2$) over experimental period

- (a) T11; no exchange of females at harvest
- (b) T12; all females exchanged at harvest(c) T14; spawned females exchanged at harvest (partial) exchange)

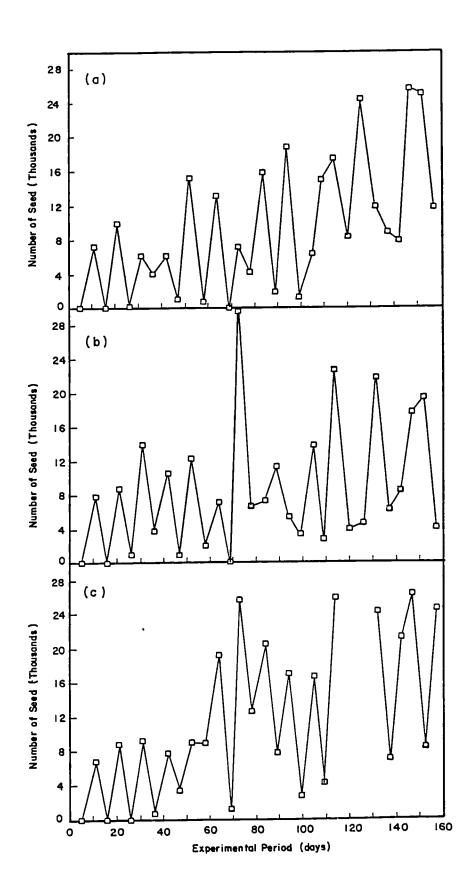


Table 2.3 Mean 5 day output of seed from concrete tanks (Area=12.57 m^2) before and after change in frequency of harvest from 10 to 5 days, for different strategies of broodfish disturbance and exchange (T1)

Suptom/Dymoniment/Theetment		No. of seed/5 day harvest interval			
Sys	System/Experiment/Treatment		5 days	% change	
T11	no female exchange	7641.5	15045.8	+ 96.8*	
T12	total female exchangeb	7483.3	12863.8	+ 71.9	
T13	undisturbed ^a	987.8	499.3	С	
T14	partial female exchangeb	9494.2	16005.6	+ 68.6*	

^{*} P < 0.05, Wilcoxon Test for paired observations

a calculated based on a mean 5 day yield

b females only

c frequency of harvest unchanged

Relationship between time of spawning and stage of seed harvested for broodfish stocked in spawning tanks and hapas for a 5 day period

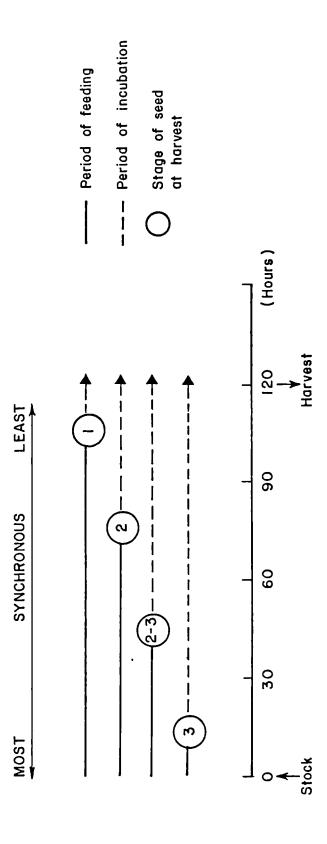


Table 2.4 The percentage of total seed production by female broodfish in concrete tanks observed at harvest as stage 3-4 and stage 1-1.5 after a change in frequency of harvest from 10 to 5 days for different strategies of female broodfish exchange (T1)

	Treatment	Mean values of percentage of seed harvested Harvest interval (n = 18)				
·	Treatment	10 days	5 days	All data		
Seed	(Stage 3-4)1					
T11	Non-exchange	38.5ª	8.8ª	28.0ª		
T12	Total exchange ³	59.8ª	25.8ª	47.8b		
T14	Partial exchange ³	58.5ª	7.5ª	40.5ªb		
Seed	(Stage 1-1.5) ²					
T11	Non-exchange	46.0ª	47.4ª	46.5×		
T12	Total exchange	28.9ª	16.0ª	23.9y		
T14	Partial exchange	27.8ª	28.4ª	29.3z		

Means within groups followed by a common superscript are not significantly different (P > 0.05, Mann-Whitney-U Test)

¹ stage 3-4 seed just before or after hatching

² stage 1-1.5 uneyed egg

³ female broodfish only

Table 2.5 Growth parameters of broodfish at stocking and at harvest after a period (t = 155 days) of seed production in concrete tanks (Area = 12.57 m²) for different strategies of broodstock exchange and disturbance (T1)

			MEAN	± SE		
Treat- TL (cm)		Ma	les	Females		
ment:	s BW (g)	Day 1 (n=132)	Day 155 (n=119)	Day 1 (n=332)	Day 155 (n=227)	
T11	Total length Body weight	22.3a ± 0.3 181.0b ± 8.0	29.9a ± 0.4 477.7d ± 16.9	20.8a ± 0.5 163.7b ± 15.9	26.4ª ± 0.4 338.6d ± 18.4	
T12	Total length Body weight	22.0a ± 0.3 164.4b ± 7.3	30.9b ± 0.4 540.6e ± 22.8	21.1ª ± 0.3 166.0b ± 7.1	24.4 ^b ± 0.3 262.9 ^e ± 9.5	
T13	Total length Body weight	22.1a ± 0.3 168.2b ± 7.7	32.4° ± 0.3 624.9° ± 22.3	21.0a ± 0.4 160.1b ± 9.0	27.6° ± 0.4 405.8° ± 19.9	
T14	Total length Body weight	22.6a ± 0.3 185.5b ± 8.9	31.5 ^b ± 0.3 559.0 ^e ± 15.9	20.7a ± 0.3 149.6b ± 7.6	$25.7^{2} \pm 0.2$ $316.3^{d} \pm 15.9$	
F	Total length Body weight	0.86 ^{n.s.} 1.58 ^{n.s.}	10.88*** 10.50***	0.34 ^{n.s.}	13.28*** 14.59***	

Means with the same superscripts in the same columns are not significantly different P > 0.05. (Duncan's Multiple Range Test)

F value one-way Anova n.s. P > 0.05

- P < 0.05
- P < 0.01
- P < 0.001
- Total length TL
- Body weight ₿₩

Table 2.6 Percentage change in body weight of mouthbrooding female broodfish during (a) the interharvest period (10 days) and (b) the conditioning period for different strategies of female broodfish exchange (T1)

	% change in body weight mean ± S.D. (n)
(a) during interharvest period	l I
T11 T12 T14	$+3.69^{a}$ ± 10.57 (81) -0.73^{b} ± 8.45 (135) -1.57^{b} ± 9.47 (152)
F 881***	
(b) during conditioning	
T11 T12 T14	$+15.84^{\times}$ \pm 8.60 (69) + 8.47 \pm 6.27 (199) + 8.97 \pm 7.97 (150)
F 24.4***	

F value one-way Anova
*** P <0.001

Means in the same group having the same superscript are not significantly different (P> 0.05, Duncan's Multiple Range Test) (T11, no female exchange; T12, total female exchange; T14, partial female exchange).

2.3.2 Results

(a) Seed yield and periodicity of spawning

Production of eggs and fry showed considerable variation with time in all treatments during both 10 and 5 day harvest intervals (days 1-125 and days 126-152, respectively) (Fig. 2.7). Mean yields of eggs and fry calculated on a daily or 5 day output basis over the first 125 days were not significantly different between treatments: T11 seed/day), T12 (1497 seed/day) and T14 (1899 seed/day); Treatment T13 had a considerably lower output (198 seed/day). Estimates for 5-day output during the ten day harvesting period (1-125 days) were made by considering harvest yields by stage. Swim-up fry were assumed to have been spawned during the first 5 day period and all other seed during the second five day period.

Fig. 2.8 shows the resulting periodicity of egg production (=breeding). All three treatments follow a similar trend with a breeding peak followed by a trough on an approximately five day cycle.

Seed production compared before and after the change in frequency of harvest for each treatment in turn shows there was a large increase in mean output (Table 2.3), which is significant (P<0.05) for Treatments T11 and T14.

(b) Synchrony of egg production

The timing of egg production within the period available for the fish to spawn enabled some estimation of synchrony of breeding within the treatment. The mean stage of all seed clutches was used as an indicator of early or late spawning (Fig. 2.9). A comparison of synchrony of spawning could be made only between treatments in which eggs and fry were harvested completely after drainage (T11, T12, T14). Over the whole experimental period (10 day and 5 day Interharvest Interval) exchange of all females significantly improved spawning synchrony, as indicated by the proportion of early or late spawned seed, compared to non-exchange; if exchange strategies are pooled (T12 and T14) they showed improved synchrony of production compared to non-exchange (T11) (Table 2.4).

An estimation of the proportion of eggs and fry produced later in the period of breeding further suggested that spawning occurred earlier with replacement than without replacement. Nearly half of all seed recovered in the non-exchange treatment (T11) were observed as Stage 1-1.5 (Table 2.4).

After the management change of reducing harvest interval from ten to five days, late spawning in the treatment replacing all females (T12) appeared to decline further. The number of Stage 1-1.5 eggs as a percentage of the total seed harvested declined from 28.9% during the 10 day harvest, to only 16% when harvest frequency was increased, indicating that synchrony of spawning was further improved by frequent harvesting.

Synchrony of spawning also improved after a reduction in harvest interval in treatments replacing only spawned females (T14) and those without exchange (T11).

The percentage of seed spawned just before harvest (Stage 1.0-1.5) after a period of 10 days in the spawning tank remained the same even when the period for spawning was reduced to 5 days.

(c) Broodstock condition and growth

Broodstock management, and resulting seed production, had a significant effect on the growth characteristics of broodfish of both sexes held in concrete tanks. Males and females in treatment T13, in which fry were removed from the edge of the tank and broodstock were not disturbed by drainage of the system, showed the best mean individual growth (males, 2.9 g/day; females, 1.58 g/day; Table 2.5). Males in treatments replacing females grew significantly better (T12, 2.42 g/day; T14, 2.41 g/day; Table 2.5) than those spawning with the same females continually (T11, 1.91 g/day; Table 2.5). In contrast, females gained more weight when left in the spawning tank throughout (T11, 1.13 g/day; Table 2.5) rather than being removed after spawning (T14, 1.08 g/day; Table 2.5) or opportunity to spawn (T12, 0.63 g/day; Table 2.5).

Monitoring of broodstock size before and after spawning showed differences between treatments. Female broodfish in replacement treatments both lost more weight during spawning (T12, -0.73%;

Table 2.7 The degree of association (r) between the change in body weight of female broodfish of <u>O.niloticus</u> during the interharvest period (10 days) and after a subsequent period of conditioning and stage of seed recovered at harvest for different strategies of female broodfish exchange (T1)

Treatment	During interharvest period r value (n)	After conditioning period r value (n)
T11	0.656 ** (81)	0.1 n·s· (69)
T12	0.522 ** (149)	0.38 ** (119)
T14	0.601 ** (152)	0.02 n·s·(150)

¹ correlation analysis

figures in parentheses are number of observations

y = a + bx

where

y = change in body weight x = stage of seed recovered

^{**} P < 0.01

n.s. P > 0.05

Fig. 2.10

The relationship between percentage change in total body weight during spawning (Interharvest period 10 days) of individual mouthbrooding females with stage of seed harvested

```
(a) T11; no females exchanged in spawning tank, y = 17.82 - 6.75x, r = 0.66, P < 0.01, n = 81;</li>
(b) T12; all females exchanged y = 11.65 - 4.49x, r = 0.52, P < 0.01, n = 149;</li>
(c) T14; spawned females exchanged y = 13.194 - 5.56x, r = 0.60, P < 0.01, n = 152 y = a + bx, where y = % change in body weight of female broodfish during spawning period x = stage of seed recovered</li>
```

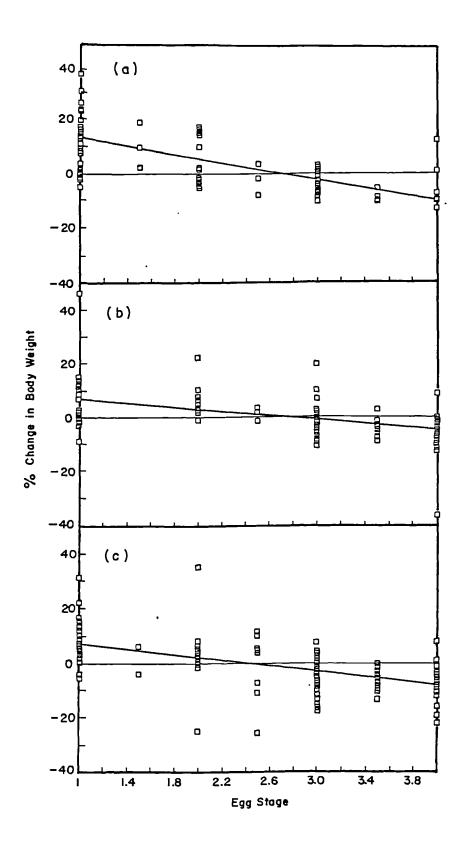


Fig. 2.11

The relationship between percentage change in total body weight of individual female broodfish after seed harvest during conditioning period with stage of seed

```
(a) T11; no females exchanged in spawning tank, r = 0.1, P > 0.05, n = 69;
(b) T12; all females exchanged y = 1.78 - 2.29x, r = 0.38, P < 0.01, n = 119;</li>
(c) T14; spawned females exchanged, r = 0.02, P > 0.05, n = 150
y = a + bx where y = % change in body weight of female broodfish during conditioning period x = stage of seed recovered during previous harvest
```

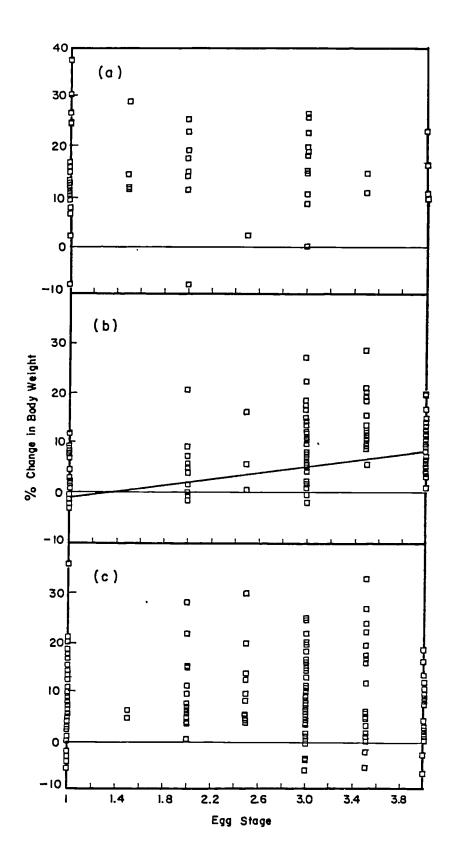


Table 2.8 The degree of association (r) between mean reproductive traits and body weight for O.niloticus females under different exchange strategies

	-			· -	-	Treat	ment				
Seed	stage		T11	· · · · · · · · · · · · · · · · · · ·		T12			T14		
		r²	r	P n	r ²	r	P n	r²	r	P	n
1	BL/BW	0.88	0.94	** 53	0.88	0.92	** 31	0.88	0.94	**	44
	ETN/BW	0.19	0.44	** 53	0.06	0.24	n.s. 31	0.24	0.49	**	44
	EIW/BW	0.03	0.17	n.s. 53	0.001	0.037	n.s. 31	0.001	0.032	v.s.	44
	ETW/BW	0.36	0.6	** 53	0.068	0.261	n.s. 31	0.26	0.51	**	44
2	BL/BW	0.89	0.93	** 24	0.84	0.92	** 16	0.79	0.89	**	25
	ETN/BW	0.62	0.78	** 24	0.32	0.57	= 16	0.09	0.3	n.s.	25
	EIW/BW	0.12	0.35	n.s. 24	0.05	0.22	n.s. 16	0.098	0.31	*	25
	ETW/BW	0.44	0.66	** 24	0.4	0.63	* 16	0.23	0.48	*	25
3	BL/BW	0.91	0.95	** 9	0.90	0.95	** 81	0.81	0.90	**	75
	ETN/BW	0.42	0.65	n.s. g	0.190	0.44	** 81	0.05	0.23	n.s.	75
	EIW/BW	0.02	0.14	n.s. 9	0.000	0.043	n.s. 81	0.068	0.26	n.s.	75
	ETW/BW	0.48	0.69	* 9	0.226	0.48	** 81	0.13	0.36	*	75
4	BL/BW	0.55	0.74	** 34	0.93	0.96	** 36	0.79	0.96	*	23
	ETN/BW	0.44	0.66	** 34	0.21	0.45	** 36	0.16	0.41	n.s.	23
	EIW/BW	0.06	0.245	n.s. 34	0.04	0.19	n.s. 36	0.23	0.48	*	23
	EIW/BW	0.38	0.616	n.s. 34	0.33	0.57	n.s. 36	0.32	0.57	*	23
AL	L BL/BW	0.77	0.88	** 120	0.89	0.95	** 164	0.84	0.92	**	193
	ETW/BW	0.32	0.57	** 120	0.17	0.41	** 164	0.1	0.322	**	193
	EIW/BW	0.00	0.00	n.s 120	0.002	0.05	n.s 164	0.018	0.133	n.s.	193
	ETW/BW	0.37	0.61	== 120	0.18	0.43	** 164	0.185	0.43	**	193
	,		•								

Correlation analysis

BL = Body length, BW = Body weight, ETN = Egg total number, ETW = Egg Individual weight, ETW = Egg total weight

y = a + bx

y = BL, EIN, EIW and ETW x = BWwhere

P < 0.01

P < 0.05

n.s. P > 0.05

T14, -1.6%; Table 2.6) and gained less weight after spawning (T12, 8.44%; T14, 8.97%; Table 2.6) than female broodstock that remained in the spawning tank continually (T11, during +3.69%, after +15.84%; Table 2.6). This can at least partly be explained by improved synchrony of spawning in replacement strategies because spawning tended to occur more quickly after stocking in the spawning tank and thus mouthbrooding was carried out for a longer period. The change in female body weight as related to egg stage, and hence period of incubation (= non-feeding) is shown for each treatment (Table 2.7, Fig. 2.10).

A relationship between change in body weight after spawning with stage of harvested eggs was only found for T12, in which a longer period of incubation led to a subsequently greater increase in body weight during the following incubation period(Table 2.7, Fig. 2.11).

Change in body weight during and after spawning showed a large range. Over a spawning period of 10 days, some females increased body weight by up to 40% and spawned in the same period. Loss in weight only became large as the incubation period extended to post-hatching (stage 3). Loss in female weight during incubation could be as high as 15% of the prespawning body weight, and some females in T14 exceeded 20%. Many females found in the spawning tanks suffered much higher weight loss over the 10 day period (>30% loss in weight over a 10 day period), presumably from mouthbrooding for much longer periods before releasing their fry into the tank. But as these

fish could not be positively identified as breeding fish they were not included in this analysis.

(d) Egg production parameters

The degree of association between various reproductive traits and body size for different seed production strategies is given in Table 2.8. Clutch size (ETN) and weight (ETW) both showed a weak but significant relationship ($r^2 = 0.18-0.62$) with broodstock size in most treatments and egg stages. Eyed eggs, those classed as stage 2, gave the best correlation with clutch size and weight for Treatments T11 and T12.

Egg individual weight (EIW) was not significantly correlated with body weight for any egg stage or treatment.

2.3.3 Discussion

Frequency of broodfish harvesting disturbance

A common restriction on the production of tilapia fry in commercial hatcheries has been the presumed need to avoid disturbance of the broodfish for prolonged periods (14-21 days). This practice, in theory, allowed the maximum number of asynchronously maturing females to be courted by the polygamous males. In contrast, the frequent disturbance of broodstock when harvesting seed from the buccal cavity in this study (T11, T12 and T14) actually increased production of seed compared to a less intrusive method in which fry were removed from the edge of the tank after natural incubation without draining and catching the broodfish (T13).

Liao and Chen (1983) claimed that too frequent removal stressed spawners which in turn affected the quantity and quality of eggs. Beveridge (1984) suggested that disruption by frequent harvest stressed broodstock and interfered with breeding in experimental breeding hapas in the Philippines, Berrios-Hernandez and Snow (1983) compared the output of fry from static water plastic pools by daily dipnetting, regular (every three weeks) draining and fry removal, and removal of seed from the spawning unit and females' mouths every 12 days (egg transfer). Fry and egg transfer gave similar yields after artificial incubation of the latter over a three month trial and the more frequent harvest of eggs was therefore discounted as not economical. However, Verdegem and McGinty (1987)

recently demonstrated that frequent collection of seed from female <u>Oreochromis niloticus</u> broodfish mouthbrooding in small hapas significantly increased spawning frequency.

The advantages in reducing the duration of natural incubation on the productivity of individual females has been demonstrated in aquaria and small tanks. A reduction of the Interspawning Interval (ISI) of females following early removal of eggs has been reported by several authors (Dadzie, 1970; Peters, 1963; Lee, 1979; Siraj et al., 1983; Rana, 1986). The productivity of these systems, despite disturbance during egg removal, was far higher than any recorded in larger and/or commercial systems in which fish remain undisturbed.

Certainly <u>Oreochromis</u> adapts readily to a variety of environments and quickly begins to breed. The establishment of territories and courtship begin within hours of broodfish release in spawning units. Under natural conditions, breeding can begin rapidly once conditions become optimal. Marshall (1979) observed new nests of <u>S. macrochir</u> on land flooded naturally in as little as 12 hours. In the AIT experiments, males took up residence within spawning 'nests' within a few hours of stocking in the concrete tanks.

The very low production of fry from the non-disturbed treatment was probably due to a combination of factors. Spawning activity may have been low, incubation success poor, and the fry released may have been cannibalised by other broodfish before they could be harvested. In the present experiments, predation on the newly released fry, despite maternal protection, was

probably high as the water was clear and there were no nursery or shallow areas in the tank. Iles (1973) reported that females commonly ate their broods soon after release in small tanks and that in larger tanks containing non-breeding adults, newly released broods were consumed in days or even hours. Philippart and Ruwet (1983) described the vegetated areas preferred by incubating Q. mossambicus females as an adaptive response to improve survival of their young; the exit of spawned females from the spawning lek under natural conditions could be for the same reason (Fryer and Iles, 1972).

Growth of individual male and female breeding fish in the undisturbed treatment (T13), was significantly greater than in the other treatments. The quantity of feed, given to appetite, also much greater in both absolute terms and as percentage of body weight. This suggests that spawning activity was lower than in the other treatments and that reproductive effort was depressed in favour of body growth. The reasons for the probable lower degree of spawning activity in the 'non-disturbance' treatment can only be speculated. The Interspawning Interval (ISI) of female fish incubating eggs naturally is likely to be long, especially if body reserves are particularly depleted by conditions unsuitable for brooding and nursing. However, the mean body condition of females and males on harvest was high, suggesting that other factors may have restricted spawning activity.

Reduction in Interharvest Interval

The reduction of Interharvest Interval (IHI) from 10 days to 5 days significantly increased output of seed in the three frequent harvest strategies treated as a composite group. The non-replacement strategy responded particularly strongly to more frequent harvests, suggesting that an extended period of natural incubation reduced egg production, especially when females were not removed from the spawning tank and conditioned after harvest.

Periodicity of spawning

A similar periodicity of seed production, an approximate five day cycle, was evident for both female exchange and non-exchange treatments during the ten day harvest period. The period lengthened to approximately 10 days when the harvest interval was reduced to 5 days. The cycle probably lengthened when harvest interval was decreased to 5 days because the period of conditioning was inadequate.

Lee (1979) observed that a small group of females in the same aquarium would usually spawn on the same day, indicating a degree of mutual stimulation and synchronisation. This suggests that females of similar condition and ripeness which are close to ovulation might be 'pulled over the top' by the nearby courtship and spawning of con-specifics. This would tend to cause the peaks in spawning every few days observed in the present study.

Male condition was probably not an important factor in the periodicity of spawning. Although fertilisation has been found to decrease with successive batches, dominant males will continue to court and spawn with up to four females a day (Rana, 1987).

Effect of hierarchy on spawning activity

Fishelson (1983) described the hierarchical structure of a group of O. niloticus kept in the same tank over a prolonged period and observed that after a while competitive interactions between fish became ritualised; a strategy by which energy flow is minimised (Dice, 1962). The dominant fish, both male and female, expended less energy than other fish to 'control' their area of the unit and the hierarchy persisted for a long time under unchangeable ambient conditions.

Turner (1986) found that dominant males in small groups of O. mossambicus maintained territories in favoured positions within the spawning unit, and for longer periods of time than subordinates, which in a natural system would be relegated to the periphery of the lek. Fernö (1987) correlated dominance amongst males in Astatotilapia burtoni, a territorial African cichlid, with improved mating success with the non-territorial females but did not describe a hierarchy between females in this species.

If a hierarchy becomes established to the extent that a few individual males and females dominate spawning behaviour in the limited area of a high density spawning unit, the sexual activity of the majority of subordinate fish would inevitably be reduced. This hypothesis can also partly explain the earlier, more synchronous spawning found in replacement treatments and the higher level of spawning activity apparent generally in treatments in which spawning and the hierarchy were regularly interrupted.

Turner (1986) suggested that catching and measuring fish in breeding colonies of <u>O. mossambicus</u> were highly stressful and entirely disrupted the structure of territories and possibly the social relationships as well.

The results of the present study suggest that disturbance may promote spawning activity. Although the 1eve1 of seed production amongst the 'disturbed' treatments was similar, the or synchrony of spawning between timing harvests was significantly different. It appeared that frequent harvest, and most importantly, further disturbance of the hierarchy by exchange of broodfish, increased synchrony of spawning. Reproductive synchrony may be a common phenomenon in natural communities of tilapia, especially those in which climatic/environmental factors the continuous prevent asynchronous breeding common in many culture systems. Silverman (1978b) suggested that synchronised reproduction would allow populations to utilize temporarily favourable conditions and in the wild enable cichlids to colonize new habitats. Females stocked into spawning tanks after a period of conditioning at high density with other females tended to spawn more quickly. Synchrony further improved as harvest frequency increased,

suggesting that the optimal conditions for spawning were being approached particularly when all females were replaced (T12).

Effect of spawning activity on body condition and growth

The differences in synchrony of spawning explained the observed variation in body condition and growth at final harvest. In replacement treatments in which spawning occurred more quickly stocking, а large proportion of females after were mouthbrooding, and therefore not feeding, for much of the time. The average growth of males in these treatments (T12 & T14) was thus greater than in the non-exchange treatment (T11) in which spawning was less synchronised and females competed for feed with the males over a longer period. This is confirmed by the faster mean growth rate of females in this treatment (T11) compared to exchange treatments.

The poorer weight gain of exchanged females after spawning suggests that the conditioning tank was a poorer environment for recovery than the spawning tank itself. Spawned females from the total exchange treatment (T12), which spawned earlier and incubated longer, regained condition faster than females that had incubated for a shorter period, but this trend was not shown by the other treatments. Because spawned females (T14) recovered in conditioning tanks in which other fish were already present, this may have moderated the attempts of the most starved (and weakest?) fish to regain condition.

Table 2.9 The relationship of seed clutch size (all stages) and body length for female broodfish spawned in concrete tanks (Area = 12.57 m²) under different exchange strategies (T1). Seed removed from the mouth of individual females every 10 days; comparative data given for Oreochromis niloticus from other workers

CS	=	-1392 + 113TL	(r = 0.56**, n = 114)
CS	=	-684 + 76TL	(r = 0.40**, n = 164)
CS	=	-529 + 71TL	(r = 0.39**, n = 180)
CS	=	-852 + 86TL	
CS	=	5.31 SL1.74	Rana (1986)
F	=	2.14 SL ^{2.25}	Payne and Collinson (1983)
	cs cs cs cs	CS = CS = CS =	$CS = -529 + 71TL$ $CS = -852 + 86TL$ $CS = 5.31 SL^{1.74}$

TL Total length

SL Standard length

CS Clutch Size = number of eggs harvested from mouth of individual female

Total fecundity = ovarian egg counts from mature ovaries

^{**} r significant (P = < 0.01)

Egg production and estimation of fecundity

Egg production parameters showed the same trends as previously reported by Rana (1986) for seed removed from small numbers of broodfish shortly after spawning. The poorer relationship between female size and seed clutch size and weight in this study reflect the more heterogeneous environment during spawning and post-spawning in tanks containing large numbers of broodfish at high stocking densities. The lack of relationship between egg size and female body weight also concurs with Rana's (1986) results for females of the same age. The usefulness of the term 'total fecundity' to describe seed clutch size of fish immediately after spawning by Rana (1988) for commercial conditions is questionable as seed cannot be collected at this time unless stripped. A different relationship was shown for seed harvested in the present study (Table 2.9) in which seed were harvested at different stages of development from large numbers of fish spawning within a similar period.

Table 2.10 The experimental design for investigation of the effect of environmental chang∈ and female broodfish exchange strategy on spawning in nylon hapas suspended ir earthen ponds (H1)

	F	Broodfish	1			Drech	anged		
System/ Experiment/	Number o		Category	Restock spawi		fro		Interva (days)	
Treatment	per spawning	Total		Tank	Hapa	Tank	Hapa	Spawning	Cond.
H11	40	40	males		х		-		
	40	120	spawned females			х		5	10
	.5	120	unspawned females			x			
H12	40	40	males		x				
	40	120	spawned females				x	5	10
	.0	120	unspawned females				x		
Н13	40	40	males	}	x			!	
	40	120	spawned females			х		5	10
			unspawned females		x				
H14	40	40	males .	{	x				
	40	120	spawned females				x	5	10
			unspawned females		х				

Cond. = Conditioning

x denotes restocking and conditioning strategy

Table 2.11 The experimental design for investigation of the effect of environmental change and female broodfish exchange strategy on spawning in concrete tanks (T2)

	E	roodfish	ı			Broke			
System/ Experiment/	Number o	of fish	Category	Restock spawni		fro	inged om tioning	Interva (days)	
Treatment	per spawning	Total		Tank	Hapa	Tank	Hapa	Spawning	Cond.
T21	40	40	males	х					
	40	120	spawned females unspawned			x		5	10
			females			х			
T22	40	40	males	x					
	40	120	spawned females				x	5	10
			unspawned females				x		
T23	40	40	males	х					
	40	120	spawned females			x		5	10
			unspawned females	х					
T24	. 40	40	males .	x					
	40	120	spawned females				x	5	10
			unspawned females	ж					

Cond. = Conditioning

x denotes restocking and conditioning strategy

2.4 Effect of conditioning environment and exchange strategy on seed production in tanks and hapas

2.4.1 Background

This trial estimated seed production from broodfish maintained in spawning and conditioning tanks and hapas under different exchange regimes. Experiment 1 (T1) indicated that disturbance of the hierarchy between spawning tilapias and frequent harvest of seed increased both the level and synchrony of seed This experiment (T2, H1) sought to investigate broodstock exchange for both tank and hapa spawning units in relation to a change in conditioning environment. It is believed that a change in environment can have a positive effect on breeding synchrony in tilapias leading to improved productivity and reduced costs. Hatchery operators in Israel have believed for many years that a change of water in aquaria can induce spawning in groups of tilapia (Mires, 1982), whilst others have realised that a short production cycle in ponds with frequent draining and refilling can optimise fry yields. The practice of holding broodfish at high density in hapas suspended in pond water rich in natural food, before breeding in clearwater units, could reduce costs compared to a totally tank-based system. Alternatively, synchrony might be improved with a change from clearwater to the low density green water conditions of the spawning hapa.

Broodfish were both spawned in hapas (Table 2.10) and tanks (Table 2.11) after tank or hapa conditioning. The effect of

total and partial exchange of females at each harvest was not clear in the first experiment (T1) and so was repeated for females conditioned for not less than a 10 day period.

2.4.2 Methods

a) Origin and Pretreatment of Broodstock

The broodfish were obtained from earthen ponds after the harvest of a duck/fish experiment on the AIT campus. Fish had been stocked as advanced fingerlings (3 months old; mean body weight - 10 g per fish) and raised at a density of 4 fish/m² over a period of 6 months with duck manure as the only input (10-20 ducks/200 m²; Edwards et al., 1987). Fish harvested from the ponds were stocked in large nylon hapas (area: 40 m²) suspended in earthen ponds. A floating catfish pellet (30% crude protein) was fed twice daily to appetite for a further three months prior to using the fish.

(b) Stocking and Harvest

Management and harvest procedures followed those outlined in sections 2.2.2.1 - 2.2.2.3.

An interharvest interval of 5 days and a conditioning period of at least 10 days was followed (Tables 2.10 and 2.11).

Each treatment required the use of three sets of females at any one time: one set in the spawning unit and two sets in the conditioning unit. All the broodfish were counted and bulk weighed on transfer from and to the spawning units and a sample of fish (initially 30%) were tagged and individually weighed and measured at each harvest. The eggs and fry of all

mouthbrooding females were preserved as individual batches. Treatments in which only spawned females were replaced were provided with equal numbers of replacement females taken at random from conditioning hapas or tanks (Tables 2.10, 2.11). A ratio of 1:1 (female:male) was used throughout the experimental period. Initial stocking data are given in Table 2.10 (H1) and Table 2.11 (T2).

At final harvest, spawned and unspawned females were separated and ovaries removed from all fish for estimation of gonadosomatic index (GSI). Samples were taken for analysis of moisture, total lipid and crude protein content using standard methods (Appendix 1).

Table 2.12 Mean total seed output per spawning unit per harvest (5 days) in tanks (Area=12.57 m², T2) and hapas (Area=40 m², H1) for different female exchange strategies and conditioning environments*

Treatment		d per unit vest (SE)	n
H11	14932	(1527)	17
н12	17194	(1317)	19
н13	15522	(1477)	20
H14	14047	(1226)	20
T21	12962	(1199)	20
T22	16274	(1543)	20
Т23	14661	(1453)	20
T24	16057	(1347)	19

F value one-way Anova = $0.49^{n \cdot s}$. Total seed = eggs and fry of all stages of development combined $n \cdot s \cdot p > 0.05$

^{*} Broodfish stocked at 80 fish per spawning unit (female:male ratio = 1); Experimental period: 116 days

Fig. 2.12

Total seed production per harvest per tank (Area = 12.57 m^2) over the whole experimental period for broodfish under different exchange and management regimes (T21 - T24)

T21 Total female exchange, conditioned in tanks
T22 Total female exchange, conditioned in hapas
T23 Partial female exchange, conditioned in tanks
T24 Partial female exchange, conditioned in hapas

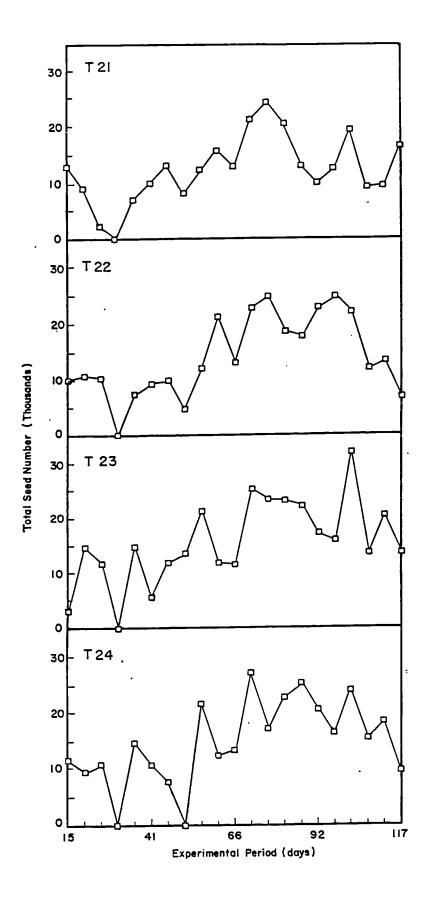


Fig. 2.13

Total seed production per harvest per hapa (Area = 40 m^2) over the whole experimental period for broodfish under different exchange and management regimes (H11 - H14)

H21 Total female exchange, conditioned in tanks

H22 Total female exchange, conditioned in hapas

H23 Partial female exchange, conditioned in tanks H24 Partial female exchange, conditioned in hapas

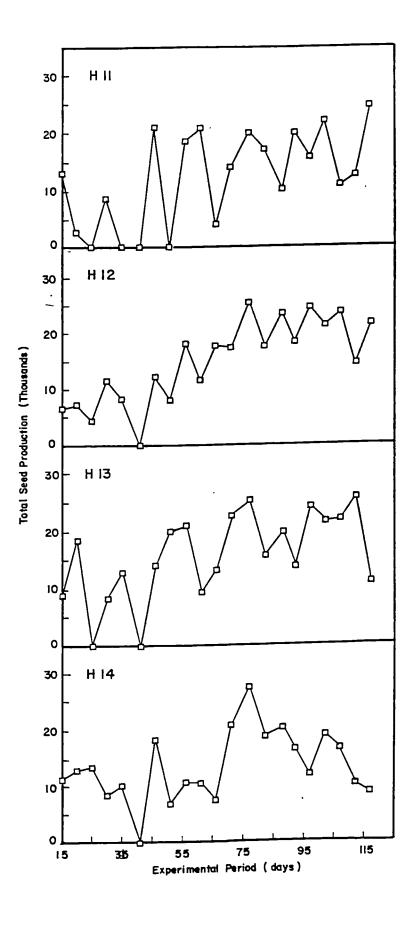


Table 2.13 Early spawned seed¹ as a percentage of total seed production harvested every 5 days from (a) spawning units² stocked with female broodfish managed under a range of different exchange and conditioning strategies (b) spawning hapas stocked with females conditioned in tanks or hapas and (c) all spawning tank and hapa treatments

Treatment	Mean percent spawned	age of early seed	Chi-square value
	%	n	
(a)			
T21	59.9	21	
T22	75.4	21	
T23	71.3	21	
T24	65.0	19	14.4*
H11	69.5	17	
H12	78.3	19	{
H13	75.8	20	
H14	80.3	19	
(b)			
Tank conditioning (H11 + H13)	79.3	37	1.6n.s.
Hapa conditioning (H12 + H14)	72.9	38	1
		_ -	
(c)	1		
All Tank spawning treatments	68.0	82	5.9*
All Hapa spawning treatments	76.1	87]

Level of significance of Chi-square value (Kruskal-Wallis one-way Anova)

n.s. P > 0.05

^{*} P < 0.05

¹ stage 2-3

² Tanks (Area = 12.57 m^2 , T2) and hapas (Area = 40 m^2 , H1) stocked at 80 fish per unit (female:male ratio =1); t = 116 days

2.4.3. Results

2.4.3.1. System data

(a) Seed production

Tank and hapa treatments were stocked with the same absolute numbers of broodstock; stocking densities were therefore considerably different in the two systems throughout the experimental period.

Stocking densities of fish of similar size at stocking (86-140 g) ranged from 0.18-0.25 kg/m² in hapas (40 m²) and 0.55-0.89 kg/m² in tanks (12.57 m²).

There were no significant differences between the eight treatments (Table 2.12) in terms of egg and fry production based on mean 5 day output over the entire 116 day experimental period. Mean output of seed varied between 2592-3439 seed/day across a range of tank and hapa treatments.

Seed production with time showed similar trends in both tanks (Fig. 2.12) and hapas (Fig. 2.13). Yields of seed tended to increase gradually with time.

No lunar periodicity was evident in either tank or hapa spawnings.

(b) Synchrony of spawning

The mean percentage of seed of different stages harvested every 5 days was calculated. Late spawning was least evident and spawning best synchronised in hapas in which spawned females, conditioned in hapas were replaced (H14); at harvest less than

Intercepts, a, (+/- S.D.); regression coefficients, b, (+/- S.D.); coefficients of determination (r²) of the linear regression analysis for reproductive traits in <u>O.niloticus</u> female broodfish maintained in tanks and hapas¹ under different exchange and conditioning regimes Table 2.14

	Clut	Clutch size/Body weight	ody weigh	ht	Clutcl	Clutch weight/Body weight	ody weigh	ıt	Clutch &	Clutch size/Total length	. length		Egg	Egg size/Body weight	weight	
	p	а	r ² /r	DF	q	ಟ	r ² /r	DF	q	ಪ	r²/r	DF	q	ध	r2/r	DF
н11	439.7 ±533.6	2.677 ±0.567	0.179/	102	2.2	0.012 ±0.003	0.174/	100	- 335.5 ± 551.5	58.02 ±16.24	0.113/ 0.34**	100	0.45	0.00024	0.031/ 0.18 ^{n.s.}	100
H12	786.6 ±527.2	2.043 ±0.590	0.071/	158	3.1	0.012 ±0.003	0.100/	158	9.4 ± 524.9	52.29 ±14.21	0.08/	158	0.42	0.00019	0.031/ 0.18n·s·	158
H13	604.6 ±498.8	2.513 ±0.574	0.196/ 0.11n.s.	141	3.2 .	0.011 ±0.003	0.100/	141	- 331.3 ± 501.7	63.26 ±15.20	0.109/	141	0.49	0.000055	0.0016/ 0.04n.s.	141
H14	1054.6 ±681.3	0.834 ±0.878	0.006/ 0.08m.s.	132	4.8	0.004 ±0.004	0.008/ 0.09".8.	132	448.4 ± 678.3	34.06 ±23.46	0.016/ 0.13ns	132	0.47	0.00004	0.0006/ 0.02m.s.	132
T21	277.7 ±526.3	4.370	0.256/ 0.51**	141	1.2	0.024 ±0.003	0.297/ 0.54**	141	-1464.2 ± 523.9	116.60 ±16.50	0.262/ 0.51**	141	0.51	0.00009	0.0035/ 0.06n·s·	141
T22	799.6 ±578.5	2.150 ±0.540	0.09/	161	3.2	0.010	0.130/ 0.36**	161	- 377.8 ± 572.1	71.42 ±15.98	0.11/	161	0.43	0.00	0.02/ 0.14n.s	161
T23	607.6 ±451.2	2.770 ±0.640	0.12/	136	2.3	0.020 ±0.003	0.198/ 0.44**	136	- 723.8 ± 438.1	84.47 ±15.99	0.17/	136	0.48	0.0003	0.03/ 0.17n.s.	136
T24	326.2 ±647.7	4.710 ±0.770	0.203/	146	0.2	0.028 ±0.004	0.318/	146	-1821.7 ± 636.8	137.70 ±20.96	0.229/	145	0.43	0.0003	0.02/ 0.15m.s.	145

n.s. 1

P>0.05 P<0.01 Tanks (Area = 12.57 m², T2) and hapas (Area = 40 m², H1) stocked at 80 fish per unit (Female:male ratio = 1); t = 116 days degrees of freedom ΡĘ

Summary of seed production parameters of female broodfish spawned in tanks and hapas under different exchange and conditioning regimes (all treatments combined) Table 2.15

		Mean ± S.E.	S.E.	
Type of Broodstock	Egg Weight ¹ (g)	Clutch Weight ² (g)	Clutch Size ³ (No.)	Relative clutch size No./g female
Tank conditioned	0.50 ± 0.003	4.95 ± 0.074	1011 ± 15.2	5,95 ± 0,13
napa conditioned	0.46 ± 0.002	5.18 ± 0.078	1154 I 17.2	6.55 ± 0.14
	133.7	4°0-	39.1.	; ; ; ;
All females exchanged Sparmed females exchanged	0.48 ± 0.003	4.93 ± 0.073	1061 ± 15.8	6.15 ± 0.13
parmica tentres evolutibed	0.48 ± 0.003	5.20 ± 0.079	1103 ± 16.8	6.41 ± 0.14
ĹŦ4	1.6".8.	6.4*	3.3n.s.	1.9n.s.
Tank spawned Hapa spawned	0.48 ± 0.003 0.47 ± 0.003	5.33 ± 0.082 4.84 ± 0.14	1126 ± 17.6 1046 ± 15.1	6.38 ± 0.13 6.16 ± 0.14
Œ,	6.2*	20.0**	11,8**	1,1n.s

P < 0.01 P < 0.05 P > 0.05 n.s.

mean weight of 100 fresh seed wet weight

mean weight of all seed in a single clutch wet weight Lotal number of seed in a single clutch

F value one way Anova Tanks (Area = 40 m², H1) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days 20% of seed were staged as just spawned (Stage 1) (Table 2.13). In contrast, 40% of the eggs from fish conditioned and spawned in concrete tanks with all females replaced (T1) were classed as stage 1 seed.

Hapa treatments, taken as a single group, showed significantly more synchrony of spawning than tank treatments (Table 2.13). Synchrony of spawning in hapas was not significantly improved by preconditioning in hapas compared to tanks (Table 2.13). Thus, the spawning unit had a greater effect on spawning synchrony than the conditioning environment.

(c) Reproductive traits and seed production parameters

The correlation (r) between female broodfish weight and egg production parameters is given in Table 2.14. A weak but significant relationship between body weight or total length and total seed number (clutch size) and total seed weight (clutch weight) was evident for fish in most treatments. No correlation was found between brooder size and seed wet weight. Combined data from fish in all hapa and tank breeding treatments respectively indicated that fish spawned in tanks produced slightly larger eggs (2%) and larger and heavier clutches of eggs than those in hapas (7.5%and 10% relative clutch size was not respectively) although significantly larger (Table 2.15).

However, clutch weight, clutch size and relative clutch size (number of seed per g female) were greater in hapa conditioned fish than individuals conditioned in tanks (Table 2.15).

The replacement strategy for female broodfish had less affect

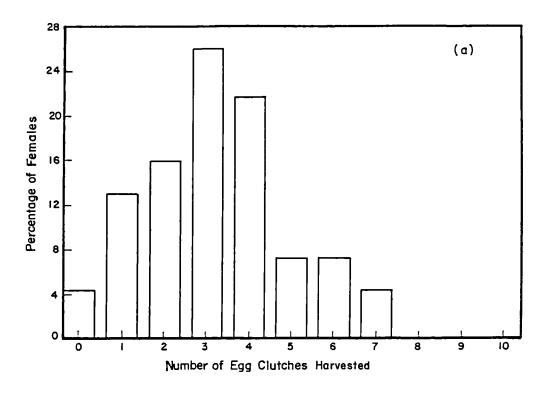
Parameters of seed production for female broodfish spawned in tanks (Area = 12.57 m², T2) and hapas (Area = 40 m², III) for different female exchange strategies and conditioning environments Table 2.16

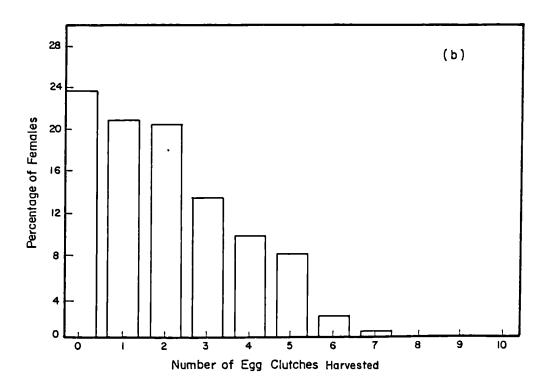
					MEAN ± S.E.	. S.E.	
	Environment Spawning Con	ument Conditioning	Exchange females	Egg weight ¹ (g)	Clutch weight ² (g)	Clutch size3	Egg number per g female
T21	TANK	TANK	ALL	0.52 ± 0.006*	5.39 ± 0.17m	1068 ± 35.6*c	5.99 ± 0.25b
T22	TANK	IIAPA	ALL	0.45 ± 0.004bc	5.09 ± 0.15bc	1168 ± 33.8m	6.49 ± 0.25b
T23	TANK	TANK	SPAWNED	0.51 ± 0.005m	5.42 ± 0.16*	1067 ± 30.0*c	6.54 ± 0.25b
T24	TANK	HAPA	SPAWNED	0.46 ± 0.050bc	5.43 ± 0.18ª	1197 ± 41.0a	6.47 ± 0.27b
н11	HAPA	TANK	ALI,	0.49 ± 0.062°	4.38 ± 0.13bc	918 ± 28.3b	5.04 ± 0.28*
H12	HAPA	HAPA	ALL	0.45 ± 0.043ba	4.97 ± 0.14mc	1099 ± 28,4ac	6.62 ± 0.28b
H13	IMPA	TANK	SPAWNED	0.48 ± 0.005°	4.78 ± 0.13°	1012 ± 27.9bc	5.99 ± 0.25b
1114	HAPA	HAPA	SPAWNED	0.46 ± 0.006ba	5,30 ± 0,16a	1170 ± 36.2ª	6.64 ± 0.33b
된				23,4***	6.2***	8.4***	3.2**

1 mcan weight of 100 fresh seed wet weight
2 mcan weight of all seed in a single clutch wet weight
3 total number of seed in a single clutch
means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)
= (P<0.01), *** (P<0.001), one-way Anova</pre>

Fig. 2.14

Frequency of seed harvest from tagged individual female broofish spawning in (a) hapas and (b) tanks over the experimental period of 116 days (total number of spawns; hapa = 69, tank = 224)





on seed production (Table 2.15). Clutch weight was marginally larger (5.4%) in treatments removing only spawned females. When considered by treatment (Table 2.16), tank spawning and conditioning produced the largest seed (T21 and T23) and tank spawning of selectively exchanged females the heaviest mean clutch weights (T23 and T24).

2.4.3.2. Data for individual fish

Data were based on spawning records for a sample of tagged female broodfish (30% of initial number stocked) from tanks and hapas, the breeding history of which was followed throughout the experimental period.

(a) Frequency of spawning

Analysis of the frequency of spawning by females in hapas and tanks revealed major differences. Across a range of treatments in hapas, the frequency of spawning over the experimental period approached normality. Over 66% of female fish spawned three times or more; and more than 95% of females were observed mouthbrooding at some time during the experiment. In contrast, it appeared that a small group of females dominated the spawning activity in tanks. Over 65% of females spawned less than three times and more than 20% of females were never observed with eggs or fry in the mouth at harvest (Fig. 2.14)

on size characteristics at final harvest of tagged female broodfish maintained in tanks¹ (all treatments combined, tagged females 30% of total number of fish at stocking) Effect of spawning frequency Table 2.17

				MEAN + S.E.	
Spamier category	g		Total length (cm)	Body weight (g)	C.F2.
Non-spawners	56		25.0 ± 0.44a	275.5 ± 13.7ª	1.72 ± 0.03ª
Frequent spawners	30		24.0 ± 0.38^{a}	246.5 ± 11.6^{b}	1.75 ± 0.02^{a}
Intermediate spawners	148	•	23.6 ± 0.19b	242.2 ± 5.8b	1.80 ± 0.01^{b}
F one-way Anova			5.97**	3,69*	5.41**

Means in the same column having a common superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

¹ Tanks (Area = 12.57m², T2) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days ² C.F. Condition Factor

* P < 0.05 ** P < 0.01

Table 2.18 Mean total and reproductive production of female broodfish maintained in tanks and hapas³ under different exchange and conditioning regimes (all tank and hapa treatments combined respectively)

	Mean +	S.E.	F
Production category	Hapas 1	Tank ²	
Total Production (g)	92.5 ± 3.8	86.7 ± 5.8	0.65n.s
Egg production as % of total production	25.4 ± 2.2	19.5 ± 1.2	5.8*
Egg production (g)	17.4 ± 1.4	15.4 ± 0.86	1.35n.s
Increase in body weight (g)	69.8 ± 5.3	77.0 ± 3.3	1.3n.s.

F value one-way Anova

n.s. p > 0.05

^{*} P < 0.05

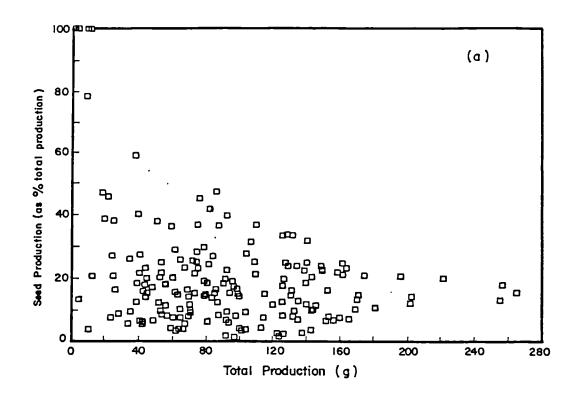
^{1 66} individual fish

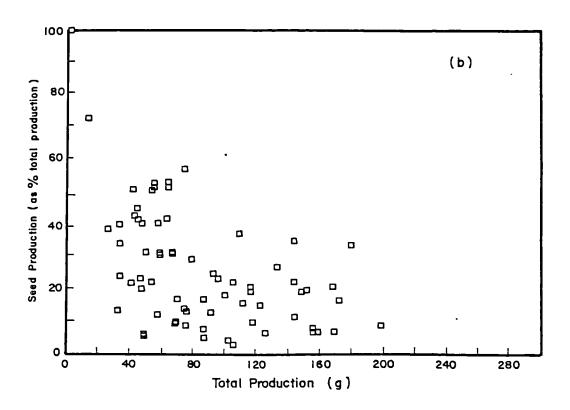
² 180 individual fish

³ Tanks (Area = 12.57 m^2 , T2) and hapas (Area = 40 m^2 , H1) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days

Fig. 2.15

Seed production as a percentage of total production¹ (from the harvest of the first seed clutch to end of experiment; 116 days) of individual female broodfish over a range of treatments in





(b) Final body size characteristics of spawning fish

Fish were divided into three categories on the basis of

frequency of spawning: those that spawned frequently

(>5 times); moderately (<5>0); and non-spawners (0). Non
spawning fish were significantly heavier and longer than

spawners at the end of the experiment (Table 2.17).

Frequently and moderately-spawning fish were not distinguishable by size, but the latters' condition, as indicated by a Fulton's Condition Factor ($K = 100 \times W/l^3$) was higher than that of both non-spawning or frequently-spawning fish.

(c) Reproductive and somatic production

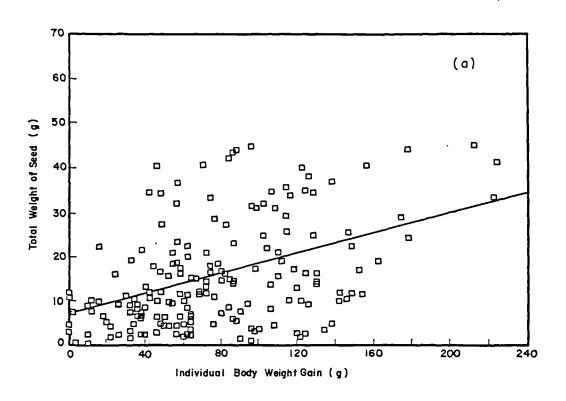
The total weight of eggs obtained from females spawned in hapas over a range of treatments was a larger proportion of total production (25.4%) than fish spawned in tanks (19.5%) (Table 2.18). However, the range of seed production (expressed as seed weight as a percentage of total production after the first spawning) between the 63 individual fish which spawned in hapas varied widely, from under 3% to 100% of total production. Seed production in both tanks and hapas was inversely related to total production (Fig. 2.15); total weight gain was defined as the total body weight increment from the first spawning to the end of the experiment plus total seed weight harvested.

The mean total weights of seed spawned in tanks and hapas were not significantly different (P>0.05; Table 2.18). In hapas there was no relationship between seed production and

Fig. 2.16

Relationship between total weight of seed harvested and individual body weight gain (from the harvest of the first seed clutch to end of experiment; 116 days) of female fish spawned over a range of treatment in

```
(a) tanks (T21 - T24),
y = 6.48 - 0.166x, n = 180, r = 0.2, P < 0.01</li>
(b) hapas (H11 - H14),
n = 66, r < 0.1, P > 0.05
y = a + bx, where y = total weight of seed harvested
(g) and x = individual body weight gain (g)
```



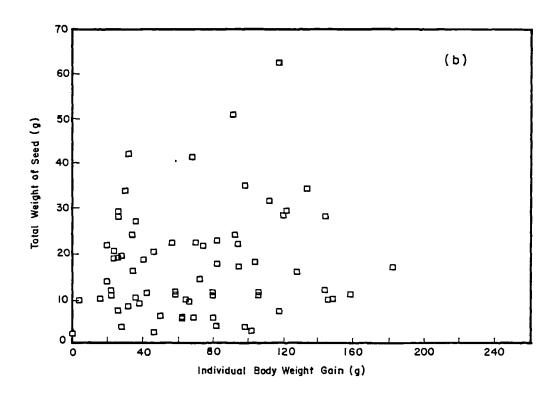


Table 2.19 Observed interspawning interval (ISI) of individual tagged female broodfish¹ maintained in tanks and hapas² under different exchange and conditioning regimes

All treatments	Mean	±	S.E.	
T21	25.09	±	1.81	bcde
T22	21.15	±	1.61	abcd
Т23	27.28	±	1.77	de
T24	25.95	±	1.65	bde
H11	21.37	±	2.39	abcd
H12	15.38	±	0.09	abcd
Н13	20.17	±	1.12	abcd
H14	29.37	±	1.52	C
F 4.78***				
All hapa spawned treatments	23.77	±	0.95	
All tank spawned treatments	24.70	±	0.86	
F 0.45 ^{n.s.}	}			
All Hapa conditioned treatments	24.25	±	0.88	
All Tank conditioned treatments	24.58			
All partial exchange treatments	26.58	±	0.87	
All total exchange treatments	21.78	±	0.97	
F 13.67**				

Any 2 means having a common letter are not significantly different (P > 0.05, Duncan's Multiple Range Test)

F value one-way Anova

- $n \cdot s \cdot P > 0.05$
- * P < 0.05
- ** P < 0.001
- *** P < 0.0001

¹ Non-spawning fish not included

² Tanks (Area = 12.57 m², T2) and hapas (Area = 40 m², H1) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days

individual body weight gain over the experimental period ($r^2 = 0.02$) (Fig. 2.16). In tanks from the spawning histories of 183 fish, seed production was positively related to individual body weight gain over a range of treatments (P<0.01; Fig. 2.16).

(d) Interspawning Interval (ISI)

The Interspawning Interval between two seed harvests from females spawning in tanks and hapas showed significant differences between the eight treatments (Table 2.19) but these were not related to the type of spawning unit. ISI was greatest where spawned females were exchanged in hapas (H14). Hapa treatments tended to have lower ISI's in three treatments (H11, H12 and H13) but taken as a group there was no significant difference between tanks and hapas (Table 2.19).

When the broodfish spawning in tanks and hapas were compared as a single group, conditioning environment had no effect on ISI (Table 2.19). However, female replacement strategy had a considerable effect on ISI; the mean interspawning interval for replacement of spawned females only, was 22% longer than if all females were replaced (Table 2.19).

(e) Gonadosomatic index

Gonadosomatic index (%) was calculated from the formula:

The GSI of females that had spawned in the period prior to harvest was less than half that of unspawned females harvested from the same spawning unit (Table 2.20). Unspawned females had

Table 2.20 Gonadosomatic index¹ at final harvest of female broodfish maintained under different exchange and conditioning regimes (a) in spawning and conditioning tanks and hapas (b) after exchange of either all or spawned females and (c) of unspawned females harvested from spawning tanks and hapas

Category of female	Treatment	GSI Mean + S.E.
(a) Spawned females from spawning tanks	(all)	1.68 ± 0.089a
Unspawned females from spawning tanks	(all)	3.67 ± 0.30^{b}
Conditioned females for 10 days	(all)	3.35 ± 0.30^{b}
Conditioned females for 5 days	(all)	3.40 ± 0.26^{b}
(b)		
Females from spawning tanks	$(T21 + T22)^2$	1.96 ± 0.25 z
Females from conditioning tanks	$(T21 + T22)^2$	$3.77 \pm 0.42 P$
Females from spawning tanks	$(T23 + T24)^3$	$2.63 \pm 0.41 \text{ yz}$
Females from conditioning tanks	$(T23 + T24)^3$	2.51 ± 0.23 q
Females from spawning hapas	$(H11 + H12)^2$	3.22 ± 0.43 y
Females from conditioning hapas	$(H11 + H12)^2$	$3.70 \pm 0.46 P$
Females from spawning hapas	$(H13 + H14)^3$	3.23 ± 0.45 y
Females from conditioning hapas	$(H13 + H14)^3$	3.61 ± 0.28 P
(c)		
Unspawned females from spawning hapas	(all)	4.40 ± 0.388 c
Unspawned females from spawning tanks	(all)	2.94 ± 0.313 d

Any 2 means having a common superscript in the same group are not significantly different. (P.> 0.05, Duncan's Multiple Range Test)

F = 5.6 (P < 0.0001) for all categories together (one-way Anova)

¹ GSI = weight of reproductive organ x 100/weight of fish

² Exchange of all females

³ Exchange of spawned females only

^{*} Spawning tanks (Area = 12.5 m², T2) and hapas (Area = 40 m², H1) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days. Conditioning tanks (Area = 1.77 m², T2) and hapas (Area = 5.4 m², H1) stocked at 40 fish per unit

Table 2.21 Crude protein, total lipid and moisture content of whole ovaries from spawned and unspawned broodfish spawned in tanks* and hapas under different exchange and conditioning regimes at final harvest

Harvested Female	Mea	an percentage ± S	5.D.
narvested relate . (n)	Moisture	Crude protein (D.M.)	Crude lipid (D.M.)
Spawned (7)	52.3 ± 0.65	46.2 ± 2.8	31.1 ± 7.6
Unspawned (21)	53.1 ± 0.63	50.0 ± 2.2	27.1 ± 3.3
F Value	8.5**	10.6**	3.8n.s.

Level of significance of one-way Anova

D.M. = Dry Matter

 $n \cdot s \cdot P > 0.05$

^{**} P < 0.01

^{*} Tanks (Area = 12.57 m^2 , T2) and hapas (Area = 40 m^2 , H1) stocked at 80 fish per unit (female:male ratio = 1); t = 116 days

similar GSI values as females conditioned for 5 and 10 days. The GSI of females removed as a group (T21 and T22) from spawning tanks (both spawned and unspawned) had significantly lower GSI's than similar fish removed from hapas (1.96 compared to 3.22)(Table 2.20).

Broodfish in treatments exchanging spawned females only in tanks (T23 & T24) had a significantly lower GSI (2.51) after a period of conditioning than other treatments (Table 2.20).

The mean GSI of unspawned females taken from the arena after 5 days was considerably higher in fish from hapas (4.40) compared to tanks (2.94) (Table 2.20).

Proximate analysis of ovaries from spawned and unspawned females showed significant differences (P<0.05) in total lipid, crude protein and moisture content (Table 2.21), the smaller ovaries of recently spawned females being higher in lipid and lower in moisture and crude protein.

2.4.4 Discussion

Hierarchical effect in green and clear water systems

The main differences in spawning characteristics between fish in tanks and hapas can be explained by the relative strengths

of their hierarchies which may be affected by water clarity and

fish density.

In the present experiment, densities of broodstock were higher in tanks than hapas $(0.55-0.90 \text{ kg/m}^2 \text{ and } 0.18-0.25 \text{ kg/m}^2 \text{ respectively})$ but were below average within the range published in terms of number or biomass (Balarin and Haller, 1982). Recirculation of tank water through a biofilter made water quality more controllable than in hapas suspended in ponds and shading ensured a low phytoplankton density in tanks relative to hapas.

The evidence from the tagging of broodstock strongly suggests that a more rigid hierarchy existed in the tanks than the hapas which affected spawning activity considerably. The normal distribution of spawning frequency in hapas contrasted with the low frequency or complete non-involvement of most of the fish in tanks with 65% of the tank fish spawning on three occasions or less over the whole experimental period.

The long term studies by Fishelson (1983) and Turner (1986) suggested that hierarchies in clearwater tanks are frequently dominated by a few larger fish over extended periods of time.

Hierarchies are maintained by mainly visual, agonistic behaviour (Aronson, 1949; Heiligenberg, 1965; Baerends, 1986; Turner, 1986), and are thus likely to be less rigid in hapas where the density of fish is lower and the water more turbid. Lowe McConnell (1959) observed that in Lake Nyasa, each species of tilapia tended to spawn in the clearest water zone of its range and to feed in the most eutrophic part, indicating the importance of breeding colours in territory advertisement and assisting synchronisation of male and female cycles. However, as sound (Marshall, 1972; Lanzing, 1974; Silverman, 1978c) and chemical stimuli (Silverman, 1978c) have been found to have a role in mate identification and spawning, they may also be important in hierarchy establishment and stability.

Hulata et al. (1985) found that the incompatibility between species in hybrid production could be reduced in green water compared to clear water suggesting that visual cues are normally of primary importance but are reduced in a more turbid water environment, making reproductive behaviour less discriminatory.

Lee (1979) described the hierarchies and agonistic behaviour between females in aquaria; competition between females resulted in a vertical hierarchy with the weaker females being displaced to the upper water level above the 'stronger' females. The subordinate fish were characterised by darker colouration and vertical bands indicating stress; they also suffered higher mortality after intensive nipping by the males. Several authors have observed that the hierarchy between

spawning groups in aquaria caused some dominant females to spawn more than others (Fishelson, 1966; Mires, 1973; Rothbard and Pruginin, 1975).

Synchrony of spawning

The improved synchrony of spawning in hapa systems compared to tanks might be explained in terms of the weaker hierarchy in the lower density and more turbid pond water environment. There was no significant effect of conditioning environment on synchrony, further suggesting that behaviourial factors within the spawning arena were the main control.

Egg production parameters

The larger clutch size of eggs produced by females spawning in tanks than in hapas suggests that interruption of spawning and/or egg stealing, as described by Balarin and Haller (1982) and Turner (1986), was reduced in the more hierarchical tank system. Relative clutch size was also higher in the tank system, although not significantly so (P>0.05), indicating that this effect was not related to body weight.

In contrast, the larger clutch weight, clutch size and relative clutch size were higher for females conditioned in hapas, indicating an environmental effect on ovarian development. A larger egg size and smaller clutch number have been correlated with a high protein diet (42.5% and 50% crude protein) in O. niloticus (Tuan, 1986) and food restriction in O. mossambicus (Miranova, 1978). In the present case, a reduction in egg size

by under 10% was matched by an increase in clutch size by 14%. The relative advantage of a larger egg size improving fry survival under conditions of starvation has been documented for O. niloticus by Rana (1988), but longer term advantages are disputed. Siraj et al.(1983) observed no differences in performance (growth and survival) after 20 days post hatching under well-fed conditions, whereas Cridland (1962) and Rana (1988) reported that size advantages continued under optimal conditions for three months in O. spilurus and two months in O. niloticus respectively.

Growth and reproductive output of broodstock

Growth of spawning fish was inversely related to reproductive output. Non-spawners tended to be larger (length and weight) than either moderate or frequent spawners. The condition factor (CF) of moderate spawners was significantly greater than that of frequent or non-spawners. This suggests that the moderate spawning fish were ready to spawn but were inhibited by more dominant fish; and that the non-spawning fish were excluded from sexual activity almost completely and substituted body growth for reproductive output. This phenomenon has been previously reported for subordinate males in groups of breeding O. mossambicus (Turner 1986).

Frequent spawners would need to be aggressive feeders to maintain reproductive output and this trait may also be accentuated by the tank environment. Frequent spawning females conditioned in tanks showed significantly improved condition compared to similar fish in hapas, even though the

opportunities for natural feeding were lower.

The relationship between reproductive and somatic output is illustrated by comparing seed output to body weight gain. Fish breeding in hapas showed a higher allocation to reproduction as more than 25% of total production was seed compared to less than 20% for fish in tanks.

Lee (1979) found that 35% of total production from nine individual females (O. niloticus) was in the form of eggs taken from the mouth the second day after spawning. Three female fish were maintained at high density in an aquarium with a single male and fed a high quality feed. Data recalculated from Tuan (1986) showed that reproductive output as a percentage of total production ranged from 12.4-20.9% depending on crude protein (CP) content of the diet (CP=27.5%, egg output=20.9%). Eggs removed daily and constituted a similar level were production as in the current study. The higher reproductive output found by Lee (1979) suggested that producing seed from small groups of females at high density was possible because of the extra 'male pressure'. The females were stocked at very high density, perhaps enough to reduce hierarchy between them, and to keep them constantly within the males' territory so that they were always in view of the males' courtship. Silverman (1978) found that the major effect of the male during courtship was to trigger ovulation of the female. The relative importance of hierarchical effects in tanks was again indicated by the trend for fish gaining most weight to also have a higher reproductive output. Fish in hapas, on the other hand showed no

significant relationship between weight gain and reproductive output.

Interspawning Interval (ISI) and frequency of spawning
Mean Interspawning Intervals (ISI) for females in both tanks
and hapas were within the range recorded by other workers for
intensive systems (as reviewed by Rana, 1988) despite fish
being conditioned for more than 60% of the experimental period.
The shortest ISI's recorded for <u>O. niloticus</u> were by Lee (1979)
and Rana, (1986) for small numbers of females maintained in
aquaria and with intensive feeding and rapid removal of eggs
after fertilisation (<48 hours).

High variability of spawning frequency amongst individuals was found in both tanks and hapas and this has also been reported for broodfish held in aquaria (Mires, 1982; Macintosh and Sampson, 1985). The management of these treatments, with 10 days conditioning between each period of 5 days spawning opportunity, meant that the minimum possible recorded ISI was 15 days.

The shorter ISI recorded for treatments in which all females were exchanged, as compared to only spawned females, indicates the importance of giving all females an opportunity to spawn regularly. Exchanging only spawned females probably allowed some unproductive (subordinate?) females to remain in the unit for long periods. The spawning of females without males is a common phenomenon (Aronson, 1949; Marshall, 1972; Silverman 1978a, 1978c; Mires, 1982) amongst isolated females in aquaria, although the ISI is reportedly increased (Jalabert and Zohar,

Table 2.22 Mean ISI of all females held in spawning units* under different exchange and conditioning regimes

Mean IS	I ± S.D. (n=3)
T21	55.7 <u>+</u> 2.3
T22	45.9 <u>+</u> 5.5
T23	47.5 ± 1.0
T24	49.7 <u>+</u> 6.7
H11	50.5 <u>+</u> 17.8
H12	43.5 <u>+</u> 13.8
H13	41.9 <u>+</u> 5.5
H14	53.7 <u>+</u> 11.6

F value one-way Anova = $0.72^{n \cdot s}$.

n.s. P > 0.05

^{*} Tanks (Area = 12.57 m^2 , T2) and hapas (Area = 40 m^2 , H1) stocked at 80 fish per unit (female:male ratio = 1); t=116 days

1982). Although this probably occurred to some extent in the conditioning nets, females were never found mouthbrooding eggs which suggests that the high stocking density may have inhibited courtship and oviposition.

The observed mean ISI's of observed females did not include non-spawning females. The mean ISI based on the average frequency of spawning of all the fish over the experimental period revealed that there was no significant difference between treatments (P>0.05, Table 2.22).

Gonadosomatic index (GSI)

The concept of a 'critical gonadosomatic index' (GSI) for females of Oreochromis spp. was expounded by Silverman (1978a) to describe the state of ovarian development immediately prior to ovulation. He observed that GSI did not change during the week of incubation after spawning but thereafter increased rapidly to a value of about 4% in O. mossambicus. No further increase was observed, but from this stage and given the 'proper stimuli' ovulation could occur at any time. critical GSI appears to be dependent on size since Peters (1983) reported GSI's for 'ripe' O. mossambicus to be between 4.9 and 10.2 for small broodfish (3.7-24.6 g). The mean GSI of spawned females found in this experiment (<2.0) is of the same order as found by Silverman (1978a) and Peters (1983) (Table 2.20). However, it is unclear whether the similar GSI's of unspawned females recovered from the spawning unit, and after various periods of conditioning (5 and 10 days) were below the critical level for O. niloticus in this experiment since the

GSI's of unspawned females in hapas were in excess of 4%.

It appears from the lower GSI of females in tanks compared to hapas that fish in condition to spawn did so more completely and that there was possibly more partial and/or interrupted spawning in hapas (higher mean GSI after spawning). These data are based on GSI estimations at harvest only and so it cannot be assumed that such GSI's were maintained throughout the experimental period. However, it concurs with the larger clutch size observed in tank spawned fish and with the hypothesis that hierarchies were more rigid in clearwater tank systems at this stocking density. Moreover the longer ISI of spawned females exchanged in tanks can be related to the significantly lower GSI of these females after conditioning.

Table 2.23 Experimental design used to study effect of conditioning period and male and female broodstock exchange on spawning in nylon hapas suspended in earthen ponds (H3)

System/ Experi- ment/	Number of	fish	Туре	Restor		Exchanged condition		Interv (day:	
Treat- ment	per spawning	total	Type	Tank	Hapa	Tank	Hapa	Spawning	Condi- tioning
н31	40 40	40 120	males spawned females		x		x	5	10
	40	120	unspawned females				x		
Н32	40	40	males spawned females		х			5	15
	40	160	unspawned females				x	ə	15
н33	40	40	males spawned		x			5	80
	40	200	females unspawned females				x	5	20
H34	40	40	males spawned		х			5	10°
	40	<120	females unspawned females		Xª		x _p	j 5	100
Н35	40	120	males spawned				х	_	16
	40	120	females unspawned females				x	5	10

^{*} Females in ripe condition

b Females in unripe condition

c minimum of 10 days

x denotes restocking and conditioning strategy

2.5 The effect of duration of female broodfish conditioning, selective female and male broodfish exchange on seed production in large nylon hapas

2.5.1. Methods

(a) Origin and pretreatment of broodstock

Broodstock were obtained from an experiment on broodfish production (3.1) in which fry of the same origin were raised to maturity under different regimes of supplementary feeding in earthen ponds. Broodfish were raised in earthen ponds fertilised with septic tank slurry and fed a supplement of a high quality floating catfish pellet (crude protein 30%), a floating herbivorous fish pellet (crude protein 16%) or fine rice bran. Equal numbers of fish were taken from the different treatments of this experiment.

(b) Stocking and harvest

Broodfish were maintained in the spawning nets at a standard total number of 80 fish per net (Table 2.23). Females and males were stocked in conditioning nets according to treatment at a standard density of 40 fish per net. All female broodstock were removed and replaced with conditioned females after 5 days spawning opportunity in Treatments H31, H32, H33 and H35 (Table 2.23).

In Treatment H34, two categories of females: (a) spawned females, and (b) unspawned but unripe females were removed at harvest and replaced with an equal number of females selected for ripeness from fish in the conditioning net.

The main criterion for selection of ripe females was a soft, rounded abdomen which was taken to be an indication of readiness to spawn, while a firm, narrow abdomen was the rationale for removal of category (a) and (b) females.

Eggs and fry were staged at harvest and preserved as combined batches.

Table 2.24 Seed production in hapas* using different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)

		MEAN	(<u>+</u> S.E.)	
Treatment	Total No. of seed per harvest per hapa	No. of seed per day per hapa	No. of seed per kg female per day	No. of seed per kg fish per day
Н31	12747ab	2854ab	233.2 ⁶ c	133.0ab
	(1092.0)	(252.5)	(27.0)	(21.3)
Н32	13092ªb	2673ªb	175.9ªc	108.7b
	(1214.9)	(287.4)	(23.9)	(19.7)
н33	10408ª	2131ª	117.6ª	61.4 ^b
	(1109.9)	(263.2)	(17.0)	(11.8)
Н34	17700ª	3671 ^b	319.9 ^b	199.1ª
	(2730.8)	(649.9)	(59.0)	(54.2)
н35	15825b	3179ab	272.4bc	119.7ªb
	(1521.0)	(334.4)	(32.8)	(13.9)
F	3.02*	2.39n.s.	5.13***	2.86*

F value one-way Anova.

Means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's multiple range test)

n.s. P > 0.05

^{*} P < 0.05

^{***} P < 0.001

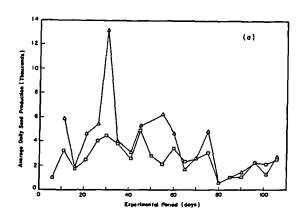
^{*} Spawning hapas (Area = 40 m^2 , H3) stocked at 80 fish per unit (female:male ratio = 1); t=106 days

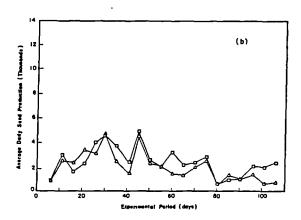
Fig. 2.17

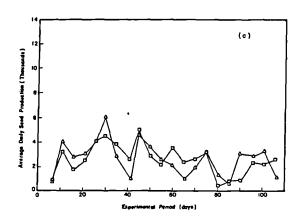
Mean daily production of seed per hapa (Area = 40 m^2) for treatments exchanging female broodfish completely after 10 days conditioning (H31; square) and

- (a) selectively exchanged females (H34; triangle) after a minimum of 10 days conditioning
- (b) completely exchanged females conditioned for 20 days (H33; triangle)
- (c) completely exchanged females conditioned for 15 days and (H32, triangle)
- (d) completely exchanged males and females conditioned for 10 days (H35, triangle)

Interharvest Interval of 5 days for all treatments







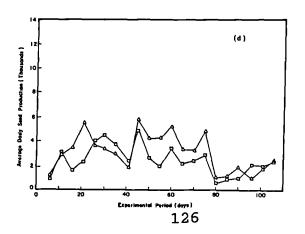
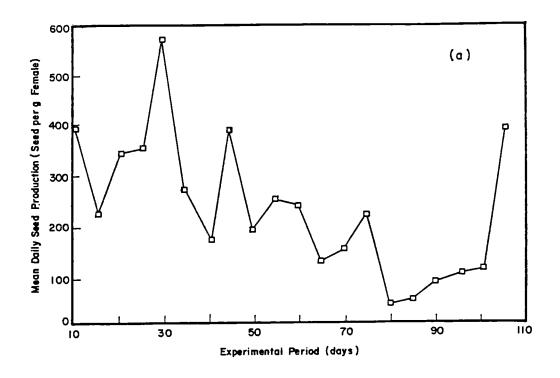
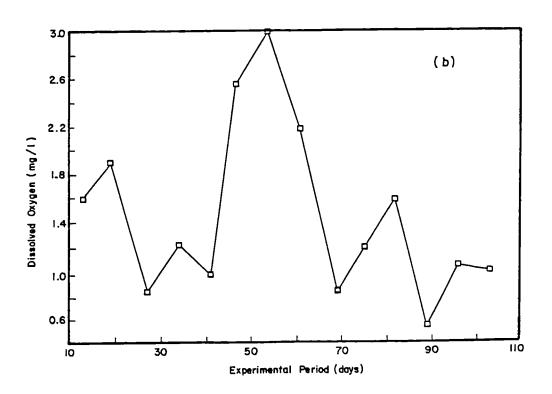


Fig. 2.18

Variation in

- (a) mean daily production of seed per ${\tt g}$ of female broodfish and
- (b) mean dissolved oxygen level at dawn in spawning and conditioning hapas under different broodstock conditioning and exchange strategies over the experimental period (all treatments combined)





2.5.2.Results

(a) Seed production

The following parameters were estimated (a) total number of seed per harvest per hapa; (b) mean daily seed output per hapa; (c) number of seed per kg total females; and (d) number of seed per kg total fish. Production parameters of the three replicates of each treatment, after evaluating for differences using Analysis of Variance (Anova), were pooled and means compared (Table 2.24). As the fish in each replicate were stocked in the spawning net a minimum of six times, the observed large variation in egg output with time probably obscured differences between treatments.

Mean daily seed output revealed some differences between treatments. Broodfish conditioned for 20 days (H33) produced less seed on a daily basis (2131 seed/hapa/day) than male exchange (H35; 3179 seed/hapa/day); although there were large differences between means of other treatments they were insignificant at the 5% level of probability (Table 2.24, Fig. 2.17). Long term conditioning also produced fewer eggs per kg of female broodstock and per kg of total broodstock than other treatments.

Variation in water quality may have contributed to the variability of egg production. Low early morning dissolved oxygen levels (<1mg/l) occurred in spawning nets throughout the experimental period although drops in egg production were not well correlated with very low oxygen levels (Fig.2.18).

Data were logarithmically transformed before Analysis of

Table 2.25 Mean percentage of total seed harvested as uneyed eggs (stage 1) from spawning hapas* stocked with broodfish managed under different conditioning and exchange regimes

Treatment	Mean percentage of total harvested stage 1 seed (n)
н31	32.8 (20)
H32	23.2 (20)
Н33	34.4 (20)
Н34	31.2 (19)
Н35	31.8 (19)
Chi-Square	5.1 ^{n.s.}

Chi-Square value of the Kruskal-Wallis one-way Anova $_{\text{n.s.}}$ P > 0.05

^{*} Spawning hapas (Area = 40 m^2 , H3) stocked at 80 fish per unit (female:male ratio = 1); t = 106 days

Table 2.26 Parameters of seed production for female broodfish spawned in hapas* after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)

		ME	AN ± S.E.	
Treatment	Egg weight ¹	Clutch weight ² (g)	Clutch size3	Number of eggs per g female
(a) All sta	ges combined			
Н31	0.37 ± 0.006	3.03 ± 0.13	673 ± 58.7 a	8.0 ± 0.62
H32	0.39 ± 0.006	3.02 ± 0.17	$704 \pm 38.9 \text{ a}$	7.8 ± 0.68
н33	0.39 ± 0.006	2.83 ± 0.13	$721 \pm 34.4 =$	6.4 ± 0.42
H34	. 0.38 ± 0.008	3.38 ± 0.18	872 ± 44.6 b	8.2 ± 1.10
H35	0.38 ± 0.007	3.08 ± 0.16	844 ± 39.8 b	8.1 ± 0.72
F	1.88n.s.	1.70 ^{n.s.}	5.10**	1.00n.s.
				
	0.38 ± 0.058 b 0.36 ± 0.049 a 0.36 ± 0.042 ab 0.42 ± 0.053 c	3.47 ± 0.13 a 3.03 ± 0.11 b 2.79 ± 0.13 b 2.96 ± 0.17 b	942 ± 35.5 a 762 ± 35.4 b 748 ± 25.5 b 589 ± 36.7 c	

F value one-way Anova

mean weight of 100 fresh seed wet weight
 mean weight of all seed in a single clutch wet weight
 total number of seed in a single clutch total number of seed in a single clutch means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

n.s. p > 0.05

P < 0.001

P < 0.0001

^{*} Spawning hapas (Area = 40 m², H3) stocked at 80 fish per unit (female:male ratio = 1); t = 106 days

Variance. The selective replacement of female broodstock (H34) was the most productive treatment on the basis of average output (3671 seed/hapa/day, 320 seed/kg female/day) but the variance of the means was also particularly high.

(b) Synchrony of spawning

There were no significant differences in synchrony of spawning between treatments (F=0.85, P=0.49; Table 2.25). Late spawning occurred within a range of 23-35% of total spawns and early spawning as indicated by the harvest of well developed seed, between 40-60%.

(c) Seed production parameters

Seed size did not vary significantly between treatments, (F=1.88, P>0.05) (Table 2.26).

Seed from females selected for ripeness (H34) had a mean clutch weight (3.38 g) larger than females conditioned for the longest period (H33, 2.83 g), although values were not significantly different from other treatments (Table 2.26).

On average, clutch sizes from treatments using selective exchange of females and male exchange (H35) were significantly larger by 30 and 25 percent respectively than clutches from the treatment with a simple exchange strategy (H31) (Table 2.26). Mean clutch weight and size varied significantly with egg stage. Late spawned clutches of eggs tended to be both heavier and larger than early spawned eggs/hatched fry (Table 2.26), even though hatched larvae (stage 4) were individually heavier than eggs on a wet weight basis. Clutch weight variability was

Table 2.27 Mean frequency of seed harvest and ISI of female broodfish spawned in hapas¹ after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35)

		MEAN + S.E. ²	
Treatment	Total number of spawns during experiment	No. of spawns/day in spawning hapa	ISI (days)
H31 H32 H33 H34 H35	116.0 ± 5.13 ab 88.5 ± 8.35 b 64.6 ± 4.35 c 129.3 ± 18.68 a 141.3 ± 12.55 a	3.29 ± 0.15 a 3.33 ± 0.32 a 3.05 ± 0.21 a 3.66 ± 0.53 a 4.00 ± 0.36 a	36.7 ± 1.7 ab 49.2 ± 4.6 b 67.0 ± 5.1 b 34.4 ± 5.6 a 30.4 ± 2.5 a
F	11.19**	1.40n·s·	11.36**

¹Spawning hapas (Area = 40 m^2 , H3) stocked at 80 fish per unit female:male ratio = 1); t = 106 days

 $^{2}n = 3$ (H31, H34, H35); n = 4 (H32); n = 5 (H33)

Means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

F value one-way Anova

- n.s. P > 0.05
- * P < 0.05
- ** P < 0.01

Individual size and growth characteristics at (a) stocking and (b) final harvest of broodfish spawned in hapas* after different periods of female conditioning (H31, H32, H33) and selective female (H34) and male broodstock exchange (H35) Table 2.28

(a) Females Males	Total length (cm) (cm) 14.5 ± 0.05 a 15.7 ± 0.13 b 83.2**	angth 0.05 a 0.13 b	Body weight (g)	ight	Condition factor	
(a) Females Males	14.5 ± 0 15.7 ± 0 83.2*),05 a),13 b	56.7 + 0			i factor
	•		72.2 ± 1.8 b 91.5**	.64 a 8 b	1.79 ± 0.01 a 1.80 ± 0.15 a 0.59n.s.	0.01 a
(a)						
					Growth g/day	rth lay
Treatment Fe	females	Males	Females	Males	Female	Male
H32 . 20.7	7 ± 0.16 a 8 ± 0.14 a	23.1 ± 0.22 a 22.9 + 0.24 a	154 ± 3.7 ab	205 ± 5,1 a	0.92	1.18
	+ 0.13	23.1 ± 0.26 a	162 ± 3.1 a	208 ± 6.4 m	0.96 0.98	$\frac{1.20}{1.24}$
	+ 11	23.5 ± 0.25 m	149 £ 5.3 b	217 ± 6.8 a	0.88	1.53
	. 9n.s.	15.7**	150 ± 4.1 b 2.03n.s.	168 ± 4.9 b 14.25**	0.89	0.82

Means in the same group with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

F value one-way Anova n.s. P > 0.05 ** P < 0.01 * Spawning hapas (Area = 40 m², H3) stocked at 80 fish per unit (female:male ratio = 1); t = 106 days

eggs. Clutch size showed a decreasing trend with stage of development, and therefore with the duration of natural mouthbrooding before harvest. Relative clutch size also declined with time and was similar between all treatments.

.(d) Frequency of spawning and ISI

Calculation of spawning frequency and ISI were based on the mean numbers of spawned fish over the total experimental period (106 days) for each replicate. Comparing conditioning periods of 10 and 20 days showed that the total number of spawns decreased with duration of conditioning and the mean interspawning interval almost doubled between a 10 and 20 day conditioning period (Table 2.27).

A longer conditioning period inevitably reduced the total time spent by individual fish in the spawning net from 35 days (10 day conditioning period) to only 21 days (20 day conditioning period) during the experimental period. But there was no significant difference between treatments if the frequency of spawning were considered in terms of number of spawns per unit time of spawning opportunity (Table 2.27).

(e) Feed input and growth

Initial body weights of male broodfish were more than 25% heavier than females (72.2 g and 56.7 g respectively) but their mean condition factors were similar (Table 2.28).

Final size was similar in all treatments except for long term conditioned females (H33) which were larger; and males used in

the replacement strategy (H35) which were smaller.

Individual female growth rates over the experimental period were below 1 g/day for all treatments (0.88-0.98 g/day). Male growth rates were higher but showed greater variation between treatments (0.82-1.53 g/day). Males in the treatment exchanging males showed least growth (0.82 g/day; H35), while the highest growth was shown by males in the selective female replacement group (1.53 g/day; H34; Table 2.28).

2.5.3 Discussion

The type of broodfish exchange used to increase spawning synchrony and productivity in tanks and hapas has consequences for ease of management and cost of fry production. Ideally, the optimum strategy would maximise breeding activity and enhance readiness to spawn, whilst reducing the number and hence cost of broodstock. The previous experiment (H11-H14) suggested that the mean ISI for fish spawned and conditioned in hapas was of the order of 15-20 days. In previous studies, broodfish were exchanged in hapas (Lovshin and Ibrahim, 1987; Batao, 1988) after an interharvest and conditioning period of 21 days. The number of broodfish required was therefore lower than for the more frequent harvesting strategies reported here.

An extended period of spawning and conditioning resulted in most broodfish naturally incubating their eggs to swim-up stage and suffering a loss in condition accordingly.

The presence of some unripe females in both spawning and conditioning nets after five and ten days respectively, suggested that exchange of all female fish, or of only spawned fish, did not always optimise the condition of fish in the spawning net. The selective female strategy (H24) therefore attempted to optimise mean female condition in the spawning net by removing unspawned and unripe fish in addition to spawned females at harvest, and exchanging them for the ripest fish from the conditioning net.

A regular exchange of male broodfish could also have potential in maintaining a high level of sexual activity in the spawning arena. Readiness to spawn and fry production have been assumed to be dependent only on female condition (Uchida and King, 1962; Peters, 1983) but the demands made on males when spawned females are continually replaced with conditioned, ripe females is likely to be greater. Moreover, regular spawning may be constrained in intensive systems by a tendency for females to prefer unspawned males with ripe gonads (Silverman, 1978a, 1978b). Nakatsuru and Kramer (1982) reported that females tended to chose males that had not recently spawned in the lemon tetra (Hyphessobrycon pulchripinnis).

The hapa is more sensitive to changes in environmental conditions than static or recirculated water tanks; this can lead to greater variability in fry production. Santiago et al. (1985) and Bautista et al. (1988) found that tilapia fry production from hapas suspended in Laguna de Bay, Philippines was more variable than in nearby concrete tanks, probably because of seasonal deterioration in water quality in the lake itself.

The high degree of variation in seed production in this experiment was probably due to variable water quality, especially early morning dissolved oxygen in the ponds in which the hapas were suspended. Dissolved oxygen ranged from a maximum of 3 mg/l to a minimum of 0.6 mg/l in the spawning nets throughout the experimental period; the effect on fry production through poorer ovarian development depressed

spawning activity, poorer incubation success or a combination of these factors.

The significant increase in ISI for periods of conditioning over ten days can be explained largely by the longer period of time the fish were outside the spawning net and thus unable to breed. Frequency of spawning did not actually change significantly, on the basis of spawns per day, of opportunity to spawn, or actual spawns per spawning period, across the range of treatments. There was no evidence that a period of conditioning longer than ten days increased or decreased the ability to spawn, or that exchanging males or females on the basis of visual maturity increased spawning activity.

There were no significant differences between egg yield and period of conditioning except for the number of eggs per kg of female per day. Long term conditioning of female broodfish resulted in a significantly lower relative output of eggs largely because the number of female broodstock required was double that necessary for a standard period of conditioning (10 days). The tendency for selective female (H34) and male (H35) exchange to show higher productivity was not significant (P>0.05).

Variation in productivity indicators was greatest for selective replacement of females, suggesting that the maintenance of ripe females ready to spawn was not always optimised. However, this was certainly influenced by the experimental design. If a high proportion of females harvested spawned, a large number had to be removed from the spawning hapa and exchanged with fish from

the finite number maintained in the conditioning net. The average degree of ripeness of exchanged fish therefore tended to decline as a function of the number required since fish were selected by simple visual ranking. The variability in the condition of fish selected for exchange could have been reduced by increasing the number of fish from which the selection was made.

Selective female (H34) and male replacement (H35) resulted in larger mean clutch size than treatments using simple replacement of all females after a standard period of conditioning (H31, H32, H33). Peters (1983) stated that 'sufficient conditions' for spawning in Oreochromis spp. were often lacking and that reabsorption of eggs was a common phenomenon. In this study, treatments in which females spawned after a fixed period of conditioning i.e. 10, 15 or 20 days, may have produced a larger proportion of overripe, poorer quality eggs after an extended period of maturation. The larger clutch size from females selected for ripeness could have been a result of the selected fish spawning more completely and/or the eggs produced being of higher quality and thus surviving better. Peters (1983) observed that reabsorption began within as little as one week of reaching a critical GSI in Oreochromis spp. but that the beginning of egg reabsorption did not necessarily imply an end to the readiness to spawn. However the eggs from 'over-ripe' fish in which the maximum GSI had been attained for more than a critical period gave rise to poor embryo survival which would result in a smaller clutch size at

harvest.

The role of the males in tilapia fry production systems has frequently been underestimated largely because of the numerous reports of their ability to spawn several times within the same day (Lowe McConnell, 1959; Peters, 1971; Polder, 1971).

The production of seed in a variety of spawning units has been shown to be quite responsive to a reduced female to male ratio. Generally, a ratio of more than one is used and fry producers in the Philippines have used up to seven to one (Guerrero, 1985). Hughes and Behrends (1983) found that seed production was depressed by 73% in small hapas when the female to male ratio was increased (2:1 to 3:1). Mires (1982) suggested that 'male pressure' had an important effect on spawning success, especially in interspecific breeding. Observations of fish spawning in aquaria indicated that when two females were ready to spawn at the same time, and only a single male was available, the eggs from one female were frequently unfertilised. Even under extensive conditions when spawning may be less synchronised, Mireş (1982) found that the mean number of fry per female was increased by more than 30% after decreasing the ratio of females to males in earthen ponds (3:1 to 1:1).

The sex ratio at stocking may be a poor indicator of actual availability of spawning males; hierarchical effects may prevent subordinate males taking part in spawning activities in intensive systems.

The ability of <u>Oreochromis</u> spp. males to mate successively is countered by a declining capacity to fertilise successive clutches of eggs. This is in contrast to the orange chromide (<u>Etroplus maculatus</u>) in which fertilisation remains constant over successive breeding cycles (Lamon and Ward, 1983). Rana (1986) reported that infertile eggs increased to over 70% of the total clutch after the third or fourth spawning of the day for individual male <u>O. niloticus</u>. Similarly, Nakatsuru and Kramer (1982) described a linear decline in the number of developing eggs as a function of spawning acts by the male lemon tetra (Hyphessobrycon pulchripinnis).

Development of the testes is not associated with the large change in size and GSI associated with oogenesis in female Oreochromis (Silverman, 1978a) but cyclical changes do occur. Peters (1971) observed no measurable weight loss of the testes after spawning although Chao et al. (1987) have since used the monthly fluctuation of average GSI in Oreochromis spp. as the basis for timing the collection of milt for cryopreservation. Dadzie (1969) reported that the lobules within the testes in O. mossambicus were empty after spawning and a period of intensive spermatogenesis followed. The replacement rate of sperm by the testes is therefore critical to the effectiveness and efficiency of the male in spawning arenas.

The period of recovery has been estimated at two days for O. mossambicus (Rana, 1987) and four days for the cichlid Aequidens portalegrensis (Polder, 1971).

The fertilisation capability of a single male Nile tilapia

increased from 22% to 100% after a rest period of one week (Rana, 1987). The replacement of males with rested individuals may therefore increase mean clutch size by improving the level of fertilisation in spawned clutches of eggs and/or by increasing the level of vigour during courtship and spawning. Rana (1986) has proposed that polyandry by females may be a mechanism by which the risk of poor fertilisation is reduced i.e. it is an adaptation to the males' declining ability to fertilise eggs during successive courtship and matings.

In this study, the replacement of males had no significant effect on spawning synchrony indicating that the time required for males to establish territories and begin courtship was not different to the non-exchange treatments. The evidence presented here, and by Batao (1988) and Lovshin and Ibrahim (1987) suggest that the conditioning period of males might be reduced to five days and to two groups of fish, with consequent savings in costs.

Rana (1986), in a study of natural mouthbrooding, described the cumulative level and type of damage to fry occurring during incubation in the buccal cavity. This study indicated that losses during incubation had a significant effect on clutch weight and size at harvest. Clutch weight and size of stage 1 eggs, in which infertile eggs and batches of eggs were included, were significantly larger than later stages, indicating the removal of unfertilised eggs from the buccal cavity. Losses after hatching were also large, the mean clutch size declining by approximately 20% below the pre-hatched size.

Contact with the pharyngeal teeth and dislodgement of the yolk-sac during the churning movements of incubation was the most common form of damage observed by Rana (1986) in the intensive tank and aquarium systems in which the fish were maintained. Marshall (1979) found no relationship between the number of eggs in the mouth and the egg stage after the harvest of O. macrochir by seine netting in a natural lake, but he rejected females that appeared to have already lost eggs or fry. Other authors have associated the decline in fry numbers during mouthbrooding to the limited size of the buccal cavity, but Rana (1986) demonstrated that this was not a limitation.

CHAPTER 3

EFFECT OF EARLY BROODSTOCK PRODUCTION AND STOCKING DENSITY IN SPAWNING TANKS AND HAPAS ON SEED PRODUCTION

3.1 Broodstock production

3.1.1 Background

The production and maintenance of good quality broodstock is an important feature of any livestock breeding system. Feeding strategies for (1) breeding and (2) fattening are different, the former requiring sound development of the reproductive system and the latter an economic growth rate (McDonald et al. 1981). This has led to a high degree of development in animal husbandry towards early selection and separate production of replacement stock. In contrast, the continuing reliance on wild spawning stock for the reproduction of many fish and shrimp means that fry producers have little control over this stage of production. In some species, such as the giant freshwater prawn rosenbergii) and (Macrobrachium walking catfish (C. macrocephalus), broodstock are usually obtained from grow-out farmers. The potential advantages of early selection and specialised husbandry of high quality broodfish are thus neglected in favour of the convenience of buying fish that ripen easily under production conditions. The lack of development in broodfish management and nutrition for many fish species and a continued reliance on wild or unimproved stock illustrate the continuing links of aquaculture to its captural origins.

The use of breeding stock removed from a grow-out system can have serious consequences for the biological and economic efficiency of fry production, especially in multiple,

non-synchronous spawners such as the Nile tilapia.

The replacement of broodstock with even-sized fish from a grow-out system in which free breeding has occurred makes the selection of homogenous broodstock problematic as the selection of even-sized individuals will not necessarily produce broodstock of even age. Rana (1986) found that female age rather than body size was negatively correlated with relative fecundities in <u>O. niloticus</u>. The management of the broodfish as a single stock for egg production would therefore be difficult to optimise as younger fish would still be spawning at their peak when older fish were in decline.

Egg size correlates well with broodfish body weight and length, but Rana (1986) found that age has the predominant effect. As size of egg is well correlated to size of hatched fry, broodfish of mixed age would tend to produce eggs of variable size. This effect is known to continue at least through the fry rearing stage, and it would therefore lead to less variation in fry size. Several workers have related variation in fry size to increased levels of mortality from cannibalism (Macintosh and De Silva, 1984; Gregory, 1987).

Variation in broodfish age may also have indirect effects on fry production by altering the hierarchy and dominance within a spawning group. There is some evidence that a mixture of age classes/fish sizes does not inhibit but may well improve fry production. Hughes and Behrends (1983) found no significant difference in fry output between a low density of mixed age class tilapia (Year I and II) and a single age class of older

fish (Year II).

The use of broodfish of the same cohort from grow-out systems in which wild spawning is controlled by density or predators should lead to an improvement in the productivity of any spawning system, but the purposeful culture of selected broodstock under optimal conditions should theoretically yield further improvements. In Taiwan broodstock are selected at an age of two months and body weight of 20-30 g (Liao and Chen, 1983) from the first spawnings of the year and are then raised for use as spawners over the following two years.

Optimal stocking densities and feeding strategies which might be uneconomic for the production of table fish could still be highly desirable for broodstock production in terms of subsequent productivity and net profit (or income).

Scott (1962) was the first to investigate the relationship between feeding rate and fecundity in fish. Earlier studies had concentrated on the effects of the reproductive cycle on the use of body food reserves in wild fish (Luquet and Watanabe, 1986). In any intensive animal breeding system, a major objective is to ensure that one reproductive cycle follows closely or overlaps the previous cycle; and thus, the opportunities for the repletion of reserves after breeding are limited. Theoretically, feeding standards should be high and less reliance placed on body reserves (McDonald et al., 1981). Management for rapid growth and the early attainment of a size appropriate for breeding would also be advantageous. Food manipulation has been shown to modify the reproduction of

female broodfish in culture. In general a restricted diet during the early stages of the life cycle has delayed the age of maturation, while the egg number has been reduced by restricting feed during oocyte differentiation. However, food restriction during the last phases of oogenesis has only a limited effect on egg size, composition and hatchability (Luquet and Watanabe, 1986).

Santiago et al. (1983) have demonstrated that egg yield in <u>Oreochromis niloticus</u> is strongly correlated with the quality of diet given to broodstock during the breeding period but the effect of the feeding regime given before maturity, despite its economic significance, has received less attention.

The concept of 'stress spawning' by tilapia has been reported by several authors (Iles, 1973; Lowe McConnell, 1982) and explained as part of the 'stunting' tactic that allows the best chance for survival in deteriorating conditions such as a drying floodplain environment. Miranova (1978) reported the more frequent spawning and greater numbers of eggs produced by underfed O. mossambicus compared to fish fed abundantly. Tuan (1986) found that Nile tilapia raised on low to medium level protein diets (20-35% crude protein) in a clear water system subsequently produced larger numbers of smaller eggs than fish fed high protein (42.5-50% crude protein). The better nourished fish also began to spawn earlier but subsequently less frequently than fish on poorer diets.

In commercial systems, however, broodstock are usually raised to maturity in earthen ponds in which natural feed may be an important part of the diet.

This section describes experiments to investigate the effect of pre-maturation production of broodstock under supplementary feeding regimes of different quality (and cost) on subsequent spawning frequency in female broodfish.

3.1.2. Methods

(a) Origin of fish and pond preparation

Fingerlings (weight range 1.5-3.0 g) were produced by spawning broodstock in a series of three small 100 m^2 ponds in July 1986. Nine identical 200 m^2 earthen ponds, which had previously been drained and dried, were filled with water from the AIT main canal.

(b) Stocking and management

Fingerlings (1.5-3.0 g each) were stocked at a density of 5 fish/m2. Three different feed regimes were compared: (a) a low cost supplementary feed (fine rice bran); (b) a low protein (16% crude protein) floating pellet; and (c) a high protein (30% crude protein) floating pellet. All three treatments were tested in triplicate and the replicates were arranged randomly within the experimental pond system. A feeding level of 4% BW/day (assuming no mortality) was used, adjusted monthly after sampling 10% of the fish biomass using a seine net. All ponds also received septage, initially at a rate equivalent to 150 kg COD/ha/day rising after one month to 300 kg COD/ha/day applied twice weekly at three times the daily rate. Water was added as required to maintain a depth of 1 m. After 5 months the ponds were drained, the fish harvested and separated into males, females and indeterminate sex fish; each. group was weighed to determine yield. Some of these fish were used subsequently in a tank spawning experiment (T3). Samples

of fish were tagged and individually weighed (to the nearest g) and measured (to the nearest 0.1 cm) allowing estimation of condition (Fulton's Condition factor $K = 100 \text{ W/l}^3$; W= total weight; l=total length). The spawning frequency of tagged fish in tank units, in which the fish were stocked at different densities, was subsequently monitored. Equal numbers of fish from each production strategy were stocked at four different densities (60, 80, 100 and 120 fish per tank), over a period of 106 days and fed a high quality catfish pellet (crude protein = 30%, D3) to appetite. The total numbers of spawns from the tagged sample fish (30% of fish in each spawning tank) were noted and used as the basis for estimating egg and fry production from females reared on each of the three test diets.

(c) Biometrics

A Chi-square test was used to test for independence (Gomez and Gomez, 1976) of previous diet and subsequent spawning density on the frequency of spawning of tagged female fish. The observed ratio was tested for homogeneity of expected spawning frequency of fish raised on the three different diets (1:1:1).

Table 3.1 Mean size of female fish raised in fertilised earthen ponds¹ using three different supplementary feeds prior to their use as broadfish in concrete tanks at different densities (T3)

Supplementary food	Female Broodfish Means (± S.E.)					
Supplementary feed	Total length (cm)	Body weight (g)	Condition factor			
Fine ricebran (D1) Floating pellet (D2)	14.28 (0.19)ª	54.2 (2.0)ª	1.84 (0.03)			
(16% CP ²) Floating pellet (D3)	14.06 (0.20)ª	52.4 (2.2)a	1.84 (0.02)			
(30% CP ²)	14.87 (0.18)b	60.4 (2.2)b	1.80 (0.03)			
F	4.74**	3.94*	0.59n.s.			

Means in the same column with the same superscript are not significantly different (P > 0.05)

¹ Earthen ponds (Area = 200 m^2) stocked at 5 fish/ m^2 , t = 117 days

² crude protein (%DM)

F value one-way Anova

n.s. P > 0.05

^{*} P < 0.05

^{**} P < 0.01

Table 3.2 Mean total net yield and survival of fish raised on three different supplementary feeds in earthen ponds¹ prior to their use as broodfish in concrete tanks at different densities (T3)

Diet	Mean ± S.D.					
Diet provided	Survival %	Total net yield (kg)				
Fine ricebran (D1)	64.1 (4.8)	32.2 (1.5)				
Floating pellet (D2)	64.3 (13.6)	34.2 (5.4)				
Floating pellet(D3) (30% CP ²)	68.7 (20.4)	39.0 (8.6)				

¹ Earthen ponds (area = 200 m²) stocked at 5 fish/m², t = 117 days

² crude protein (%D.M.)

Table 3.3 Frequencies of observed and expected harvested seed clutches1 from females fed different supplementary diets prior to their use as broodfish in concrete tanks at different densities (T3)

STOCKING	SUPPLEMENTARY FEED							
DENSITY	D. CERDAN	PELLETED FEED						
(females/tank)	RICEBRAN	16%C.P. ²	30%C.P. ²	TOTAL				
30ª obs.	2.5	4.3	6.0	12.8				
exp.	3.6	4.4	4.9					
chi2	0.336	2.27 x 10-3	0.25					
40 ^b obs.	5.0	5.4	4.8	15.2				
exp.	4.2	5.2	5.8					
chi2	0.15	7.7 x 10-3	0.17					
50 ^b obs.	4.4	4.3	3.7	12.4				
exp.	3.4	4.2	4.7					
chi2	0.29	2.3 x 10 ⁻³	0.21					
60° obs.	2.0	3.1	4.5	9.6				
exp.	2.7	3.3	3.6					
chi2	0.18	0.012	0.22					
TOTAL	13.9	17.1	19.0	50.0				

Chi-square value

- independence of density and feed = 1.85n·s·

a - 30 and 60 only (combined) = 30.14***

b - 40 and 50 only (combined) $= 1.13^{n \cdot s}$.

n.s. P > 0.05 *** P < 0.001

¹ number of seed clutches as proportion of tagged fish

² crude protein (%DM)

3.1.3 Results

The growth of fish raised on the floating pelleted feed of comparatively higher quality and cost (D3) was greater than the other two treatments (D3=0.5 g/day, compared to D2=0.43 g/day and D1= 0.45 g/day, Table 3.1) and their final size, before use as broodfish, was significantly larger (mean body weight P<0.05, mean total length P<0.01). Fish condition was not significantly different (P>0.05) however (Table 3.1). Total net yield and survival from the three treatments were not significantly different, largely because of the more variable survival in the higher quality feed strategies (Table 3.2). The subsequent spawning frequencies of broodfish maintained at different densities in concrete tanks (3.3.2) as a proportion of tagged fish are given in Table 3.3. A test for homogeneity of the expected ratio of spawning frequency of fish raised on the three different diets (1:1:1) of the four densities taken together revealed that they were heterogeneous and that the data could therefore not be pooled.

There was a measurable effect of early diet quality on subsequent spawning frequency at high (T34) and low (T31) densities but not at medium stocking densities (T32,T33).

3.1.4 Discussion

Early nutrition had no measurable effect on the subsequent spawning frequency of female broodfish at the optimal stocking densities in the spawning tank. But if the stocking density was either higher or lower than the optimal range, the fish raised on a high quality diet spawned significantly more frequently than those raised initially on a lower quality diet (P<0.001). Initial stocking densities in concrete tanks of between 80-100 fish per tank (sex ratio 1:1, female biomass 0.21-0.87 kg/m²) gave significantly higher output of seed per harvest than suboptimal stocking densities treated statistically as one group (either lower, 60 fish per tank or higher 120 fish per tank). This different response to early feeding on broodstock subsequently spawned as mixed groups at different densities might again be interpreted as a result of differential hierarchy. The larger initial size of broodfish fed a higher quality diet may have allowed these fish to dominate and spawn more frequently as a result. The stability of a hierarchy in such systems may be very sensitive to slight changes in density. The hierarchical effect of a few dominant fish within a clear water arena may be proportionally greater at a lower stocking density than when numbers are fractionally higher and the hierarchy less stable.

However, the effect was also clearly shown at the highest density in which the hierarchy seems to have been destabilised. Poorer productivity, lower synchrony and reproductive frequency (% of fish spawning) suggested that successful spawning

activity was reduced compared to lower densities. Possibly, the larger fish were more capable of competing for food at this density and of spawning successfully and evading disturbance by subordinates.

Lee (1979) described the behaviour of Oreochromis breeding at high density in aquaria and observed that the 'strongest female', or dominant female was always the first to accept the courtship of the male. During the spawning period, both the male and the female defended the eggs from other females attempting to steal them. Thus, in the present experiment, although a few individuals could not dominate large parts of the tank as seem possible at low densities, dominant, larger females might still outcompete weaker individuals for the best spawning sites and be more capable of successfully incubating Rana (1986) reported the eggs once spawned. higher fertilisation rates and survival of fry in larger, older female brooders than in smaller, younger fish. He suggested that the differences could be explained by behaviourial differences broodfish of different age-classes. between Similarly, broodfish size, independent of age and in this case relating to early rearing conditions, could explain the differences observed in the present experiment.

3.2 Effect of broodstock density on tilapia seed production

3.2.1 Background

The optimum stocking rate (number or biomass) of broodfish per unit area for seed production is very system specific. Natural tilapia communities congregate for spawning at higher than normal density at the lek site. Fishelson (1983) observed colonies of <u>Oreochromis aureus</u> males to frequently number over ten individuals in an area of around 30 m² and females to congregate at densities of more than 1 individual/m². The spawning act is followed by mouthbrooding during which the female becomes solitary and leaves the main group. In spawning arenas, Fishelson (1983) noted that incubating females often dramatically increased their position within the social hierarchy.

These observations suggest that optimal densities for spawning success might be different to those required for maximal fry production.

The design of the 'Baobab arena' was an attempt to allow some degree of self-selection by fish within an intensive system following observation of the fishes' natural spawning behaviour (Haller and Parker, 1981). This concrete unit consists of a series of concentric rings in which the central male nesting and spawning area is separated from a middle ring by graded gates into which the smaller females can pass post-spawning, and an outer, shallower ring into which only released fry can pass. The design allows mouthbrooding females to escape from

male pressure and young fry to escape predation.

Theoretically, the stocking density in spawning units can be increased until courtship behaviour is inhibited and/or spawning or incubation is less successful because of density dependent factors. Several workers have used the area per male as the major criteria for determining stocking density as males require a minimum size of territory to successfully make and defend a nest (Uchida and King, 1962; Fishelson, 1966; Balarin and Haller, 1982). In practice, some types of unit, especially larger ones are operated with much lower densities of broodstock (Table 1.2).

A large range of broodfish stocking rates in tanks has been reported. Snow et al. (1983) used a stocking density of $0.24 \, \text{kg/m}^2$ in static water, plastic tanks, a factor of ten lower than that used in tanks with a recirculated water supply in Stirling, U.K. ($2.52 \, \text{kg/m}^2$; Balarin and Haller, 1982). However, the upper range of densities used for fry production is still far below those used for grow-out (>100 $\, \text{kg/m}^2$), indicating that the density dependent nature of natural spawning in Oreochromis inhibits fry production below the limits of water quality in intensive systems. Over the range of tank systems in operation, however, deterioration in water quality maybe a major factor reducing fry production. Complete water exchange at fry harvest may be important in maintaining water quality in broodstock systems.

Bautista et al. (1988) maintained actively spawning <u>Oreochromis</u> niloticus in small static tanks (area = $4m^{\frac{1}{2}}$) using aeration and

a complete change of water at harvest (every 21 days); higher stocking densities produced less fry over the range investigated $(0.42-1.04 \text{ kg/m}^2)$. Ridha et al. (1985) kept two size classes of broodstock (<u>Oreochromis spilurus</u>) in similar tanks for a 6 month period with aeration and a complete change of water at each weekly harvest.

Larger tank systems (e.g. area = 100 m², Guerrero and Guerrero, 1985) have been stocked at lower densities for extended periods of time with no water exchange, but this may have contributed to poorer fry production over the latter half of the period. Substantially increased stocking densities, with a concomitant increase in fry production, have been achieved in tank systems which maintain high water quality by continual replacement of water using a flow through or recirculated supply.

Simple air-lifts to drive biological filtration within the breeding system are frequently used for aquaria and small tanks. Larger systems tend to use filtration external to the spawning unit for greater flexibility and efficiency (Uchida and King, 1962; Balarin and Haller, 1982).

Water quality, particularly with respect to dissolved oxygen, is more difficult to manage in outdoor tanks or in hapas containing densely stocked broodfish receiving high quality diets (Uchida and King, 1962; Sampson, 1983; Bautista et al., 1988). Guerrero and Guerrero (1985) associated the heavy accumulation of bottom scum and thick growth of algae with deterioration of water quality in large static outdoor tilapia breeding tanks after prolonged periods.

The best production from small, intensively managed pond systems in the Philippines occurs when ponds receive flow through underground water (Guerrero, 1985) and a high density of small broodfish is used (4 fish/ m^2 , 50-100 g each).

A high stocking density of broodfish is used in large earthen ponds in Israel (Table 1.2) for batch production of swim-up fry. Water quality presumably remains high as the system is completely drained after every harvest (17-19 days) and is refilled with high quality well water (Rothbard et al., 1983). Bautista et al.(1988) found that fry yields were significantly lower in nylon hapas in Laguna de Bay than in static water concrete tanks over a similar range of densities. They suggested that production was affected by temperature and water quality. The level of water quality and exchange inside the hapas was not reported but the quality of the Lake itself was reported to be subject to periodic algal blooms and their dieoff and subsequently depleted levels of dissolved oxygen. In contrast, water was completely exchanged in the tanks at harvest.

Guerrero and Garcia (1983) tested a double net hapa design in which the broodfish were held in a coarse mesh net within a fine mesh hapa (30mm mesh). The design attempted to decrease the suspected high level of early cannibalism of released fry by the densely stocked broodfish. After spawning and mouthbrooding, the released fry could escape cannibalism by moving outside the broodstock holding/spawning net. At the same stocking density of broodstock, this design doubled the fry

production per female, although the actual output of fry from the system was the same as a normal single net hapa. Beveridge (1984) suggested that such a net design might further impede exchange of water into the nets and described a simpler design with a single large mesh wall.

The present trial attempted to study seed production over time in simple tank and hapa units stocked and maintained at different densities of broodfish. High seed production was maintained in earlier trials (T1, T2, H1, H2) by intensive management and the same approach was used in this trial. Water quality and broodstock growth were monitored throughout the experimental period with the objective of estimating the optimal carrying capacity of such spawning systems under commercial conditions.

3.2.2. Methods

(a) Origin and pre-treatment of broodfish

Broodstock were obtained from an experiment on broodfish production (3.1) in which fry from the same background were raised to maturity under different regimes of supplementary feeding in earthen ponds. Equal numbers of fish from the different treatments in this experiment were used. Samples of fish from each of the treatments were tagged so that their subsequent seed production in tanks could be monitored.

(b) Stocking and harvesting

Broodstock were separated after harvest into spawned and unspawned females and males before enumeration and bulk weighing.

The stage of eggs and fry was assessed at harvest, the number of clutches of each stage was counted and then pooled into one sample. All eggs and fry were preserved in 4% formaldehyde except those used for incubation studies which were counted immediately.

All fish used in the tank experiment (T3) were maintained for the duration of the trial within concrete tanks receiving recirculated water inside the hatchery. Fish in the hapa experiment were maintained in hapas suspended within a single large earthen pond (1740 m²). In both cases, females were divided into three groups of broodfish with alternated periods (10 days) of conditioning in small units and spawning in larger units (5 days). Males remained in the spawning unit throughout

Table 3.4 Experimental design used to study effect of broodstock density on seed production in circular tanks (Area = 12.57 m^2)

System/ Experi- ment/	Number of	Number of fish		Restocked to spawning		Exchanged from conditioning		Interval (days)	
Treat-	per spawning	total	Туре	Tank	Нара	Tank	Нара	Spawning	Condi- tioning
T31	30	30	males spawned females	х		x		5	10
	30	90	unspawned females			x			
Т32	40	40	males spawned	x					
	40	120	females unspawned females			x		5	10
Т33	50	50	males spawned	х					
	50	150	females unspawned females	 		x		5	10
T34	60	60	males spawned	х					_
	60	180	females unspawned females			x		5	10

x denotes restocking and conditioning strategy

Table 3.5 Experimental design used to study effect of broodstock density on seed production in rectangular hapas (Area = 40 m^2)

System/ Experi- ment/	Number of fish		There is	Restocked to spawning		Exchanged from conditioning		Interval (days)	
Treat-	per spawning	total	Туре	Tank	Hapa	Tank	Нара	Spawning	Condi- tioning
H21	30 30	30 90	males spawned females unspawned females		x		x x	5	10
H22	40 40	40	males spawned females unspawned females		x		x x	5	10
н23	50 50	50 150	males spawned females unspawned females		x		x x	5	10
H24	60 60	60 180	males spawned females unspawned females		х		x x	5	10

x denotes restocking and conditioning strategy

the experimental trial.

<u>Tanks</u>

All females were stocked in the hatchery for conditioning and spawning (Table 3.4). Female broodfish were maintained in conditioning tanks at a standard density throughout the experiment by manipulation of depth. Flow rate was increased according to the number of fish to maintain a standard residence time for each treatment.

<u>Hapas</u>

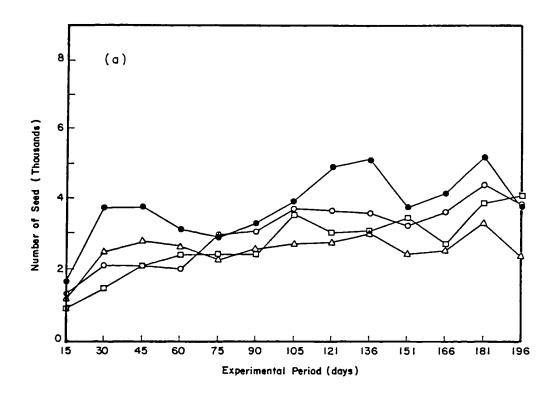
All females were stocked in net hapas for conditioning and spawning (Table 3.5). In all treatments, female broodfish were conditioned in small hapas at the same density by adjusting the hapa depth in the water.

Fig. 3.1

Effect of stocking density on mean daily output of seed from

- (a) tanks (Area = 12.57 m^2 ; Interharvest Interval 5 days; sex ratio = 1:1 and
- (b) hapas (Area = 40 m; Interharvest Interval 5 days; sex ratio = 1:1)

(where open square, 30 fish/unit; open circle, 40 fish/unit; closed circle, 50 fish/unit; open triangle, 60 fish/unit)



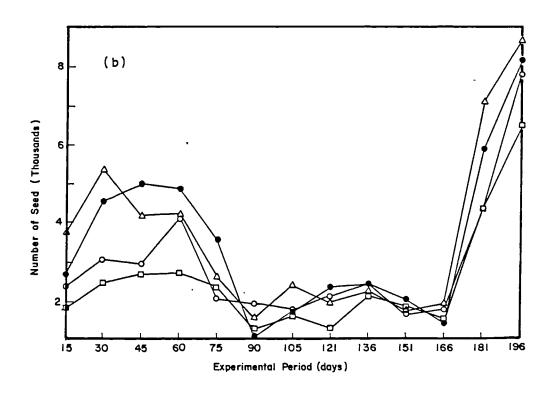


Table 3.6 Productivity of tank and hapa seed production systems over a range of broodstock densities (T3, H2)

The experimental period has been divided into 3 phases; values shown are means (± S.E.). Spawning arena area; tank = 12.57 m², hapa = 40 m²; sex ratio 1:1, t = 201 days

PERIOD		RLY 1-60	MID Day 61		LATE Day 167-201			
	TANK	НАРА	TANK	НАРА	TANK	НАРА		
Total number of seed per harvest	11019 (760)	17531 (1099)	16514 (575)	9824 (581)	19587 (1073)	35960 (3068)		
	F	F 24.4***		.9***	3:	32.4***		
Mean daily output of seed	2248 (170)	3506 (220)	3271 (114)	1941 (117)	3917 (215)	7192 (614)		
	F	20.9***	66	.4***	3:	32.4***		
Number of seed per kg of spawned female	7255 (307)	9018 (458)	5562 (135)	4577 (233)	5216 (186)	6150 (253)		
per harvest	F	F 10.8*		.64*	9.29*			
Number of seed per kg of total female	198 (12.4)	324 (15.8)	175	102 (7.3)	163 (14.5)	301 (38.3)		
per day	F 40.29***		51.6***		14.3**			
Number of seed per kg of total fish	151 (19·2)	242 (20.9)	115 (5.1)	78 (5.6)	95 (7.4)	176 (14.7)		
per day	F 1	F 10.1*		23.8***		28.8***		
Number of seed per m ² per day	175 (12.1)	88 (5.7)	260 (9.6)	49 (3.0)	312 (17.1)	180 (15.3)		
	F 4	0.1***	412.	1***	30.1***			

¹ mean of 4 densities, 30, 40, 50 or 60 females per unit.

F value, one-way Anova
* P < 0.01
** P < 0.001
*** P < 0.0001

Table 3.7 Productivity of tank and hapa seed production systems over a range of broodstock densities (T3, H2).

	MEAN VALUES (S.E.)										
	Total number of seed per harvest!	Number of seed per kg female spawned	Number of seed per kg female total ²	Number of seed per kg fish total							
H21 H22 H23 H24 T31 T32 T33 T34	12934 (1607)ab 15044 (1667)abc 17626 (1998)bc 18311 (2121)bc 14034 (990)ab 15468 (999)abc 19040 (946)c 12983 (649)ab 2.68***	6886 (549) 5230 (428) 6340 (525) 5740 (391) 6105 (210) ^b 5980 (239) ^{ab} 6701 (336) ^b 5253 (252) ^a 5.076***	210.8 (26.2)a 188.3 (21.9)a 193.5 (24.3)a 178.7 (22.8)a 203.2 (12.3)a 190.6 (10.5)a 203.8 (10.6)a 119.5 (7.8)b 3.34*	131.9 (15.2)b 120.7 (15.4)b 113.6 (16.2)b 105.3 (14.7)ab 139.4 (8.9)b 121.1 (8.1)b 124.0 (8.7)b 74.1 (7.8)a 3.9****							

Means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

(Spawning area; tank = 12.57 m^2 , hapa = 40 m^2 ; sex ratio 1:1, t = 201 days)

¹ Total number of seed per 5 days interval

² Number of seed per kg of female total including exchange females

F value one-way Anova of log10 transformed data

^{*} P < 0.05 ** P < 0.01

^{***} P < 0.001

3.2.3. Results

The production of tilapia seed over an extended period (201 days) using a range of broodstock densities was found to be far more variable in hapas than in concrete tank systems. This was correlated with a prolonged period of poor water quality in the hapa environment especially with respect to early morning low dissolved oxygen mid-way through the experiment. Such variability in hapas contrasted with a general increase in daily egg output with time (mean of all treatments; day 1-60: 2248 seed/tank/day, day 167-201: 3917 seed/tank/day) for all but the highest density tank treatment (T34). Seed production was therefore considered over three phases (Table 3.6); early, mid and late in addition to the experimental period taken as a whole.

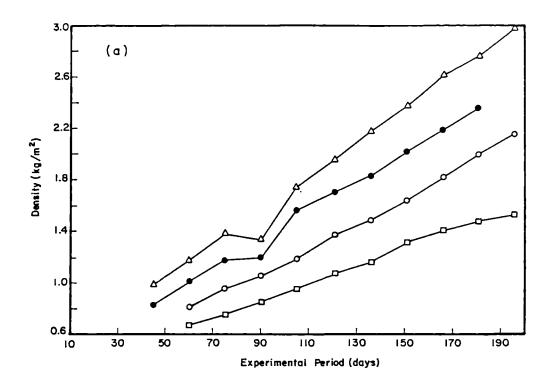
Density effect on broodstock productivity

Egg production per harvest period (5 days) over the whole experiment was not significantly different except for low-broodstock density in hapas·(H21) and low and high densities in tanks (T31 and T34) which gave significantly poorer yields (Table 3.7).

These results suggest that the optimal stocking density for seed production was exceeded in tanks but not in hapas. The daily fry production of the highest stocking density treatment (60 females/tank; T34) was lower than that of the treatment consistently producing most seed (T33) (Fig. 3.1). The highest yielding treatment continued to outperform the others even

Fig. 3.2

Density of all broadfish (g/m^2) in (a) tanks (Area = 12.57 m⁻) and (b) hapas (Area = 40 m²) maintained at four different densities (by number, sex ratio = 1:1) (where open square, 30 fish/unit; open circle, 40 fish/unit; closed circle, 50 fish/unit; open triangle, 60 fish/unit)



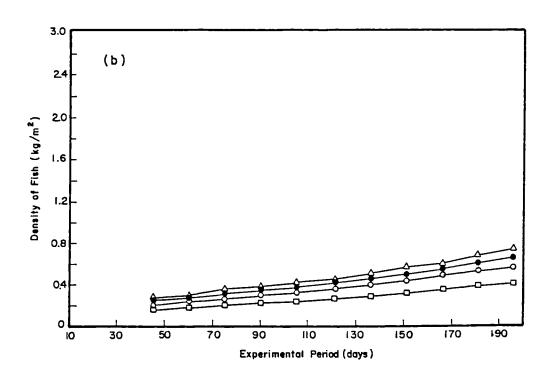


Table 3.8 Variation in clutch size of seed harvested from broodfish in tanks and hapas
(a) seed harvested of different stage (mean of all density treatments) and (b) seed harvested from broodstock spawned at different densities (T3, 112)

(a)

Seed Stage	Mean seed cl.	utch size (S.E.)
	Hapa	T'ank
1	1171 (50.4)	1118 (51.8) *
2 2-3	957 (39.2) 897 (32.9)	927 (29.3) y 835 (23.8) y
3	743 (33.2)	712 (33.8) z

(b)

The second second	IIA	PA .	TANK		
Treatment	Maximum broodfish density (kg/m²)	Mean seed clutch size (S.E.)	Maximum broodfish density (kg/m²)	Mean seed clutch size (S.E.)	
30	0.42	1039 (55.1)	1.5	1055 (48.4)ª	
40	0.57	919 (36.2) ^{nb}	2.1	918 (35.2)	
50	0.66	929 (39.4) ^{nb}	2.4	909 (33.7)	
60	0.76	868 (38.2) ^b	3.0	725 (27.0)°	
F		· 2.9*		13.91***	

Means in the same column with the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range Test)

F value one-way Anova

Spawning area; $tank = 12.57 \text{ m}^2$, $hapa = 40 \text{ m}^2$; sex ratio 1:1; t = 201 days

^{*} P < 0.05

^{***} P < 0.001

Table 3.9 Synchrony of spawning in (a) tanks and hapas² stocked with broadfish at different densities and (b) in hapas over a range of stocking densities for different periods within the experimental period

(a)

The state of the s		Mean stage of harvested seed							
Treatment	n	1	2	2-3	3				
T31 T32 T33 T34 Chi-square ¹	40 40 40 39	18.5 17.8 22.2 28.5 11.7**	18.5 26.2 24.1 21.6 6.0 ⁿ ·s	35.6 35.9 34.8 30.3 2.6 ^{n.s}	25.5 18.4 17.2 15.8 5.1 ^{n.s}				
H21 37 H22 36 H23 40 H24 37 Chi-square ¹ Z		28.3 a 23.9 24.1 24.3 a 0.8 ^{n.s} 0.47 ^{ns}	22.2 a 20.4 20.0 21.1 a 2.0 ^{n.s} -0.03 ^{ns}	18.4 a 28.3 25.2 25.2 a 4.9 ^{n.s} -1.87 ^{ns}	20.1 a 27.4 30.6 29.4 b 4.2 ^{n.s} -2.2*				

(b)

Time Period	Percentage Stage 3 Seed
within experiment	Mean + S.D. (n)
Early (day 1- 60) Middle (day 61-166) Late (day 167-201) Chi-Square = 16.34**	36.09 ± 25.3 (38) 20.05 ± 19.3 (85) 36.15 ± 25.9 (24)

n.s. P > 0.05

^{*} P < 0.05

^{**} P < 0.01

Z value of Mann-Whitney test between means in the same group marked with different superscripts are significantly different (P < 0.05)

¹ Chi-square value of the Kruskal-Wallis test

² Spawning area; tank = 12.57m², hapa = 40m²; sex ratio 1:1, t = 201 days

Table 3.10 Mean frequency of females spawning in tanks and hapasa stocked at different densities as a percentage of fish stocked per spawning period (5 days)

Treatment	No. of broodfish/unit	% Female spawned/harvest Mean ± S.D. (n)					
T31 T32 T33 T34 Chi-square	60 80 100 120 9 = 36.2***	$\begin{array}{c} 49.3 \pm 13.2 & (40) \\ 47.3 \pm 11.5 & (40) \\ 45.7 \pm 11.1 & (40) \\ 33.5 \pm 10.0 & (40) \end{array}$					
H21 H22 H23 H24 Chi-squareb	60 80 100 120 = 1.2 ^{n.s.}	39.0 ± 18.6 (40) 42.8 ± 17.1 (40) 38.9 ± 18.3 (40) 38.6 ± 16.9 (40)					
All tanks (g All hapas (g Z° - 2.13*		43.9 ± 12.9 (160) 39.7 ± 17.8 (160)					

n.s. P > 0.05

^{*} P < 0.05

^{***} P < 0.001

a Spawning area; tank = 12.57m², hapa = 40m²; sex ratio 1:1, t = 201 days

b Chi-square value of Kruskal-Wallis test

c Z value of the Mann-Whitney-U test

after exceeding a biomass equivalent to that which inhibited spawning in the high density treatment. At 110 days, for example, the density of T33 was over $0.5~{\rm kg/m^2}$ and fry production remained high; the density of T34 was below this level for the first 70 days but seed production remained lower at all times.

Considering the experimental period as three parts (early, middle and late), hapa production exceeded mean tank production across a range of densities in the early and late periods, while tank production exceeded that of hapas over the middle period (Table 3.6).

Seed production per unit area in tanks exceeded that in hapas throughout the experiment (mean number of seed/ m^2 /day: tanks 175-312, hapas 88-180).

Relative productivity (calculated as seed produced per kg of spawned female) showed the lowest variation of the parameters monitored, with mean hapa and tank productivity differing by less than 25%. In tanks, the mean relative productivity declined throughout the experiment whereas actual seed production increased.

Density effect on clutch size

Clutches of stage 1 eggs tended to be larger than those of other seed stages, irrespective of broodstock density in both tank and hapa treatments (Table 3.8). The average reduction in clutch size with time during incubation indicated that egg losses during mouthbrooding were approximately the same in hapas and tanks at around 36% from uneyed eggs to yolk-sac

Fig. 3.3

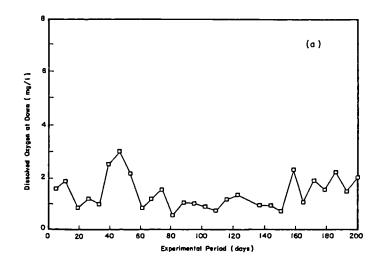
(a) dissolved oxygen level at dawn,

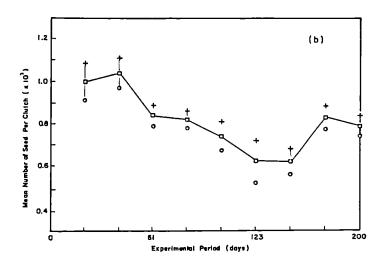
(b) mean harvest clutch size of seed and

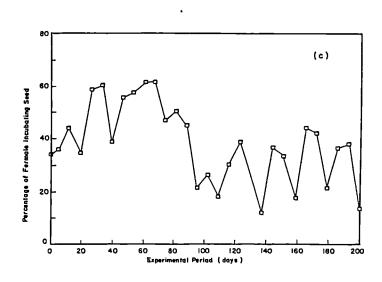
(c) frequency of female broodfish harvested with seed in the mouth

from female broodfish maintained at variable density (density of all broodfish $< 800 \mathrm{g/m}$; sex ratio 1:1) in nylon hapas (Area = 40 m²) suspended in an earthen pond

^{*} mean of all treatments







larvae.

Clutch size of seed harvested from brooding females was inversely related to stocking density in tanks and hapas (Table 3.8). The relationship was similar although more pronounced in tanks, despite actual densities being considerably different (final stocking density; tanks $1527-2984 \text{ g/m}^2$; hapas $416-756 \text{ g/m}^2$).

The mean clutch size declined during the period of poor water quality and increased after early morning dissolved oxygen levels were maintained above 1 mg/l (Fig. 3.3).

Synchrony of spawning

In tanks synchrony of spawning declined significantly with density. The percentage of hatched fry at harvest was greatest (25%) in the lowest density whereas the number of stage 1 eggs was greatest (28.5%) in the high density treatment (Table 3.9a).

There were no differences in synchrony of spawning in hapas over the broodstock range 1.5-3.0 fish/m² (Table 3.9a).

The proportion of seed harvested as hatched fry in all the hapa treatments when combined as a single group, was significantly greater (36%) in the early and late periods of the experiment compared to the mid-period (20%) (Table 3.9b).

Frequency of spawning and ISI

The number of fish spawning as a percentage of those stocked showed considerably more variation in hapas compared to tanks (Table 3.10). The percentage of females spawning in hapas

Table 3.11 Mean Interspawning Interval (ISI) of female broodfish stocked at different densities in tanks and hapas 1 (T3, H2)

Treatment	Mean + S.E.	N
T31 T32 T33 T34 F = 4.66*	31.3 ± 3.5 a 32.2 ± 7.4 a 33.2 ± 3.6 a 46.1 ± 0.6 b	3 3 3 3
H21 H22 H23 H24 F = 0.03n.s.	38.9 ± 2.7 c 41.1 ± 1.5 c 39.6 ± 1.3 c 39.1 ± 1.7 c	3 3 3 3
All tanks (grouped) All hapas (grouped) F = 1.48n.s.	35.7 ± 2.3 39.7 ± 2.4	12 12

F value one-way Anova. Means in the same group having the same superscript are not significant different (P > 0.05, Duncan's Multiple Range Test)

n.s. P > 0.05

^{*} P < 0.05

¹ Spawning area; tank = 12.57 m²; hapa = 40 m²; sex ratio 1:1; experimental period 201 days

Body characteristics of frmule broodfish stocked at different densities in tanks and hapas at final harvest. Table 3.12

					_					
	GSI 1	+	+	+	۱+	1 +	1+	۱+	3.3 ± 0.35 a	0,38n.s.
S.E.	Gonad weight (g)	8.1 + 1.2 в	6.6 + 0.8 a	7.2 + 0.8 a	8.7 ± 1.1 a	8.9 + 1.5 a	8.7 ± 1.0 a	8.4 ± 0.9	8.8 ± 1.0 a	0.62n.s.
Mean ± S.E.	Body weight (g)	± 23.1 a	+!	± 11.3	+ 11.2	+		265 ± 14.9 ac		2,71**
	Total length (cm)	6 ± 0.75	4 ± 0.57	1 ± 0.38	+1	3 ± 0.52	+I	24.3 ± 0.48 ac	۱+ ع	2.59
		T31	T32	T33	T34	1121	H22	1123	1124	្ន

Means in the same column having the same superscript are not significantly different (P > 0.05, Duncan's Multiple Range 'Fest.)

F value of one-way Anova n.s. P > 0.05 ** P < 0.01

 1 GSI = Goradosomatic index $^{'}$ Spawning area; tank = 12.57 m², haya = 40 m²; sex ratio 1:1, t = 201 days (T3, H2)

Table 3.13 Mean feeding rate, actual feed input and biomass of broodfish stocked at different densities in tank and hapa system³

Treatment	Total number of broodfish	Total feed ³ input (kg)	Mean total biomass² (kg)	Feeding rate ¹ %BW/day
T31 T32 T33 T34 H21 H22 H23 H24	120 160 200 240 120 160 200 240	63.2 74.3 88.3 102.6 56.7 62.9 68.1 69.6	17.5 22.6 27.7 32.6 19.8 27.4 31.4 37.3	1.8 1.7 1.6 1.6 1.5 1.2 1.1

¹ calculated over 201 day period

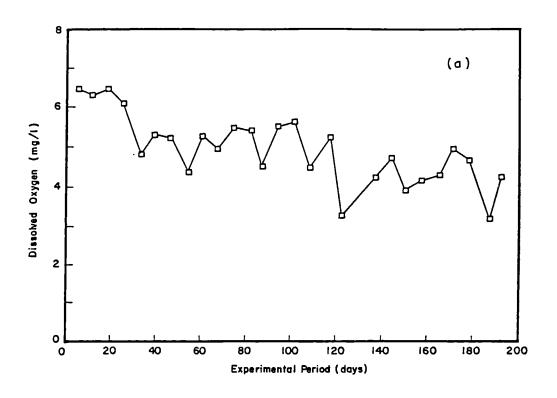
Spawning area; tank = 12.57 m^2 , hapa = 40 m^2 ; sex ratio 1:1, t = 201 days (T3, H2)

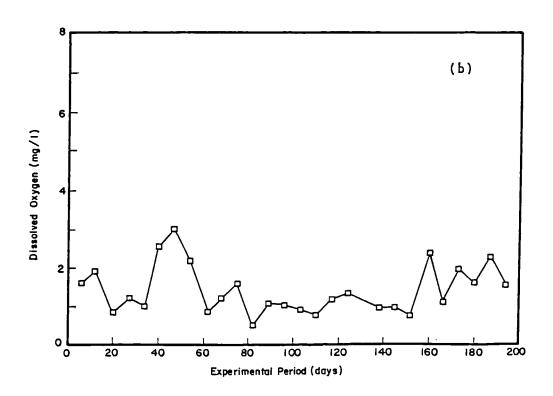
^{2 (}initial weight + final weight)/2

³ total feed into spawning and conditioning units

Fig. 3.4

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Mean dissolved oxygen in (a) spawning tanks (Area = 12.57 m²) and (b) spawning hapas (Area = 40 m²) stocked at variable densities (tanks < 3 \text{ kg/m²}; hapas < 0.8 \text{ kg/m²}) measured weekly at dawn and at 09.00 a.m. respectively (mean of all treatments)
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varied with the level of early morning dissolved oxygen (Fig. 3.3, c). There were no differences in spawning frequency with density in hapas. In tanks, where densities were much greater, frequency of spawning was significantly lower at the highest density (H34).

The mean ISI of broodfish groups (three groups per treatment) spawned in hapas was generally longer than in tanks, with the exception of the highest density treatment (T34) (Table 3.11).

Growth

The body size of broodfish at harvest showed variation between treatments. The average total length and body weight of female broodfish in hapa treatments were greater than fish spawned and conditioned in tanks, although gonad weight, GSI and condition factor were not significantly different (Table 3.12). Fish in hapas had higher mean growth rates than fish in tanks despite their consumption of pelleted feed being lower in both absolute and relative body weight terms (Table 3.13).

The density and biomass of stocked fish showed a steady increase in both tank and hapa treatments (Fig.3.2, Appendix 2). Growth was inversely correlated to egg production, with the latter increasing with early morning dissolved oxygen levels.

Water Quality

<u>Tanks</u>

Dissolved oxygen in the spawning tanks showed a gradual decline over the experimental period (6.5-3.0 mg/l; Fig. 3.4). In the higher density treatments (T33, T34), oxygen fell to its lowest

level towards the end of the experimental period, but was never measured at less than 2.0 mg/l (Appendix 2) in spawning tanks and 1.8 mg/l in conditioning tanks.

Total ammonia increased steadily to a maximum of 0.6 mg/l in the highest density spawning tanks, and 0.9 mg/l in conditioning tanks (Appendix 2). pH ranged between pH 6.0-8.6 and thus ammonia in the unionised form was unlikely to be at toxic concentrations to fish (Appendix 2).

The decline in dissolved oxygen and increase in total ammonia was consistent with the increase in biomass of fish in the system and the correspondingly higher application of feed and levels of waste feed and excretory products. Phytoplankton concentration as indicated by chlorophyll a levels remained low in the tanks throughout the experimental period (Appendix 2). Temperatures remained fairly constant throughout the trial although there was a temporary rise on refilling the tanks with warm tap water after each harvest.

<u>Hapas</u>

Early morning dissolved oxygen concentrations fluctuated throughout the experimental period in the hapa environment (Fig.3.4). Initially early morning levels of dissolved oxygen were generally above 1 mg/l in the hapas; but from days 60 to approximately 150 the level often fell below 1 mg/l. Measured water quality parameters inside spawning and conditioning hapas were usually slightly different but consistent with those outside the cages. Levels of dissolved oxygen, for example, were typically slightly lower in conditioning cages and outside

the nets than inside the spawning hapas (Fig. 3.5). This was probably related to the higher densities of broodfish within conditioning nets and the presence of escaped broodfish and their progeny outside the nets.

Chlorophyll a levels followed the same trend over the experimental period as dissolved oxygen (Appendix 2), with two peaks at the beginning and end of the experiment (250-350 mg/m 3) and much lower levels in the middle period (40-200 mg/m 3). Total ammonia reached high levels initially (1.8 mg/l) but declined erratically over the duration of the trial such that levels were under 1 mg/l for the latter half (Appendix 2). Temperatures measured at 20 cm below the water surface were relatively constant over the duration of the experiment, varying from daily minima of 26-32 $^{\circ}$ C to daily maxima of 29.5-34.5 $^{\circ}$ C. Maximum temperatures were more variable in the latter third of the trial reflecting heavy rainfall during the main monsoon period (August-September) (Appendix 2).

3.2.4 Discussion

(a) Variability in seed production over time

The extreme variability of fry production has been a major limitation to the development of practical hatchery systems for Oreochromis spp.; observed at the level of both individual fish (Lee, 1979; Mires, 1982) and small and large spawning units, there is still confusion as to its cause. Environmental factors may be of most significance in the seasonality of breeding in some natural populations but in culture systems the major controlling factors are probably density-dependent.

In tilapia breeding groups, the common phenomenon of an early peak followed by gradual decline in fry production with time (Broussard et al., 1983; Guerrero and Guerrero, 1985) may be influenced by the increasing density of broodfish. There is evidence that in spawning ponds that become overcrowded with broodfish and recruitment, spawning is inhibited (Mires, 1982). However, there is a tendency in experimental and commercial systems alike to compensate for the low individual fecundity of Oreochromis with a higher stocking density of broodfish.

Changes in temperature and rainfall have been most associated with onset or cessation of peak spawning activity in natural systems but water quality may be of equal significance, accounting for variation in seed production, in high density systems. The striking correlation between daily output of fry in hapas and the early morning dissolved oxygen level indicated this to be the major reason for variation in seed output in the hapas during the present study.

Hapa seed production was far in excess of that obtained in tanks (6000-8000 seed/spawning unit/day) when dissolved oxygen was not limiting, but was low during the prolonged period of poor water quality (1000-2000 seed/unit/day). In contrast, seed production in tanks, in which water quality was more constant, was less variable. Output gradually increased over the duration of the experiment but was maintained between 2000-4000 seed/unit/day over the range of densities operating. The importance of water quality control for improving the homogeneity and predictability of output extended over production periods is clear. Bautista et al. (1988) explained seasonal variation in fry output from static water tanks and hapa nets largely in terms of temperature, and in the case of hapas, in terms of water quality. The hapas were suspended in Laguna de Bay, a water body prone to seasonal algal blooms, die-offs and depleted dissolved oxygen (Nielsen quoted in Bautista et al., 1988).

The decline in early morning dissolved oxygen and chlorophyll a levels over the experimental period was probably related to a large increase in biomass of recruits spawned by escaped broodfish outside the nets. Regular harvest of the fish also increased turbidity in the pond. Suspended fine clay particles would have further inhibited light penetration and therefore photosynthetic oxygen production. Semi-diurnal studies of dissolved oxygen levels were not made but super - saturated levels were common by mid-day in the same ponds fertilised using the same regime in previous experiments

(Edwards et al., 1987).

(b) Other effects of changes in water quality

Low dissolved oxygen in hapas appeared to affect seed production, both by reducing the number of females that spawned and by reducing the mean clutch size of seed that was harvested.

Lowe McConnell (1959) suggested that the mouthbrooding habit made Oreochromis well adapted to spawning in their natural environment of water lily swamps (which are typically deoxygenated), whereas the eggs of the substrate spawning Tilapia spp. would die quickly in such conditions. However, the association between males and females during oviposition and fertilisation under natural conditions is brief and further development of the fertilised seed is carried out away from the site of spawning (Fig. 1.1).

Lower spawning frequency may be a result of poorer fish condition and/or a reduced period of spawning opportunity or vigour during the hours of low dissolved oxygen concentration. The rapid increase in spawning frequency after a return to improved conditions suggests the latter. Ross and Ross (1983) described reduced oxygen consumption by tilapia, as a respiratory response, to levels of dissolved oxygen below 4 mg/l; spontaneous activity increased at levels of between 2.5 mg/l and 1.0 mg/l which the authors associated with an avoidance reaction. Gulping at the surface was only observed in the present trial when oxygen levels declined to below 1.0 mg/l

in the hapas and this is the normal response to these near lethal levels (Chervinski, 1982). Oral incubation of seed would certainly prevent mouthbrooding females gulping and suggested that they respired anaerobically for short periods of time (Kutty, 1972).

The lower mean clutch size recorded in this study indicated that incubation was possible but less successful under such adverse conditions. Uchida and King (1962) correlated low dissolved oxygen with the frequent occurrence of dead yolk-sac fry found in the detritus on the floor of certain spawning tanks. Fry production may be depressed by low levels of dissolved oxygen, especially when the period of production is extended; when feeding is intensive; and/or when there is little or no exchange of water. Thus, the poor productivity of the Baobab outdoor arenas in Kenya may be partly caused by the low dissolved oxygen at dawn (0.3 mg/l; Sampson, 1983) causing loss or mortality of developing eggs and fry. However, gonadal development may have been retarded as the fish were probably under stress until mid-morning and spawning frequency reduced as intense spawning activity was restricted mainly to late afternoon (Sampson, 1983). A decline in fry production from large static tanks over extended breeding periods (72 days) in the Philippines was also related to poor water quality (Guerrero and Guerrero, 1985).

In contrast, relatively stable water quality was maintained in the concrete tanks receiving filtered and recirculated water throughout the present experiment. A gradual decline in dissolved oxygen and increase in total ammonia levels were apparent in both spawning and conditioning tanks. Feed inputs, and thus load on the filter, increased as the experiment progressed, but a build up of settled solids also occurred which probably decreased the efficiency of the horizontal biological filter (Muir, 1982). Dissolved oxygen remained within the tolerance levels of tilapia for growth (Balarin and Hatton, 1979) and levels of unionised ammonia remained below levels considered toxic (Appendix 2); therefore, it is unlikely water quality had any major effect on spawning or mouthbrooding.

The gradual increase in seed output with time in tanks correlated with a steady increase in broodfish biomass and density. The tendency for clutch size to increase with time has been reported for individual fish (Lee, 1979; Mires, 1982; Siraj et al., 1983) but this would be expected as under most conditions broodstock continue to grow and clutch size and weight is correlated with the weight of the female (Lowe McConnell, 1955; Dadzie, 1970; Peters, 1983; Rana, 1986). The faster increase in clutch weight over time and overall larger mean size in the low density treatment is explained by the fact that this treatment contained the fastest growing fish.

Several workers have suggested that spawning at high density reduces mean clutch size through increased interference during spawning and/or stress during incubation (Balarin and Haller, 1982; Rana, 1986). Macintosh and Sampson (1985) described the lower mean clutch size (fry) of females maintained at medium to

high $(3-12 \text{ fish/m}^2)$ compared to very high $(16-30 \text{ fish/m}^2)$ levels in aquaria. Spawning of very small fish in aquaria at these densities is quite likely to invite experimental aberrations that may not occur at the lower densities used in larger and commercial systems.

(c) Frequency of spawning and I.S.I.

The frequency of spawning in tanks (i.e. the mean percentage of females mouthbrooding at harvest) increased in all treatments over the first 30-45 days and then oscillated, mainly above the 40% level. In contrast, spawning frequency in hapas was more erratic but also tended to be higher initially (>45%) until water quality started to decline; during the period of early morning dissolved oxygen deficit, frequency of spawning varied between 10-40%. A trend of increasing production of fry over the first few harvests after stocking was also found in the sequential harvesting of fry from hapas by Hughes and Behrends (1983). Santiago et al. (1985) and Bautista et al. (1988) observed the same phenomenon in tanks and hapas. This trend is probably associated with differential maturity of new brooders and gradual familiarisation of fish to the spawning unit environment since it is noted to be most pronounced in treatments stocking young females (year class I, Hughes and Behrends, 1983).

Mean Interspawning Interval (ISI) of fish in tanks was within the range reported for aquarium-held individuals from which eggs were removed within 48 hours (Mires, 1977, 1982; Rana, 1986). On average, fish spawned every second time in the spawning unit, i.e. once a month, in all treatments except the highest density in which the ISI was significantly longer; the previous study (T2) indicated that variability in frequency of spawning was high probably because of hierarchical effects.

(d) Synchrony of spawning

Synchrony of spawning showed variation with density in tanks but not hapas. The lower percentage of stage 1 eggs (18.5%), in the low-density compared to high density tank (28.5%) suggested that spawning was delayed in the latter, suggesting that at a certain critical density the hierarchy becomes more unstable and territory establishment, courtship and spawning take longer. The poor synchrony of spawning found during periods of low early morning dissolved oxygen in hapas could also be explained by a decrease in the time available for courtship and spawning. The data presented by Hughes and Behrends (1983) for spawning of two densities of broodfish showed no clear indication of improved synchrony of spawning at the lower density in cages (although production was significantly different) based on relative numbers of fry, yolk-sac fry and eggs after a 21 day period. However, differences in synchrony are likely to be less clear using such a long interharvest interval.

(e) Effect of broodstock density and change in density over time

Comparison of the productivity of tanks and hapas during the early and late periods of the experiment showed that the

relative clutch size, number of eggs per kg of female and kg of fish declined with time but that productivity per unit area increased. A decline in relative fecundity with increasing age and size of female broodfish has also been reported previously by several authors (Hughes and Behrends, 1983; Siraj et al., 1983; Rana, 1986) but greater productivity per unit area for larger fish was explained on the basis that smaller fish were understocked (Hughes and Behrends, 1983). Guerrero and Guerrero (1985), stocking on the basis of biomass in large static tanks, found no significant difference between productivity using different sized broodstock.

The use of number or weight of fish as a basis for stocking is still unclear. At 110 days, T33 had a density 50% higher by weight than T34 at 45 days but a considerably higher seed output (4000 cf. 2750 seed/tank/day). Clearly, the number of fish is as important as biomass in intensive systems where hierarchy is an important control mechanism. In tanks, the optimal spawning density on first stocking continued to produce the most seed over the duration of the experimental period, despite the density weight more than doubling bу $(0.8 \text{ kg/m}^2 -)2 \text{ kg/m}^2$). This was much higher than the densities of small broodfish in 'spawning families' used in Israel in intensive aquaria and tanks $(0.4-0.64 \text{ kg/m}^2; \text{ Rothbard})$ Pruginin, 1975; Mires, 1977; Hulata et al., 1985). The optimum density of O. mossambicus breeders used by Uchida and King(1962) was about 12 fish/ m^2 (1.65 kg/ m^2).

The optimum density of broodfish for seed production in hapas appeared to depend on management and water quality maintenance.

An adequate exchange of water through the net is essential for grow-out cages holding large stocking densities of fish, but may be less critical for the spawning of relatively low densities of broodfish. The use of fine-mesh netting has been universal for spawning hapas but such material clogs rapidly under most conditions, reducing water exchange (Snow et al. 1983).

No significant differences were found in mean seed production using the three higher density treatments (H22, H23, H24) suggesting that spawning was relatively insensitive to densities between 1.5-3.0 fish/m in such a hapa environment. Hughes and Behrends (1983) described the lower output of seed using a higher stocking density (10 compared to 5 fish/ m^2) and ratio of female to male breeders (3 compared to females:males). Guerrero and Garcia (1983) stocked at a density of 4 fish/m² and Batao (1988) at 3 fish/m² (0.438 kg/m²); both densities were based on a sex ratio of 3 females: 1 male. Allison et al. (1976) found that fry production was inversely related to stocking density in concrete ponds over the range 4-20 fish/ m^2 . This suggests that at a critical density the hierarchy weakens, perhaps as dominant males cannot effectively control their territories, and aggression and interruption of spawning (Balarin and Haller, 1982) combine to reduce the number of successful spawning acts and/or fertilised seed.

(f) Growth and condition of broodfish

Growth and condition of broodstock after a period of spawning may be valuable indicators of the efficiency of the system. The improved mean growth rate and Food Conversion Ratio (FCR) in hapa treatments compared to tanks was probably related to the higher level of natural feed available to the fish in hapas. Feeding response, and thus intake, was higher in tanks across the range of densities tested, but remained below 2% body weight per day (BW/day). In hapas, a mean feeding to appetite could be calculated as a feeding rate of 1-1.5% over the period of production. Feeding levels given in intensive systems in other studies vary considerably. Hughes and Behrends (1983) fed fish 3% of initial BW/day in hapas suspended in clear water, static tanks. Guerrero and Guerrero (1985) reduced an initial feeding rate of 3% BW/day to 2% BW/day over the course of a 72 day breeding period in large static water tanks, and Snow et al. (1983) fed broodstock 2-3% BW/day in small plastic pools.

The increased growth observed in hapas during the period of poor water quality, and the subsequent pulse of spawning (with cessation of growth) after conditions improved may be an important reproductive strategy. Rana (1986) linked the conservation of gonadal products during periods of unfavourable spawning conditions and the channelling of energy into growth as an adaptation since larger fish are able to subsequently produce more eggs of a larger size when conditions improved. The experience of workers in Israel (Mires, 1982) that a change

of water in aquarium systems stimulates spawning supports this idea. Similarly, the breeding peak of fish held in hapas in Laguna de Bay follows the onset of the rains and subsequent improvement in water quality (Bautista et al., 1988). Evidence from field studies suggests that this may be a common phenomenon under natural conditions (Lowe McConnell, 1959) but it is contrary to the concept of 'stress-spawning' or runting (Fryer and Iles, 1972). Early maturation and precocious breeding of tilapia under unfavourable conditions, such as occur during the drying up of natural pools or in shallow fish ponds have been advanced as the basis for runted populations. However, the results of the present experiment suggested that spawning is halted before growth inhibition during unfavourable environmental conditions.

CHAPTER_4

SWIM-UP FRY PRODUCTION IN EARTHEN PONDS

4.1 Background

Earthen ponds are the main commercial systems for producing fry suitable for hormone treatment and mixed-sex fingerlings alike. Although yields of fry tend to be low compared with those possible in tanks and hapas (Table 1.3), management can be simple and cost of production low.

Most ponds in use are conventional shallow fish ponds, although various adaptations have been proposed to increase the efficiency of fry harvest (Lovshin and de Silva, 1975). A facility for easy water exchange is advantageous as for any fish pond. In Israel, the survival of swim-up fry is improved during batch harvest of fry by incorporating a large capture sump (5% of total pond area) in which the fry can be efficiently collected.

Stocking densities in earthen ponds are typically lower than in other spawning units and feed inputs can also be used economically in view of the contribution of natural feed to the fish diet. As stocking densities and feeding rates are lower, water exchange can be reduced or, in the case of total harvest and drainage, eliminated during the spawning cycle.

Earthen ponds can also be managed more intensively by increasing the frequency of fry harvest and by increasing the stocking density of breeders and rates of feeding and water exchange. The spawning of tilapia broodfish over a prolonged period, with intermittent harvest of fry or fingerlings by seining, is a common method of management (Table 1.2). Although many authors report a decline in production with time using

this technique (Lovshin and de Silva, 1975; Broussard et al., 1983) the simplicity and low-cost of the method is attractive. In addition, if the ponds have adequate fertiliser the reduced number of fry may be compensated for by their larger individual size (Broussard et al., 1983). Partial or intermittent harvesting tends to be conservative in terms of water and energy use.

The total production and proportion of fry suitable for sexreversal can be increased from systems in which fry removal is more frequent and the ponds are drained regularly restocked. Popma (1987) found that a high proportion of fry removed regularly with a large seine were sex-reversible even when they exceeded 14 mm total length after grading. Verdegem and McGinty (in press) reported that weekly seining of fry from the pond perimeter over a ten week period produced a fry harvest from which 75% could be sex-reversed. In Taiwan, fry are removed from earthen ponds on a daily basis by netting the perimeter of the pond several times during the day or at night with the aid of a light (Liao and Chen, 1983). Fry are collected with a dip net up to 6 times a day at two-hourly intervals starting early in the morning in the most productive pond systems in Pililia, Rizal Province, Philippines (Guerrero, 1985). In less intensive systems, fry are removed for nursing in hapas only once a day but regular draining (every 30-60 days) of the small ponds is a common feature.

The intensity or frequency of fry harvest in earthen ponds appears to be of major significance to the overall yield of

sex-reversible fry and an important variable cost, but has yet to be quantified under controlled conditions.

Earthen ponds in Israel are managed by regular total harvesting of the fry after drainage of the pond. Large numbers of fry suitable for sex-reversal can be produced at the same time. Grading fry is also unnecessary since harvested fry are spawned during the same period. The method may be constrained by the high requirement for labour at the harvest period and for water and energy during each spawning cycle. Partial harvesting is probably more appropriate to the needs of fry operations using family labour in most tropical developing countries.

Stocking densities in pond systems are generally dependent on the level of intensification, and the size of broodstock used appears to be highly variable. In Israel, two broodstock sizes (0.2-0.4 kg and 1.5-3.0 kg) were found in successive years to have the same productivity (1500 fry/female), suggesting that large numbers of the larger size class did not participate in spawning (Rothbard et al., 1983).

Large broodfish (0.6-1.0 kg) stocked at high density are used in commercial systems in Taiwan (Liao and Chen, 1983), although fry production is concentrated during the early part of the year. Guerrero and Guerrero (1985) found much higher fry production per individual for larger females when two size classes of broodfish were spawned for 30 days in large static water tanks at equivalent stocking densities (0.27 kg female/ m^2). Broussard et al. (1983) found no correlation between either broodstock number or biomass and fingerling production

in open-pond spawning over a 250 day production period.

The size of broodfish stocked may also be affected in practice by other factors such as availability and market value. In Thailand, larger tilapia can have a considerably higher market value than smaller fish and most farms producing tilapia fry cull broodstock for the market as a by-product. Silliman (1975) suggested that this practice constituted selection for slower growing individuals as broodfish.

Wongsaengchan (1985) has described the same negative selection practices with <u>Oreochromis niloticus</u> on farms in Northern Thailand, where large fish are removed for sale leaving the smaller fish of the same cohort for breeding. The consequences for fry production of this form of selection have yet to be investigated.

The management and production of swim-up fry using frequent partial harvesting was investigated in the present study because of the commercial importance of fry production from earthen ponds. Two sequential trials were carried out in large (0.17 ha) earthen ponds at AIT. The first trial estimated the effect of harvesting intensity (=fishing effort) on fry production.

The second trial studied the effect on fry production of stocking an equivalent biomass of broodstock of two-size classes of fish from the same cohort. Large broodfish are considerably more expensive per unit weight than small broodfish and they have been found to have a lower relative

clutch size (seed/kg female). Larger broodfish are commonly used commercially however, and this trial attempted to assess the relative advantages of broodstock size.

4.2 Methods

4.2.1 Pond preparation

The ponds used for Experiments 1 and 2 were rectangular earthen ponds (water surface area = 1740 m^2). A free-breeding monoculture of <u>O. niloticus</u> had previously been raised on septage in the same ponds (Edwards et al. 1987). A plan of the ponds in relation to inputs of water and septage is given (Fig. 2.5).

Two of the ponds were drained and dried for a period of two weeks before refilling with fresh water obtained from the AIT canal and commencing the first of the present fry production experiments (P1; section 4.3.1).

In preparation for the second experiment (P2; section 4.3.2), following the harvest of the first, the ponds were treated with sodium cyanide (approximately 8 mg/l) to kill any remaining fish. After this the ponds were left to dry and the sediments to crack for two months before refilling; sediments were not removed.

4.2.2 Broodstock

Origin and pretreatment

Experiment 1

Broodstock were obtained and pretreated for Experiment 1 after the harvest of a duck-fish experiment on AIT campus (see section 2.4.1.(a)).

Experiment 2

Broodfish from the first experiment (P1) were reused in the second experiment (P2). After harvest, the broodfish from the first experiment were separated by sex and placed in large nylon hapas (40 m²) suspended in an earthen pond. The fish were conditioned for a period of 2 months by feeding with a floating catfish pellet (crude protein content 30%) to appetite 3 times per day before beginning the second experiment.

4.2.3 Selection and pond management

Experiment 1 (P1)

After pond preparation was completed broodstock were selected, anaesthetized and batch weighed before release. A sex ratio of 1:1 was used, on the basis of number of fish, or 1:1.4 (female:male) on the basis of weight. Initial stocking densities of both treatments were based on using the same numbers of broodstock (317 fish/pond) although the biomass was also comparable (P1, 141; P2 375 kg/ha). A floating catfish pellet (crude protein 30%) was fed twice daily at a rate of 0.5% of initial stocking weight.

Septage was used as an organic fertiliser at a rate of 150 kg/ha/day loaded into the pond on a twice weekly basis at three times the daily rate.

Water was added from the AIT canal system as required to maintain a depth of 1-1.25 m.

Experiment 2 (P2)

The broodstock were removed from the conditioning hapas and

divided by eye into two size groups. The fish were then weighed in small groups after being anaestheti (initial stocking density 529 kg/ha). The numerical sex ratio was 1:1 equivalent to 1.14-1.25:1 by weight (female to male). Feeding, pond fertilisation and water management were the same as for Experiment 1, except that feeding was given twice daily to appetite.

4.2.4 Fry removal

Fry were removed from the shallow perimeter of the pond on a daily basis (6 days/week) by the same worker. Two types of hand-held nets were used to capture the fry. At first, a large (0.25 m² aperture) bag net was employed. This type of net, made of green mosquito netting material, is used in Taiwan on commercial farms. Later, this was replaced with a smaller net (0.06 m² aperture), manufactured from white mosquito netting which is widely available in Thailand. It was found easier to capture large groups of shoaling, recently released fry with a smaller net that could be moved more easily in the water. After stocking, the broodfish ponds were inspected daily and harvesting was begun from first sight of free-swimming fry at the pond perimeter.

Harvest frequency, or fishing effort, was based on the time that the fry harvester spent in the water removing fry. In practice, fry were removed at high frequency on the basis of harvesting six times per day (6 circuits) and three times a day

for the low frequency.

During fry removal, the net was kept almost continually in the water, the captured fry being maintained in the net by the velocity of the net moving through the water. Every few minutes, the harvester would remove the fry to a floating aluminium bowl attached by a string to his belt. Fry were removed from the bowl after each circuit and held in another shaded bowl on the pond dyke. Each circuit was timed to allow quantification of fishing effort.

Fry were separated from water boatman (Notonectidae) and other insects caught along with the fish fry using a method observed in the Philippines by fry producers. A small quantity (1-2 ml) of gasoline was poured into the aluminium bowl containing fry and insects. After a few minutes, the dead and floating insects could be separated from the swimming fry quite easily. It was found necessary to maintain low densities of fry in the bowl during the process (<5000 fry/bowl) and adequate water quality; a shortage of dissolved oxygen in the water would cause the fry to surface and become contaminated with the gasoline. Assorted pond debris was removed at this stage and other species of fish were removed for preservation and later identification. The tilapia fry were then preserved in 4% formaldehyde in sealed glass bottles for later analysis.

Fry were harvested in the same way in Experiment 2, except that both treatments were subjected to 'heavy fishing effort' comprising 6 harvest circuits a day.

4.2.5 Estimation of fry numbers

Fry preserved in formalin were first rinsed and washed to remove excess chemical. The fish were then passed through a fine net (mesh size 4 mm) to separate fry suitable for hormone treatment from those that were too large (old) for effective sex-reversal. Small freshwater prawns (Macrobrachium lanchesteri) were removed from samples with forceps.

Samples of the small fry were then taken randomly from the group and counted (3 x 100 individuals). Excess moisture was removed by drying the sample in a small net on paper tissue before weighing to two decimal places on an electronic top balance (Mettler model Pe 3600). The rest of the fry were then bulk weighed in lots not exceeding 30 g. Large fry were similarly treated, except that fewer fry were counted (3 x 50 individuals). Estimation of total fry numbers could be made to an acceptable degree of accuracy (1.5-3.5%, depending on number of seed according to Tuan, 1985) using this method.

4.2.5 Broodstock harvest

Experiment 1

After the experimental period (116 days from stocking), the ponds were drained on successive days and all the fish harvested. Stocked and recruited tilapia were separated, counted and bulk weighed. Other species of fish found on harvest were identified, counted and weighed. These included some cultured species which had been released accidentally in the ponds (4.3.1). Samples of broodstock were taken for individual weight, total length and gonad weight determination,

and body and gonad composition analysis.

Experiment 2

After an experimental period of 105 days, the ponds were drained on successive days and the surviving fish harvested. Stocked tilapia were separated from recruits which were sorted into three size classes (<10 cm; 10~<15 cm; 15->15 cm) and bulk weighed. No other species of fish were present at harvest.

Water Quality

Water quality parameters were measured regularly throughout both experimental periods (Appendix 1). Total ammonia, pH, chlorophyll a and dissolved oxygen were measured weekly from samples taken at two points within each pond. Temperature was measured daily using a maximum-minimum thermometer suspended 30 cm below the water surface.

Table 4.1 Mean number of fry per 5 days of two size classes removed by dip-net from the shallow margin of large earthen ponds (Area = 1740 m^2) over an experimental period of 105-116 days

System/ Experiment/ Treatment	MEAN + S.E.			
	Small fry ¹	F	Large fry ²	F
P12 P13	21403 ± 3763 13369 ± 2062	3.3n.s.	132 ± 14.3 106 ± 10.4	2.1 ⁿ ·s·
P22 P23	19704 ± 4516 48131 ± 7914	8.9**	130 ± 13.1 207 ± 17.7	11.9*

- P12 Fry removed 6 times per day; broodstock of mixed size class
- P13 Fry removed 3 times per day; broodstock of mixed size class
- P22 Fry removed 6 times per day; broodstock of large size class
- P23 Fry removed 6 times per day; broodstock of small size class

F value one-way Anova

P < 0.01

- n.s. P > 0.05
- P < 0.05

small fry (mean weight <0.015g) suitable for hormone treatment

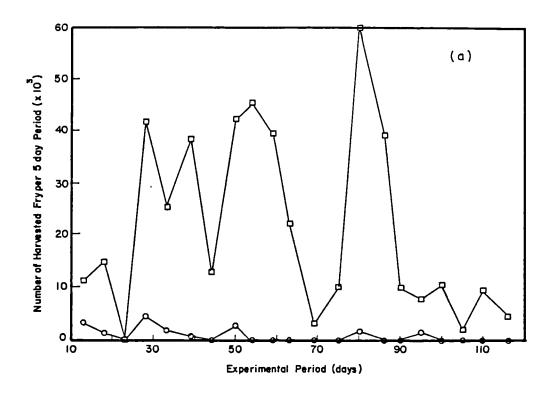
² large fry (mean weight > 0.015g) unsuitable for hormone treatment

Fig. 4.1

Production of two size classes of swim-up fry removed by dip-net from the margin of large earthen ponds (Area = 1740 m²)

- (a) at high harvest intensity (6 times per day) and(b) low harvest intensity (3 times per day)

Stocking density 4215 fish/ha; sex ratio 1:1 (by number) square: small, treatable fry (mean weight < 0.015 g), circle: large, untreatable fry (mean weight > 0.015 g)



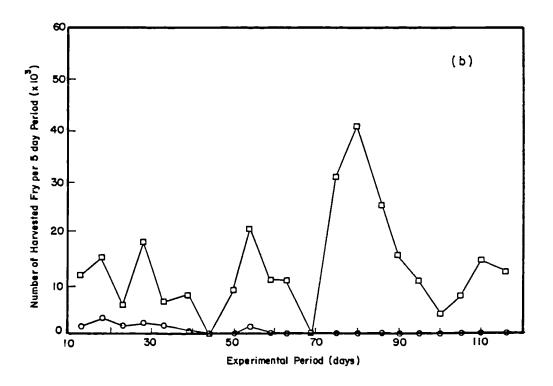


Table 4.2 Oreochromis niloticus fry suitable for hormone treatment as a percentage of total fry harvested by dip-net from the shallow margin of large earthen ponds (Area = 1740 m^2) over a study period of 105-116 days

Experiment/ Treatment	Mean ± S.D.	F
P12 P13	96.1 ± 6.3 93.6 ± 29.4	2.85n·s.
P22 P23	84.7 ± 29.4 99.9 ± 0.2	4.8*

F value one-way Anova n.s. P > 0.05

* P < 0.05

Table 4.3 Efficiency of removal of two size classes of fry by dip-net from the shallow margin of large earthen ponds (Area = 1740 m²) at two levels of harvest intensity (t = 116 days)

Treatment	Harvest intensity	Number of fry harvested per minute (MEAN ± S.E.)		
		SMALL FRY®	LARGE FRYb	
P12 P13	HIGH	43.21 ± 3.9 45.56 ± 8.0	3.66 ± 0.57 10.96 ± 1.66	
F		0.07n.s.	28.26***	

a small fry (mean weight <0.015g) suitable for hormone treatment

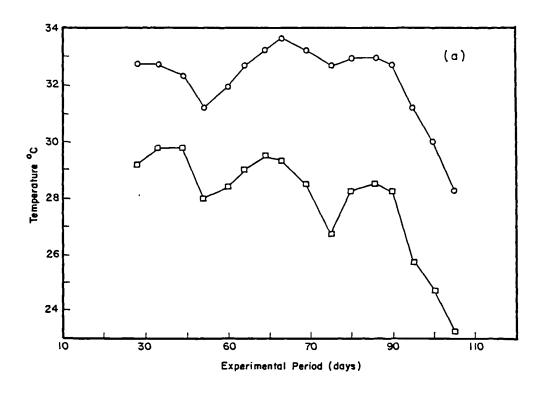
F value one-way Anova

n.s. P > 0.05 *** P < 0.001

b large fry (mean weight >0.015g) unsuitable for hormone treatment

Fig. 4.2

Mean maximum (open circle) and minimum (open square)temperatures measured daily at 20 cm below the water surface during swim-up fry production in large earthen ponds (a) at different levels of harvest intensity (P1) and (b) using two size classes of broodfish (P2) (5 day means plotted)



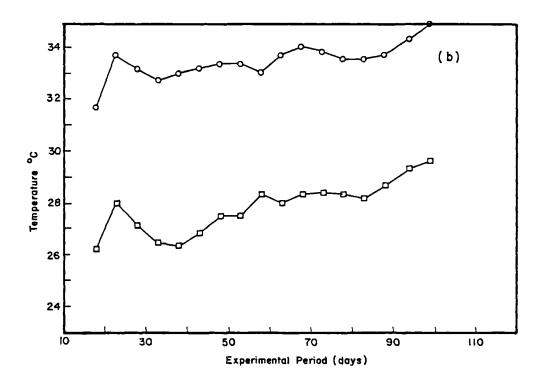


Table 4.4 Characteristics of female broodfish at harvest following fry production in large earthen ponds (Area = 1740 m²) using (a) different harvest intensity (P1) and (b) broodfish of different size class (P2) (P1 = 116 days, P2 = 105 days)

	MEAN ± S.E.				
Treatment/ Experiment	Total length (cm)	Body weight (g)	Gonad weight (g)	GSI ¹	Condition factor ²
(a)	19.2±1.4	194± 9.6	3.3±0.5	1.7±0.3	1.64±0.04
P12	19.2±1.4	1941 9.0	3.310.0	1.720.5	1.0420.04
P13	20.7±1.1	197±10.3	3.2±0.5	1.7±0.3	1.76±0.03
F	0.74n.s.	0.07n.s.	.002 ^{n.s.}	.01n.s.	4.6n.s.
(b)					
P22	24.0±1.1	300± 8.5	6.8±0.6	2.4±0.2	1.62±0.02
P23	19.4±1.3	218± 8.6	5.0±0.5	2.5±0.2	1.78±0.03
F	7.31*	45.9***	5.5n.s.	0.5 ^{n.s.}	18.6***

F value one-way Anova

 $n \cdot s \cdot P > 0.05$

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

¹ GSI = weight of reproductive organ x 100/weight of fish

² Condition Factor = Body weight x 100/total length³

4.3 Results

4.3.1 Effect of harvest intensity

The treatment removing swim-up fry by frequent harvest (6 times daily, P12) of a large earthen pond (1740 m²) produced a mean 5 day harvest 60% higher than the treatment removing fry less frequently (3 times daily, P13). However, the difference between means was not significant (P>0.05; Table 4.1, Fig. 4.1) because of the high variability over the experimental period. Fry production was estimated for five day intervals to make data comparable with the tank and hapa experiments in which harvesting was on a five day basis. Output ranged from less than 5,000 fry to 60,000 fry over a five day interval. The ratio of small fry (those judged suitable for hormone treatment) to larger fry caught at the same time was slightly greater for intensive harvesting, although not significantly so (P>0.05; Table 4.2).

The number of small fry removed per unit effort (number/min harvesting) was not different between treatments (P>0.05), but the harvest efficiency of larger fry was doubled in the low labour treatment (P<0.001; Table 4.3). Water quality remained high throughout the experiment (Appendix 1). A fairly rapid decline in water temperature in the last month of the experiment may have contributed to the fall off in production observed in the last 20 days of the trial (Fig 4.2 (a)).

At harvest, the growth and gonadosomatic index of broodstock in the two treatments was found to be similar although the

Fig. 4.3

CS = other species

Main categories of harvested net production (wet weight) from partial and final draining of large earthen ponds (Area = 1740 m) after experiments using (a) two levels of harvest intensity (P1) and (b) two size classes of broodfish (P2)

Stocking density:
P1, 4125 fish/ha (sex ratio 1:1 by number)
P2, 528 kg/ha (sex ratio 1:1 by weight)
Experimental period: P1, 116 days; P2, 105 days

R = recruits
M = stocked male broodfish
F = stocked female broodfish
UF = untreatable fry
TF = treatable fry

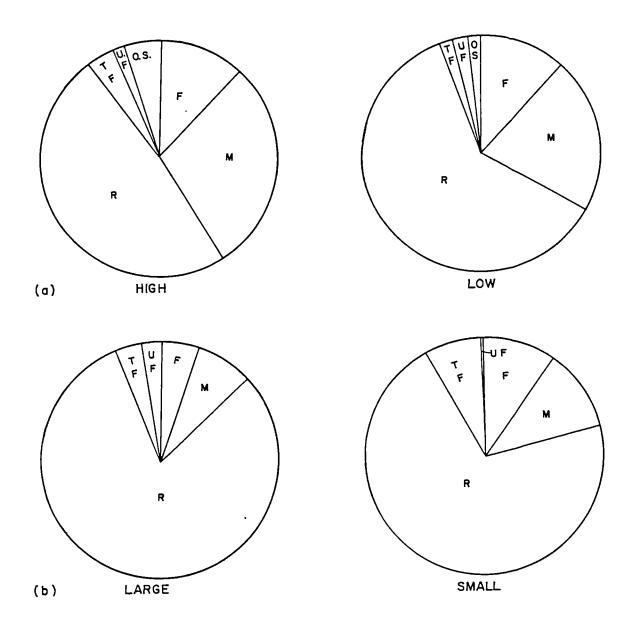
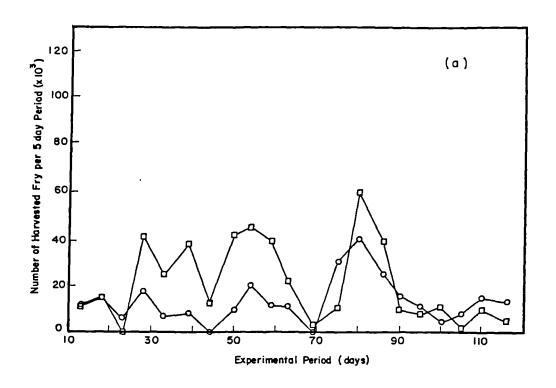
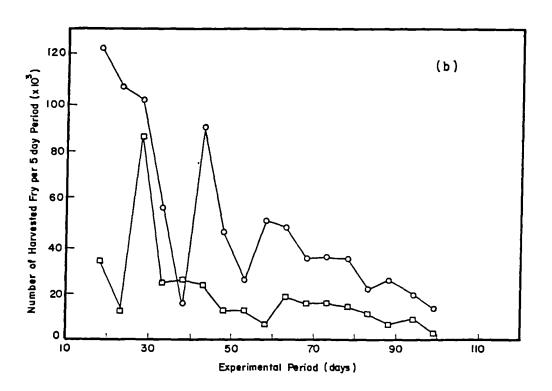


Fig. 4.4 Production of swim-up fry suitable for hormone treatment (mean weight < 0.015 g) in large earthen ponds (Area = 1740 m²) (a) at different levels of harvest intensity (P1; square , high intensity; circle, low intensity) and (b) using two size classes of broodfish (P2; square, large fish; circle, small fish) Stocking density: P1, 4125 fish/ha (sex ratio 1:1 by number); P2, 528 kg/ha (sex ratio 1:1 by weight)





condition of females in the low harvest intensity treatment was significantly higher (Table 4.4). The total harvested biomass on draining the ponds was found to be similar in both treatments (High, P12, 1298 kg/ha equivalent; Low, P13, 1480 kg/ha equivalent). Female broodstock maintained a similar growth rate during the trial in both treatments (High, P12, 0.84 g/day, Low, P13, 0.95 g/day).

At harvest, tilapia recruits, i.e. fish that had been spawned in the pond but evaded capture as swim-up fry, were found to be the largest source of total net production from the pond in both treatments (Fig 4.3). A larger percentage of net production were present as recruits for the low intensity treatment (>61%) than the high intensity (<50%).

The other species of fish found both throughout the experiment and at harvest included a variety of wild fish (e.g. <u>Anabas testudineus</u>, <u>Channa striata</u>) and some omnivorous/herbivorous cultured species (e.g. <u>Puntius gonionotus</u>, <u>Labeo rohita</u>, <u>Aristichthys nobilis</u>).

4.3.2 Effect of broodstock size

The stocking of small broodfish produced more than double the output of swim-up fry than a similar biomass of larger broodfish stocked in ponds of equal size and harvested with equal intensity (Table 4.1). In both treatments, production declined over the experimental period but breeding appeared to be more synchronised.

Table 4.5 Synchrony of fry production in large earthen ponds (Area = 1740 m^2) as indicated by the proportion of total fry harvest removed by dip-net in the shallow pond margins during the first 33 days (Experimental period; P1 = 116 days; P2 = 105 days)

Total number of fry harvested				
Experiment/Treatment	synchronous production(a)	total production(b)	Ratio of (a):(b) (%)	
P12	92977	449462	20.7	
P13	58397	280745	20.8	
P22	159465	334962	47.6	
P23	387571	866363	44.7	

- (a) number of swim-up fry produced in days 1-3(b) number of swim-up fry produced over total experimental period

In the first 50 days of the experiment, fry output from both treatments peaked at a level twice that found in either of the treatments in the previous trial (small broodfish, 120,000 fry/5 day period; large fish, 90,000 fry/5 day period; Fig. 4.4). More than 40% of the total fry produced were harvested in both treatments within the first 33 days of the second experiment compared to around 20% during the same period in the first experiment (Table 4.5; Fig. 4.4).

The ratio of small, sex-reversible fry to large fry was significantly greater (P<0.05) in the treatment using small broodfish and this was independent of the much greater production of small fry <u>per se</u> (Table 4.2).

Very few larger fry (mean individual weight >0.1 g) were caught during daily harvest in this treatment (<1000 fry throughout the whole experiment).

Water quality remained high throughout the experiment although both minimum and maximum temperatures climbed to a mean above 27° C and 33° C respectively, for the last 60 days of the experiment (Fig. 4.2 (b)).

The gonadosomatic index (GSI) of female broodfish at harvest was similar between treatments. However, average GSI in Experiment 2 (P2) was greater for both treatments than in the previous experiment (P1) (Table 4.4). There was an average weight loss of 0.2 g/fish/day among large females (P22) and an average weight gain of 0.11 g/fish/day among small females (P23); the mean condition factor of large females was lower (P<0.001) than that for small females.

Final total biomass at harvest was of the same order as the previous trial (1353-1532 kg/ha) although recruits formed a much larger proportion of total net production (P12, 48.5%; P13, 61.2%; P22, 80.6%; P23, 71.0%) (Fig. 4.3)

4.4 Discussion

Effect of harvest intensity

A freely breeding population of tilapia constitutes a 'micro-fishery' for which the concept of increased 'yield' for increased harvesting effort should apply.

The higher yields of fry removed for increased effort in the first experiment were therefore characteristic of any capture fishery (Rothschild, 1986). Greater fishing effort yielded more fry throughout the experimental period although variability with time was extremely high (CV=82%).

Rothbard et al. (1983) harvested all swim-up fry after draining earthen ponds on a 21 day cycle and also reported high variability of F1-hybrid (O. niloticus x O. aureus) yields with time (mean 513,000 fry/ha/harvest; S.D.=629,000; n=9). Verdegem and McGinty (in press) seined O. niloticus fry from the periphery of ponds (0.7 ha) on a weekly basis and reported a similar level of variability of output between seine hauls (CV=87%). Undoubtedly, a large part of the variability in spawning activity was related to a degree of synchrony of breeding in populations of Oreochromis.

Environmental factors, especially temperature, may have been important in synchronising broodfish maturation and spawning. The two major declines in fry yield in both treatments at around days 45 and 70 correlated with a decline by about 2°C in mean water temperatures on each occasion. The corresponding spawning peaks that followed these declines suggested that a small rise in temperature can again stimulate synchronised

spawning of breeders.

Behrends and Smitherman (1983) concluded from their studies of Oreochromis maturation in warm water effluents that spawning became more synchronous as environmental conditions approached an optimum. Hughes and Behrends (1983) correlated a spawning pulse in hapas to a sudden rise in temperature followed by a period of low temperatures. McAndrew (quoted in Balarin and Haller, 1982) claimed that breeding could be synchronised (> 50% of females spawned) by reducing ambient temperatures from 25°C to below 18°C for two weeks before increasing it again. Srisakultiew and Wee (1988) found that maintaining breeders for such long periods at low temperatures (22°C) merely caused gonad regression, but that synchrony could be improved (10-25%) by a short period of cooling (6-24 hours) before a return to high ambient temperatures (30°C). The short period required for oocyte maturation prior to breeding of Oreochromis therefore appears to be sensitive to short-term, and relatively small changes, in temperature.

More intensive harvesting had a negligible impact on the proportion of 'large' fry. The number of large fry that had to be removed at both levels of harvesting intensity was a very small proportion of the total catch (4-8%) compared to a weekly harvest by seining the periphery of the pond in which over 20% were considered too large for hormone treatment (Verdegem and McGinty, in press).

The difficulty of removing larger fry at either harvesting intensity suggests that larger fry can more easily evade

capture and/or no longer congregate along the perimeter of the pond in the same way as very young fry. Shoals of just-released fry were relatively easy to remove from the shallows of the ponds and efficiency of harvest was relatively high.

Uchida and King (1962) found very few recruits at final harvest after removing swim-up fry on a daily basis from the edge of broodstock tanks but did observe some variation in the size of fry netted. In more intensively stocked clear water tanks predation by other broodfish may be a very important form of control of fry left in the system. In this study recruits represented a smaller proportion of total production in the high effort treatment but were still the largest component of net yield from the pond.

A high density of recruits might eventually depress fry yields through competition with the broodfish for feed and territory. Mires (1982) reported that when ponds became overcrowded with fry in Israel, stocked broodfish ceased to spawn. Regular draining of ponds in which swim-up fry are produced appears to be the most effective way to reduce the negative effects of recruits. Farmers in the Philippines drain their ponds, harvest remaining recruits and restock broodfish on a monthly basis (Guerrero, 1985). In Israel the total harvest of ponds every 21 days insures that recruits are not a problem. The cost of regular draining may be high in terms of water use (Verdegem and McGinty, in press) but actively spawning tilapias are maintained at depths as low as 50 cm on commercial farms in the Philippines (Guerrero, 1985) and pond effluents are recycled

within the wider agricultural system in Israel (Hepher and Pruginin, 1981).

GSI's of females at harvest in both treatments were low (<2%) which corresponded with a decline in production and temperature over the last month. Overall condition of female broodfish at harvest was high especially in the pond harvested at low intensity (P13). Spawning activity may have been inhibited proportionally more by the larger numbers of recruits present in this treatment, thus allowing more diversion of food resources to body growth. The level of harvest intensity may be critical for both the quantity and sustainability of fry output, although this would have to be confirmed by longer term studies and under a more stable temperature regime.

Effect of Broodfish Size

The stocking of broodfish on the basis of biomass rather than number has been proposed for earthen ponds by Broussard et al. (1983) and for large static water tanks by Guerrero and Guerrero (1985). This study revealed that large productivity gains were made in terms of swim-up fry yields by using smaller broodfish (female mean size 207 g) rather than larger fish (female mean size 262 g).

The inverse relationship between relative clutch size and broodfish size in <u>Oreochromis</u> has been reported by many authors (Lowe McConnell, 1955; Peters, 1983; Hughes and Behrends, 1983; Siraj et al., 1983; Rana, 1986). Most of these reports relate to larger fish which were also <u>older</u>, but in the present study

the two size classes originated from the same group and the differences in fry production were considered to be too large to be accounted for simply in terms of a larger relative clutch size.

Frequency of spawning may also vary with size of fish smaller females have been observed to complete spawning more quickly than larger females (Rothbard and Pruginin, 1975). Siraj et al. (1983) described a decline in the frequency of successive spawns with larger females although initially their spawning rate was higher. The results of this study were in contrast to the findings of Guerrero and Guerrero (1985) who found no significant differences in fry production (fry/m²/day) using an equal biomass of different sized fish over a 21-30 day breeding period. But over such a short period of spawning, the initial spawning rate was probably more important than recovery time and frequency of spawning and a lower relative clutch number may be compensated by better survival of the eggs produced. Lee (1979), Siraj et al. (1983) and Rana (1986) all demonstrated the improved survival of the progeny of larger, older breeders. Rana (1986) proceeded to show that it was due improved incubation success rather than any inherent superiority in egg quality among larger fish.

In a system in which brooders are stocked for extended periods of time the improved productivity of smaller breeders probably stems from their ability to recover more quickly after incubation. In an semi-intensive spawning environment in which natural feed contributes a larger proportion of total feed

requirements, this may be of particular importance.

An early pulse of spawning followed by a gradual decline in production is a well recognised phenomenon in both extensive tilapia breeding systems (fertilisation only; Broussard et al., 1983;) and semi-intensive systems (additional feeding; Guerrero and Guerrero 1985) in which broodstock are not exchanged and this was shown clearly in the second experiment. The small broodfish treatment showed a succession of gradually weaker pulses indicating a steady reduction in spawning activity. The ability of broodfish to recover after spawning in ponds may have been reduced after an initial spawning pulse in which a proportion of fry evaded capture to quickly form a large cohort of recruits that then competed with their parents for feed. In the first experiment this effect was probably less important as mean broodfish size and standing stock were smaller and breeding, and hence recruitment and competition, started later and were less intense.

The much larger ratio of small to large fry removed daily from the small broodfish treatment, compared to those produced from larger fish, suggested that the fry from the latter managed to evade capture during the early stage more easily. This may be explained by the higher incubation success apparent in larger fish (Rana, 1986) continuing into the nursing period and/or because larger fish tend to nurse their individually larger clutches for a longer period or release them in deeper water. Lowe McConnell (1955) suggested that extended nursing was common under natural conditions when cover, in the form of weed

beds, was lacking but in this experiment the amount of cover available for nursing females and fry was the same. She also commented that extended retention of fry by the mother might be related to the frequency of spawning, and that many batches produced in succession might be a greater stimulus to cease brooding.

CHAPTER 5

INCUBATION

5.1 Background

The high fecundity of many fishes is countered by high mortality during the pelagic larval stage in natural environments (May, 1974), and the controlled incubation of eggs offers potentially vast increases in larval survival. The incubation of eggs in suitably designed vessels is commonplace for many farmed fish species but the hatching of <u>Oreochromis</u> eggs on a commercial-scale has yet to become a reality.

Mouthbrooding has been described as the most advanced type of parental behaviour in fishes (Oppenheimer, 1970) and has considerable implications for the mass production of high quality tilapia fry (Rana, 1986). The development of artificial incubation for Oreochromis spp. has mainly been limited to small experimental systems in which individual clutches of a few hundred eggs have been hatched. Poor survival has been commonly reported for seed that has been naturally incubated for less than 48 hours before transfer to an artificial incubator. Methodologies for the large-scale production of eggs and yolk-sac fry, necessitating facilities that can incubate seed that has been just spawned, have been described in the preceding chapters.

Artificial incubation of fish eggs has a far longer history in salmonid culture than in carps the other major 'traditional' group of cultured fish (Huet, 1972), but carp hypophysation from which vast numbers of eggs are produced at the same time and hatched over a few hours has undoubtedly stimulated the development of artificial incubation for fish in general.

Salmonid eggs were, and still are, incubated by placing static eggs in a flow of running water in different types of hatching trays.

Static incubation is often used for incubation of sticky or adhesive eggs such as those of the common carp (Cyprinus carpio) or the walking catfish (Clarias macrocephalus) in which survival can be improved by an exchange of water around the substrate and/or a low density of eggs during incubation. Substrates that increase the surface area for attachment of incubating eggs, such as kakabans for common carp eggs, may further improve efficiency (Huet, 1972), but in general the incubation of fish eggs in up-welling systems has become the dominant hatchery method world-wide.

An important objective of any incubation system is that water movement is reliable, smooth and of constant velocity to prevent embryos or larvae being stressed by mechanical, chemical or thermal disturbances (Devauchelle et al., 1986). This can be achieved by the use of high quality water in a suitable up-welling incubator e.g. Weiss-type or down-welling Macdonald jars (Bagenal and Braum, 1968).

The up-welling principle is now used for incubating eggs of many species of fish, especially the Chinese and Indian Major carps but also those of common carp after removal of stickiness (Woynarovich and Horvath, 1980) and even salmonid eggs (Poxton and Murray, 1987). However, it has proved a less successful system for Oreochromis eggs in experimental tests.

The specialised oral incubation habit of <u>Oreochromis</u> spp. has proved a difficult process to imitate. The susceptibility of the large yolky eggs of mouthbrooding tilapias to infection by bacteria and fungi has long been known (Shaw and Aronson, 1954) but more recently the poor survival during incubation has been linked with the mechanical trauma induced by most designs of incubator (Rana, 1986).

Systems in which tilapia eggs remain static (Shaw and Aronson, 1954), or are moved by aeration (Lee, 1979), mechanical shaking (Lee, 1979; Suliman, 1987) or by the movement of water (Rana, 1986; Suliman ,1987; Don et al., 1987), have all recorded poor or highly variable incubation success. A downwelling design in which recirculated water is filtered to remove micro-organisms has shown greatest potential on a smallscale but has yet to be proved a practical alternative to harvest of swim-up fry after natural incubation under largescale conditions. However, the removal of eggs has been shown to reduce the Interspawning Interval (ISI) of broodstock under experimental conditions (Dadzie, 1970; Lee, 1979; Macintosh and Sampson, 1985; Rana, 1988). Egg removal, if followed by successful artificial incubation, could therefore reduce broodstock costs under commercial conditions and has more potential for multispawning fish such as tilapias that produce small clutches of eggs regularly, than for annual spawners. The notion that cultured tilapias, with adequate inputs of technology and genetics, are set to follow the broiler chicken and become a major animal food commodity is not new (Maclean,

1984). An analogy between mouthbrooding tilapias and egg producing chickens is of greater significance to this study since it suggests that the adoption of artificial incubation is a key step towards intensification of tilapia fry production. In poultry the separation of egg production from incubation has had a major and fundamental impact on the productivity. Naturally, parental broodiness and post-hatching nurture are inseparable activities but artificial incubation and intensive nursing together with selection in modern breeds of hen have made them redundant (Cole, 1966). The onset of broodiness and natural incubation in birds normally suppresses further egg production, a strategy that enables a clutch of young to be . incubated and raised together. This behaviour tends to reduce the level of feeding possible by the incubating parents and, in turn, the number of eggs produced. The ancestor of the modern chicken, the red jungle fowl (Gallus gallus) had an annual egg production of 25-50 which was increased to 100-150 by removal of eggs alone; subsequent selection and improved nutrition have in turn allowed this production figure to more than double (Cole, 1966).

Experimental design

The main aim of these experiments was to construct and test a down-welling incubation system for <u>Oreochromis</u> eggs suitable for commercial application. This would then allow a comparison of the different seed production strategies using tanks and hapas with swim-up fry production from static water earthen ponds. The design criteria included (1) a capacity for

incubating a large number of egg clutches of different stages at the same time (>100,000 eggs) i.e. equivalent to the eggs harvestable from a series of large hapas, (2) conservative water use, (3) low cost and constructible from available materials and (4) capable of simple management and maintenance. The system design was required to provide adequate quantities of bacteriologically clean water and avoid mechanical trauma to the eggs.

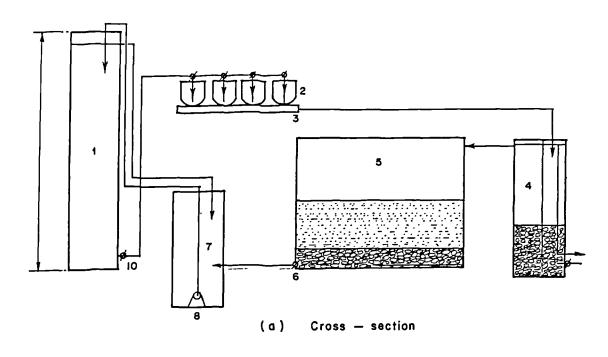
A down-welling design of incubator using recirculated water filtered through a UV-filter (Subasinghe and Sommerville, 1985) was used as the basic model; this had given good results for incubation of Oreochromis eggs on a laboratory scale (Rana, 1986; Suliman, 1987). However, ultraviolet light sterilisation, used widely in hatchery production of high value salmonid and marine fish fry (Muir, 1982) was not considered a practical alternative for hatcheries in most developing countries. A slow-sand filter was substituted as an alternative method of filtration.

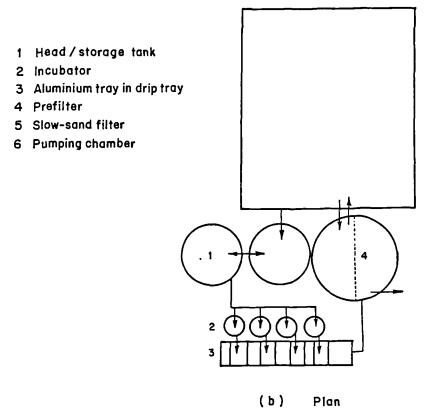
Slow-sand filters have been used extensively for safe drinking water production in both developed and developing countries (Ellis, 1987). Their low maintenance demands and ease of construction and operation make them appropriate for a wide range of fry production situations.

The design of incubators, piping, valves, pump and drip trays was influenced by their availability and cost. All materials used in the construction were available at the local district level (Amphur) throughout Thailand.

Fig. 5.1

Incubation system used for experimental trials in the AIT hatchery suitable for incubating > 100,000 seed at a time (a) cross section (b) plan

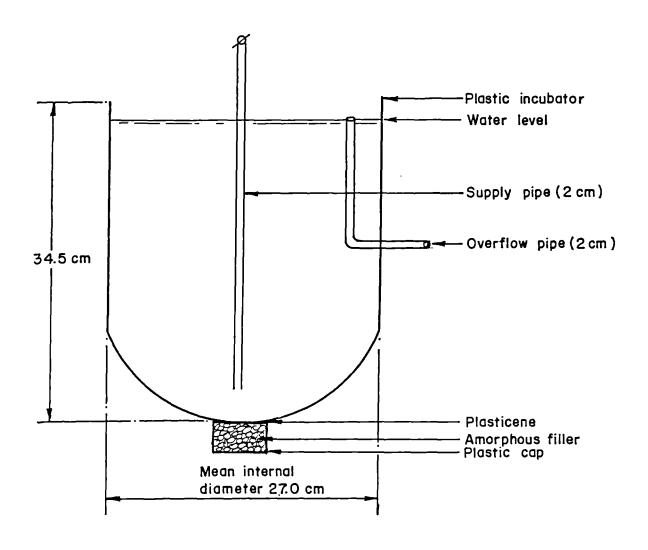




INCUBATION SYSTEM

Fig. 5.2

Design and dimensions of incubation bottle (Volume = $20\ 1$) used in experimental trial in the AIT hatchery



5.2.1 Methods

5.2.1.1 Experimental system

The apparatus used for the incubation experiment is illustrated in Fig.5.1. It was positioned within the AIT hatchery as shown in Fig. 2.2. Incubators were fabricated from 20 1 food-grade plastic water bottles. The bottom of the bottle was cut off and a hole bored in the side for the outlet pipe (Fig 5.2). The neck of the inverted bottle was filled with inert material and covered with an oil-based filler ('plasticine') to form a round bottomed container. Four incubators were placed in parallel within a closed recirculated water system. Water was delivered to the base of the incubators via a PVC pipe, from a concrete header tank (1). Swim-up fry and wastewater left the bottle via a small internal standpipe into a perforated aluminium tray lined with a fine nylon mesh. Water then flowed through a primary gravel filter (4) into a slow-sand filter (5). Flow rate through the filter was controlled by a valve (6) at 25 1/min. Clean water was collected in a concrete sump (7) and pumped continuously into a header tank with a 0.4 kw electrical submersible pump (8). A standard head of water pressure was maintained by allowing the overflow to cascade through a pipe (9) back into the sump. The system was drainable at two points via valves (6 and 10).

5.2.1.2 Experimental methods

Clean, chlorinated water was obtained from the AIT water supply; the system was filled and run for 12 hours before each incubation experiment. Dead eggs were removed from viable eggs as far as possible by careful washing before weighing and counting eggs of the same stage prior to placing them in the incubators. Flow rates for each incubator were adjusted to a minimum to avoid mechanical trauma, but actual flow rates varied according to the numbers of seed in the incubator.

Hatched fry were washed into aluminium trays where they continued to develop until yolk-sac absorption was complete. Fry numbers and individual size were then estimated (see above). Incubators, aluminium trays and the drip tray were cleaned out before the next incubation experiment. Top up water was added to the system from the AIT supply as required; the slow-sand filter required no maintenance or cleaning throughout the experimental period. Flow rate was adjusted and measured each day with a stopwatch, beaker and graduated flask. Temperature, total ammonia, pH and nitrite were also monitored daily from samples of water taken from inside each incubator. Total bacteria were estimated over a single incubation trial. Water were collected from the outlet pipe using a sterile samples 300 ml BOD bottle every six hours. Total bacteria were enumerated using a standard plate count of 1 ml of sample following standard methods (APHA, 1981).

Table 5.1 Percentage survival to swim-up of <u>Oreochromis</u> seed of different stage harvested from (a) tanks and (b) hapas and placed in 20 1 downwelling plastic incubators supplied with recirculated, slow-sand filtered water in three sequential trials (n = 3)

Seed stage stocked in incubator	Percentage survival to swim up fry stage				
	MEAN \pm S.D. (n = 3)				
Incubator	TANK	IIAPA			
1	26.2 ± 22.9	31.8 ± 18.0			
2	71.7 ± 1.7	68.8 ± 22.3			
2-3	82.5 ± 11.3	48.9 ± 45.2			
3	22.6 ± 27.0	33.9 ± 40.6			
Overall	48.9 ± 32.1	44.8 ± 31.9			

Seed stages

Stage 1: uneved; Stage 2: eyed; Stage 2-3: imminent hatching;

Stage 3: hatched

Table 5.2

- (a) Number of seed and percentage survival to swim-up stage of <u>Oreochromis</u> seed harvested from spawning tanks and hapas (combined) and incubated in 20 l plastic incubators supplied with recirculated, slow-sand filtered water in 10 sequential trials
- (b) Maximum density of seed during incubation in the same vessel

Sand		Stocked number of seed¹ per incubator							
Seed stage		1		2		2-3		3	
Trial	Number	Survival*	Number	Survival%	Number	Survival%	Number	Survival%	
1 2	92810× 58238	42.5 83.1	56444 ^y 30703	85.7 69.8	14404 49287	96.6 69.9	48986 690912	80.7	
3 4	18476 72765	47.4 57.2	6064 28416	77.3	22981 42913	66.0 77.3	5288 10900	84.2 60.7	
4 5 6	25608 20417	71.8 66.0	53437 25098	92.0 71.4	72468² 24375	76.4 88.1	23738 29176	77.9 73.2	
7 8	68948 19757	53.1 53.5	8952 11025	64.1 81.6	34943	81.6	8393 26543	64.2	
9 10	20566 3229	56.4 50.9	49205 5895	90.4	38463 28324	54.7 82.4	22094 48576	83.7 63.4	
Overall Mean S% S.D.		58.18ª		80.23° 11.10		77.00b 11.68		72.87° 8.24	

(b) Maximum density of seed during artificial incubation in 20 litre plastic vessels in trials as (a)

Max. number of seed	mean wet seed weight	Density of seed (g/l) per		
stocked	(g)	incubator	2 1 of capacity	
	0.41 .	23.0	190	
x	0.40	14.1	113	
) y	0.36	30.0	240	
z	0.39	16.9	136	
]	·		ļ	

¹ Seed stages: Stage 1, uneyed; Stage 2, eyed; Stage 2-3, imminent hatching; Stage 3, hatched;

percentage of seed survival to swim-up

F value one-way Anova ** P < 00.1

weight of 100 seed with surface water removed

Table 5.3 Total bacteria in incubation water as measured by standard plate count¹ during a complete incubation cycle in four 20 l plastic vessels supplied with recirculated, slow-sand filtered water

Period of incubation		Number of cells/ml incubation water Mean (S.D.)							
Hours after stocking	n	1	2	3	4	Mean	Control		
0- 30	6	185 (177)	326 (250)	433 (482)	86 (95)	257 (207)	23 (20)		
36- 60	5	699 (366)	3317 (2998)	628 (419)	857 (684)	1375 (590)	25 (13)		
66-108	8	194 (135)	231 (92)	249 (142)	181 (116)	214 (114)	15 (6)		

¹ estimated by standard plate count (APHA, 1981)

Table 5.4 Physico-chemical parameters of the system water during incubation of <u>Oreochromis</u> seed in 20 l plastic vessels supplied with recirculated, slow-sand filtered water

Parameter	Range	. Mean
Temperature (°C)	27.5 - 31.3 6.4 - 7.9	28.7
Dissolved Oxygen (mg/l) Total ammonia-N (mg/l)	0.01 - 0.01	0.01
Total nitrite-N (mg/l)	0.00 - 0.05	0.02
pH	8.0 - 8.5	8.4

5.2.2.Results

Eggs and yolk-sac fry obtained from tanks and hapas showed no significant difference in hatchability and survival (Table 5.1) and batches were subsequently combined for expediency.

Egg density within the range used (11,000-92,810 seed/incubator, equivalent to 688-5800 seed/1) also had no observable effect on hatching success ($r^2=0.05$).

However, survival to swim-up fry was related to the stage of egg development at harvest, the survival of stage 1 (uneyed eggs) to swim-up fry being significantly lower than the other stages (P<0.05; mean stage 1 = 58%; other stages combined 77%) (Table 5.2a).

Total bacterial levels within the incubator were monitored through only one trial and results are therefore only preliminary (Table 5.3). Bacterial levels peaked (mean 1375 cells/ml) from 36-60 hours after placing the seed in the incubators before returning to much lower levels (<500 cells/ml). At the times high levels of bacteria were measured, clumps of infertile eggs were observed in the incubators that were not being washed out efficiently. Control bacterial levels in a similar incubator containing no eggs remained below 50 cells/ml for the duration of the trial.

Physio-chemical parameters remained stable throughout the period of experimentation; the level of dissolved oxygen in the water remained at or near saturation and the level of nitrogenous compounds was low (Table 5.4). Density of seed in the incubators varied between 14.1-30.0 g/l of water in terms

Table 5.5 Range of water flow rate and residence time in downwelling 20 l plastic vessels supplied with recirculated, slow-sand filtered water in the present trial with comparative data from other studies.

Seed Stage	RANGE			
stocked in incubator	Flow rate (1/min)	Residence time (min)		
1	5.6 - 10.3	2.9 - 1.6		
2	5.4 - 9.4	3.0 - 1.7		
2-3	5.7 - 10.2	2.8 - 1.6		
3	4.8 - 9.6	3.3 - 1.7		
1	0.2 - 1.2	3.8 - 0.6 (Rana, 1986)		
1	0.44	1.75 (Suliman, 1987)		
l		}		

Seed stages:

Stage 1, uneyed; Stage 2, eyed; Stage 3, imminent hatching; Stage 4, hatched

of total incubator volume. The 'working' volume in which eggs were actually moving was much smaller (approx. 2 1) and egg density was correspondingly higher (Table 5.2b).

Hatched fry required less water movement, therefore flow rates were reduced for this stage.

The effective residence time of water in the incubators averaged between 44 and 82 seconds (Table 5.5).

Table 5.6 Summary of Methodology and effectiveness of artificial incubation of Orgocoromis seed compared to natural incubation

· 	Hethods					
	To move eggs/embryos	To clean water	Hacchadility	Hotes	Sources	Connercial implications
Haturai	Respiratory movements and periodic back flushing in tuccal cavity	None but eggs may be cleaned by physical polishing in the mouth	bign	dependent on age/size of female; fertility rate of clutch	Fisheisen 1958 Varute and Jirge 1971, Rana 1986.	Loss in conditioning of proopers and increase in [S]
Artificial	Hone - (Static)	None (pond water)	yery poor	dependent on stage removed from female	Shaw and Aronson 1954	experiments carried out with very few eggs. Little applicability with larger numbers of eggs.
	Name - (Static)	Use of salt water (40% optimal)	nigh (44-87%)	O.macrocegnela	Shaw and Aronson 1954	
	None - (Static)	Use of bateriocides and fungicides use of formaldehyde sterile	poor 2 885° 2 825°	dipped at 0.1% for 10 mins emoryos dipped at 0.1% formaldebyde before placing in sterile water	Rana 1986	
	Aeration	Hone	8.8	mechanical injury causing post-hatching mortality - only successful (for seed) 60 hrs. post-fertilisation	Lee 1979 Nussaau & Chervinsky 1968	If eggs are few and/or females are synchromised such that the proportion of newly spawned eggs is small (i.e. seed removed) 48
	Mechanicai Shaxing Table	ippt. formalin/10 mins daily Daily removal of spoiled eggs, refill with freshwater if turnid	74-96%*	only good hatchability of eggs used 48 hrs. post fercilisation. Whole clutches sometimes lost.	tee 1979 Berrios Hermandez and Snow 1953, Siraj et al 1983. htboard & Prwginim 1975	ars post-fertilisation) may have limited application
		Recirculated water exchange Recirculated water exchange, Yater U.Y. treated	647s 40Zs	c	Suliman 1987 Suliman 1987	
	Water corrent	clean ground water, not recordulated	0.2.	upwelling, small bottles	Ridha ec al 1985	cleam flow through water may be unavailable and/or expensive
		Recirculated vater, unfiltered Recirculated water-pre- disinfection of eggs by dipping in antiseptics	≤ 628 75-868°	downwellings formalin, Malachite green acriflavin & buffadine over a range of doses	Subsingne 4 Sommerville 1985	- availability of antiseptics may constratin applicability recirculation systems have higher technical requirements
		recirculated water + U.Y. light (43556 susec/cm²) Recirculated water + U.Y.	z 90X*	^c st lower U.Y. dose, fungi ^c still viable, suggested bacteria are major cause		- u.y. light filters may be expensive or unavailable
		light (67112 pwsec/cm²)	ı	of mortality		- U.Y. light dependent on availa- bility of electricity
		Recirculated water U.V.light (6186;wsec/cm ²)	752*(602)* 922*(852)*	conical incubator/up-welling round bottomed incubator/ down-welling	Rana 1966	- incubator design and flow rate are important factors in reduc- ing mechanical trauma on eggs
		Recirculated vater, untiltered	24%* 54%* 42%*	flowrate 500L/min ^c flowrate 440L/min ^c flowrate 380L/min ^c	Suliman 1987	
		Recirculated water + U.Y. light (5188pwsec/cm²)	48% 85% 62%	flowrate 500 L/min ^c flowrate 440L/min ^c flowrate 380L/min ^c	Suliman 1987	
		Recirculated water + Charcoal filter Recirculated water + Charcoal filter + U.Y. light [30,000pwca+)	32-22% (88-9%) 91-66% (88-60%)	efficiency of both systems declined over 5 week period or operation	Don et al 1987	- maintenance of U.Y. systems is important for for high effectiveness

based on total number of eggs (i.e. including unrertilised), natural fertilised.
Dased on fertilised eggs to hatched larvae
Commediting incubator
Dased on fertilised eggs to swim-up fry
Dased on total number of eggs (artificially fertilised) to hatched larvae
Dased on total number of eggs (artificially fertilised) to swim-up fry.

5.3 Discussion

Successful artificial incubation of <u>Oreochromis</u> eggs must involve the reduction of levels of bacteria and fungi, and mechanical stress (Rana, 1986). In many incubation systems, these problems are often interrelated; methods that involve agitation of the eggs may damage the chorion, the protective outer layer of the egg, leading to the death of the egg by bacterial infection or disruption of the egg contents (Hempel, 1979). This problem is compounded by the nature of <u>Oreochromis</u> eggs, which are heavy and yolky and thus require a heavy flow rate to keep them buoyant (Fig.5.3).

Most workers have assumed that egg movement is important for successful incubation and have attempted to simulate the movements of natural mouthbrooding. Fishelson, 1966 (cited by Rana, 1986) suggested that the natural churning of eggs in the mouth helps to maintain the internal organisation of heavy lipids within the egg whereas Shaw and Aronson (1954) and Rana (1986) reported high hatchability of eggs after static, but sterile, incubation (Table 5.6). The maintenance of sterile conditions is likely to be practically difficult with large batches of seed which inevitably include unfertilised eggs. The control of bacterial and fungal infection in static water systems using various treatments has been described by Shaw and Aronson (1954) (Table 5.6) for very small numbers of eggs. Clumping and breakdown of infertile eggs correlated with increased levels of bacteria in down-welling incubators in this study and it is likely that the 'loss of whole batches'

reported by several workers (Lee, 1979; Peters, 1983) was related to seed that contained a high proportion of unfertilised eggs (Rothbard and Pruginin, 1975). It may also explain the improved results obtained when eggs are removed for artificial incubation 48 hours or more after fertilisation since by that time viable embryos probably have developed past the stage most sensitive to infection and infertile eggs have been lost from the buccal cavity.

The reduction in clutch size with duration of natural incubation (=stage of seed on harvest) observed in these studies indicates that eggs, many of which are probably unfertilised, are gradually lost during mouthbrooding. Presumably, differences in density of developing viable embryos and disintegrating infertile eggs allows separation and removal of the latter from the mouth. The same process allows removal of unfertilised eggs in incubators using water currents to move the eggs. The prior removal of unfertilised eggs during mouthbrooding also explains the difference found between the success rate of incubation for stage 1 eggs and other seed (stage 1 = 58%, other stages = 75%) in the present trials. Infertile eggs in the more developed seed (stage 2, 2-3 and 3) could be easily observed at the pond bankside but it was not possible distinguish to between recently spawned unfertilised batches. A slightly lower survival (P>0.05) for seed harvested as yolk-sac larvae may have been a result of the greater sensitivity of post-hatched seed to handling stress during harvest. Just-hatched fish larvae are particularly

sensitive to physical damage, yolk-sac of herring, plaice and salmon, for example, were found to suffer 50% mortality from skin damage on only 1% of the body surface (Hickey, 1978). On an experimental scale it may be possible to improve overall hatchability by incubating only viable eggs. However, the removal of infertile eggs after mass production of seed from natural spawnings is not possible unless each batch is viewed under a microscope.

Stripping of eggs and sperm from ripe broodfish, followed by artificial fertilisation, has been shown to result in very high fertility (mean 93.7%; Rana, 1986) but synchronising the ovulation of large enough numbers of females to make this costeffective has yet to be demonstrated. Rothbard and Pruginin (1975) observed that only fish that showed three 'on-heat' signs simultaneously could be stripped successfully.

The fertility rate of eggs after natural spawning is variable and appears to be dependent on several factors. Maternal age (Rana, 1986) and possibly size may be important. availability of sperm from the male, which is related to sex ratio and density effects is also likely to have an effect and other factors relating to the behaviour of fish within hierarchies of different stability are probably important (see above). Rana (1986) found that a mean of 6% of all egg batches taken from groups of females held in 1 m2 spawning tanks (sex ratio 4:1) were lost due to low fertility and that for broodfish over a range of age-classes mean fertility rate per clutch was only 66.5% (range 56.4% at age 6-9 months to 72.3%

at age 20 months).

Ideally, the design of incubators suitable for large numbers of eggs from natural spawning would allow for self-removal of unfertilised, decomposing seed that provide a medium for bacterial growth. This appears to be a major advantage of designs using water exchange to both move the viable eggs and remove the dead eggs (Table 5.6).

No relationship between seed density and survival was shown over the range of stocking density investigated, suggesting that provided, that bacterial levels can be kept below a critical value, very high densities are possible or even preferable. Shaw and Aronson (1954) suggested that churning causes mechanical injury to gram-negative bacteria and thereby protects the embryos from infection. The degree of contact, and thus removal of bacteria by physical polishing, between eggs would also be increased at higher egg densities but this must be countered by a necessary increase in flow rate to keep the eggs agitated. The flow rate of water required to agitate such densities of eggs may have detrimental effects hatchability and early fry mortality. Rana (1986) suggested that this explained the improved performance of down-welling, round-bottomed vessels compared to conical incubators receiving an up-flow of water in which eggs are in constant motion. In down-welling incubators, the inflow can be positioned such that eggs are gently banked before being moved again; and this more closely mirrors the pattern of natural churning in the mother's buccal cavity.

Suliman (1987) observed that the chief forms of mechanical trauma within a down-welling incubator were due to collision between egg and incubator and between eggs. Such collisions were five and two times more frequent respectively, at a flow rate higher than the optimum. The relative success of using higher densities of eggs in the wider, higher volume incubators used in the present experiment might be explained by the greater egg-to-egg but reduced egg-to-incubator contact compared to incubators with a lower surface-area to volume ratio.

In general, the maintenance of adequate water quality in terms of physio-chemical parameters has not proved to be a problem in flow-through systems (Subasinghe and Sommerville, 1985; Rana, 1986; Suliman, 1987) since nitrogenous wastes are mainly limited to the by-products of hatching. The same was found in this study.

In down-welling incubators in which mechanical trauma can be minimised, the level of bacteria and fungi appears to be the most important constraint to improved performance. Some workers have achieved good results using clean ground water which is then reused elsewhere in the culture system (Ridha et al., 1985; Table 5.6) but more normally water is recirculated. This can quickly lead to a build-up in the level of microorganisms responsible for egg loss.

Hatchability and larval survival are dependent on both flow rate and ambient bacterial levels (Table 5.6, Suliman, 1987) but flow rates alone do not adequately describe conditions in

incubation systems. The residence time of water and flow rate per unit weight of seed are also necessary to adequately assess the incubation environment. The flow rates used in this study varied with both time, density and stage of seed initially stocked, but were much higher than other studies (4800-10300 ml/min; Table 5.5) in keeping with the higher volume and numbers of seed used.

Flow rate was reduced with time in accordance with the observations of Lee (1979), showing that the frequency of churning during mouth brooding decreases as incubation proceeds; Rana (1986) related this to a progressive thinning of the chorion as hatching is approached.

When the spawning period is deliberately shortened to decrease mean ISI of breeders, and increase productivity, incubation systems designed to reduce mechanical trauma and maintain high bacteriological water quality continually are required. The technical performance of properly maintained UV light filters is high (Kimura et al., 1976; Table 5.6) but they are expensive (UK price US\$ 0.12/litre of water filtered/hour) and in many places units or replacement tubes are unavailable. The efficiency of the filter can also decline significantly with time as a result of slime and mineral deposits (Don et al., 1987; Table 5.6). Membrane filters have become available (Brock, 1983) and have been used for shellfish seed incubation (Garland et al., 1986) but have yet to show uniform and reliable results.

Slow sand filters have long been used for the production of

potable water of high bacteriological quality and although used throughout the world have particular advantages for developing countries in the tropics (Skeat, 1961; Ellis, 1987). Their inherent advantages include extremely low maintenance requirements and, under most conditions, independence from imported materials or equipment. Capital cost can be relatively high (local cost US\$ 0.26/litre of water filtered/hour) but they have effectively no maintenance costs and construction is simple and unsophisticated. The efficacy of the filter in these experiments was such that levels of bacteria were kept low in the supply water at all times (<50 cells/ml) in the incubator cycle studied and only climbed to higher levels $(>3x10^3/m1)$ within a single incubator for a brief period during the breakdown of large numbers of unfertilised eggs. Suliman (1987) reported that total viable bacterial cells were never lower than 5 x 10^3 cells/ml in an unfiltered incubation system.

Table 5.7 Summary of estimated swim-up fry production from experimental treatments in concrete tanks, nylon hapas-in-ponds and earthen ponds³

Treat- ment	Female stocking density (kg/m²)	Mean swim-up fry output (fry/m²/day)	Mean female productivity (fry/kg fem ¹ /month)	Mean swim-up fry output/ system/ day
T112	0.64-1.27	106	3344	1332
T122	0.86-0.96	118	1941	1483
$T13^2$	0.84-1.48	14	361	176
T142	0.68-1.06	132	3034	1659
T21	0.66-0.77	142	2152	1790
T22	0.57-0.87	185	2589	2322
T23	0.56-0.80	167	2524	2096
T24	0.55-0.89	172	2412	2160
H11	0.20-0.20	58	3322	2318
H12	0.18-0.24	71	3171	2835
H13	0.21-0.21	64	3108	2560
H14	0.18-0.25	59	3021	2364
T31	0.19-0.60	168	4523	2106
T32	0.21-0.69	185	4079	2329
T33	0.27-0.80	224	4138	2818
T34	0.31-0.96	156	2398	1963
H21	0.04-0.20	45	3815	1783
H22	0.06-0.27	53	3330	2133
H23	0.06-0.32	63	3430	2513
H24	0.08-0.37	65	2906	2593
Н31	0.05-0.12	46	5215	1834
H32	0.05-0.12	50	4145	1993
H33	0.05-0.13	40	2636	1595
H34	0.05-0.13	65	8463	2592
H35	0.06-0.12	61	7039	2421
P12	0.02-0.03	2.6	3351	4524
P13	0.02-0.03	1.5	2022	2610
P22	0.03-0.03	2.3	2328	4002
P23	0.03-0.04	6.2	5077	10788

All treatments, except P12-P23, harvested as eggs and yolk-sac fry Swim-up fry numbers estimated from mean survival after incubation losses

^{1 (}Mean stocking weight + mean harvest weight)/2 of all females used in treatment.

² Includes 10 day and 5 day harvest period

³ Concrete tanks $(A = 12.57 \text{ m}^2)$; nylon hapas $(A = 40 \text{ m}^2)$ and earthen ponds $(A = 1740 \text{ m}^2)$

5.4 General discussion of systems investigated

Seed production and broodstock productivity

A trend towards intensification from ponds, to hapas, to tanks is shown clearly from the estimated swim-up fry yields of each system. This is broadly in agreement with the results of other workers (Table 5.7) although management and unit size may have a greater effect on yields. The broodstock density (0.22 kg/m^2) and fry yields $(9-16 \text{ fry/m}^2/\text{day})$ in static water tanks in the Philippines (Guerrero and Guerrero, 1985) were more similar to that found for earthen ponds $(0.04 \text{ kg/m}^2, 1.5-6 \text{ fry/m}^2/\text{day})$ than most of the tank treatments in this study $(1.5 \text{ kg/m}^2, 100-250 \text{ fry/m}^2/\text{day})$. This undoubtedly reflects the efficient management, especially with respect to seed removal, broodfish condition and water quality, that is possible in a small, easily drainable system.

The importance of management is indicated by the very low yields in the tank treatment removing swim-up fry (T13). These are in the range recorded for the Baobab arena (Balarin and Haller, 1982) and earthen ponds in this study. It appears that high production from tanks requires a high degree of management: broodstock exchange, water clarity and stocking density are important not only in promoting spawning activity but also improving synchrony of breeding and individual broodfish productivity. The yields from the Baobab arena suggest that potential economies of scale with larger tank systems may be difficult to attain, especially when the breeding arena is also used for conditioning, spawning and

incubation. However the response to intensified management is also shown in smaller systems. Seed yields from tanks within a recirculated water system in which broodfish were held continuously in small tanks and seed harvested every 14 days $(78.3 \text{ fry/m}^2/\text{day}; \text{ Balarin and Haller ,1982})$ were also far lower than those recorded in the present study utilising a shorter interharvest interval plus broodstock exchange.

Hapas produced high yields of fry per unit area, and enhanced female productivity compared to tanks in the present study. Exchange of broodfish in spawning hapas has improved productivity in other studies (Lovshin and Ibrahim, 1989), but production and female productivity (45-52 seed/m²/day and 2673-3177 seed/kg female/month) were lower than for the present study, especially as yields were estimated in terms of fry rather than seed of mixed stages (45-71 fry/m²/day and 2906-8463 fry/kg female/month). Female productivity was high despite individual broodfish being in the spawning net for only one third of the time. This confirms that conditioning is an effective strategy for improving the synchrony of spawning, and if the interharvest interval is also reduced, for increasing productivity (fry/m²/day).

The lower production reported by Lovshin and Ibrahim (1989) probably reflected the longer Interharvest Interval which allowed an extended period of natural incubation and subsequent considerable loss of condition. Improved yield with increased frequency of harvest in hapas has recently been confirmed by Vergedem amd McGinty (1987). An increase of nearly 50% in the

number of total spawns after harvesting every 4 days rather than every 10 days was recorded over a 5 week period. However, these workers suggested that weekly harvests were more cost effective, particularly as a greater proportion of the seed collected was more advanced and easier to incubate.

The importance of incubation as a constraint to more frequent harvesting has been shown by other studies. Hughes and Behrends (1983) achieved very high production in hapas (73.4 seed/ m^2 /day) over a 73 day experimental period, but a high proportion of the seed was eggs and the highest swim-up fry production was much lower (29 fry/ m^2 /day) than that achieved in the present study.

The response of intensive management, in particular the conditioning of broodstock and frequent harvest of swim-up fry, in earthen ponds, indicated both the major constraints and potential of such a system. The separation of conditioning and spawning, such as in Treatments P22, P23, and large ponds in Israel (Rothbard et al., 1983; Table 5.7) can be effective in enhancing synchrony of spawning and producing high quantities of fry. If the period of production is prolonged, however, mean productivity is drastically reduced. This poor 'sustainability' of production is related to a lack of control of fish density. Unharvested fry soon become recruits and increase fish density above the optimal for spawning, with consequent negative effects on water quality and broodfish condition. In contrast, tank and hapa production show greater potential for sustainable yields, especially the former, as complete and frequent harvest of seed can complement optimal water quality management. In

Table 5.8 Indicators of system efficiency for experimental treatments in concrete tanks, nylon hapas-in-ponds and earthen ponds

Trea ment	t- Net broodstock biomass change (kg/day) ¹	By-product yield ² (kg)	Swim-up fry³ as % total net yield	System FCR ⁴	Fry output/ kg feed/ day
T11	0.12	0.0	9.5	2.2	29.3
T12	0.12	0.0	10.8	3.2	22.3
T13	0.16	0.0	5.7	1.9	3.8
T14	0.16	0.0	8.8	2.1	29.3
T21	0.16	0.0	6.7	2.5	35.3
T22	0.16	0.0	7.2	2.5	47.1
T23	0.16	0.0	8.5	2.5	41.2
T24	0.16	0.0	7.2	2.4	44.1
H11	-0.04	0.0			52.9
H12	-0.02	0.0			64.7
H13	-0.04	0.0			67.6
H14	-0.02	0.0			58.8
Т31	0.10	0.0	11.6	2.7	33.3
T32	0.13	0.0	9.9	2.7	31.3
T33	0.15	0.0	10.6	2.7	31.8
T34	0.18	0.0	6.1	2.7	17.9
H21	0.10	0.0	8.7	2.7	31.3
H22	0.16	0.0	6.4	1.8	32.8
H23	0.18	0.0	7.2	1.7	34.8
H24	0.21	0.0	6.4	1.5	35.3
Н31	-0.00	0.0			67.0
H32	-0.00	0.0			59.4
H33	0.00	0.0			39.6
H34	-0.00	0.0			96.2
Н35	-0.06	0.0			81.1
P12	0.57	156.6	4.0	0.2	128.4
P13	0.54	129.5	1.7	0.2	78.4
P22	0.19	127.5	3.5	0.2	139.2
P23	0.38	134.5	7.8	0.2	367.6

¹ negative figures indicate loss of broodfish during handling and/or from cage

Concrete tanks (A = 12.57 m^2); nylon hapas (A = 40 m^2) and earthen ponds (A = 1740 m^2)

² by-products include recruits at harvest (large tilapia other than stocked broodfish), larger fry and wild species

³ swim-up fry suitable for hormone treatment

⁴ feed input given to appetite except pond treatments includes 10 day and 5 day harvest period

practice, poor water quality may depress fry production both in ponds as shown in this study, H21-24, H31-35 and in larger water resources (e.g. Laguna de Bay; Bautista et al., 1988). Recruits can be controlled in the pond environment if a piscivorous fish species such as snakehead (<u>Channa striata</u>) is stocked ($1/100 \text{ m}^2$; Balasuriya, 1988).

The range of different by-products from earthen ponds geared to produce swim-up fry suitable for sex-reversal distinguish these systems from intensive tanks and hapas (Table 5.8). In hapas and tanks in which swim-up fry are harvested daily 'left-over' fry are removed irregularly or at final harvest (Guerrerro and Garcia, 1983; Uchida and King, 1962).

Recruits may reduce broodstock productivity but they are a byproduct, as human food or as replacement broodfish under some
circumstances. Larger fry, unsuitable for hormone treatment,
may be sold as normal tilapia fry after removal by grading but
they are of minor consequence. Earthen ponds are also
susceptible to contamination with 'weed' species such as small
prawns and competitive or predatory species of wild fish,
insects, and birds. Insects must be removed, sometimes at high
labour cost, from the fry before hormone treatment. Wading and
diving birds e.g. egrets, herons may also be important
consumers of swim-up fry; this loss is avoided if seed are
removed as eggs and yolk-sac larvae.

The biomass gain of broodstock during fry production is another indicator of system 'efficiency' (Table 5.8). The experiments in which this aspect of production was analyzed (T1, H3, P1)

revealed that high biomass gain was often negatively correlated with mean broodfish productivity (e.g. T13, H33, P12, P13). Non-spawning fish in concrete tanks were found to be larger than either moderately spawning or frequently spawning fish (T21-T24).

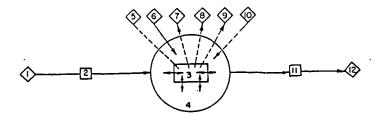
The broodstock weight gain during periods of poor water quality and low seed output in hapas (H21-H24) further suggested that rapid growth in broodfish can be indicative of low reproductive output. This is a strategy that may allow tilapias to optimise survival under fluctuating environmental conditions (see 3.2.4.). However results from other trials are contradictory. Santiago et al. (1985) found that a broodfish diet containing a high level of crude protein (40% rather than 20%) optimised growth and fry production in concrete tanks and hapas.

The systems tested in the present experiments showed a large variation in feed consumption (Table 5.8). The natural feed available in ponds increased the efficiency of pelleted feed use (tanks 4-47 fry/kg feed/day; hapas 31-96 fry/kg feed/day; ponds 78-368 fry/kg feed/day respectively) and considerably improved system mean FCR (tanks, 2.5; hapas, 1.9; ponds, 0.2). In situations where high quality pelleted feeds are unavailable or expensive their use as supplements in pond fry production, rather than complete feeds in intensive systems, could be advantageous. The necessity of using high quality feeds to maintain tilapia broodstock productivity in clearwater systems has been reported from several sources (Roberts, 1982; Tuan, 1986) but a variety of feeding strategies have been used in

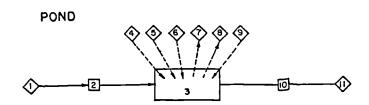
Fig. 5.3

A flow diagram of major system processes for <u>Oreochromis</u> seed production in recirculated water supplied tanks, static hapa-in-pond and earthen pond systems

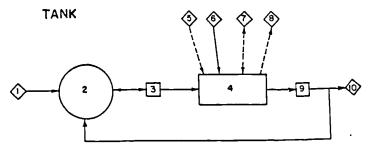
HAPA



- I. AIT water supply canal
- 2. Concrete supply channel
- 3. Nylon hapa
- 4. Earth pond
- 5. Feed input 6. Make up water
- 7. Broodstock
- 8. Eggs/yolk sac fry
- 9. Recruits from wild breedin;
- IO. Fertiliser
- II. Drainage channel
- 12. Outfall



- I. AIT water supply canal
- 2. Concrete supply channel
- 3. Earthpond
- 4. Feed input
- 5. Make-up water
- 6. Broodstock
- 7. Swim-up fry
- 8. Recruits
- 9. Fertiliser
- IO. Drainage channel
- II. Outfall



- .1. AIT deep well water supply
- 2. Settling tank and biofilter
- 3. Head/storage tanks
- 4. Concrete spawning tank
- 5. Feed input
- 6. Nake up water
- 7. Broodstock
- 8. Eggs/yolk-sec fry
- 9. Collecting channel
- 10 . Outfall

LEGEND

A rearing unit

A unit of process that changes water quality so as to make it significantly more or less suitable for rearing organisms within the system

A process necessary to the system but has no significant direct effect on water quality

Input of output from the system

--- Movement of water or other liquid

--- Other types of transfers

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systems in which natural feed is important. Pelleting a high quality feed (35% crude protein) did not improve fry production in <u>O. aureus</u> compared to its use as a powder (Allison et al., 1979) and Guerrero and Garcia (1983) reported that the improvement in fry yields in hapas associated with feeding a high quality pellet rather than rice bran were only apparent when natural feed availability was low. Fry producers using ponds in Taiwan reduce feeding rate to only 0.3-0.5% body weight during breeding to maintain good water quality and save feed costs (Liao and Chen, 1983).

However, all of these systems reported female productivities lower than in the present experiments in which broodfish were fed to appetite with a high quality floating pellet.

System Management

intensification of <u>Oreochromis</u> breeding has The several implications for management. The harvest of eggs and yolksac larvae after a short interharvest interval, the maintenance of exchange broodfish in separate groups, and incubation of seed, increases the required level of management. Access electricity facilitates incubation and refrigeration hormone-treated feed but is not absolutely necessary for any of the swim-up fry production systems investigated.

Intensification and selection of pond hapa or tank methods has consequences for water use and reuse (Fig 5.3) and requirements for fertilisers and feeds.

Both Snow et al. (1983) and Verdegem and McGinty (in press) in attempting to intensify O. niloticus fry production, found that

the greater cost of frequent seed harvest was not compensated sufficiently by higher yields over periods of twelve and six weeks respectively. The higher production achieved by intensification does not necessarily mean higher profitability. Sarig (1988) observed that the profitability of Israeli fish farms was related to management rather than yield (t/ha/y). Management and economics of the systems examined are addressed more fully in Chapter 6.

CHAPTER 6

ECONOMIC ANALYSIS OF FRY PRODUCTION

6.1 Introduction

Fry, or seed fish, are one of the major inputs of a grow-out system, accounting for between 28-40% of the total production costs for herbivorous species (Shang, 1981). Hence, the economics of the grown-out product strongly affect the viability of fish seed operations. If the profitability of the grown-out product drops, demand for fish seed will inevitably decline, forcing fry producers to sell at lower prices. Thus, fish seed operations are influenced by the stability of the grow-out production system.

An example of this dependency is the recent decline of smallscale tilapia fry production in the Philippines following the collapse of many of the grow-out cage operations in Laguna de Bay (Guerrero pers. comm., 1989). Another example is the recent halving in the price of shrimp post-larvae in Thailand followed a drastic decline in consumption of tiger shrimp (Peneaus monodon) in Japan, the main export market for the grown-out product, following overproduction in S.E. Asia and the demise of the Emperor of Japan. A third example is red tilapia $(0, niloticus \times 0, mossambicus hybrid)$ in Thailand. The fish resembles the high value marine red snapper (Lutjanus spp.) and initially the price of broodfish rose to a level well above that of normal Nile tilapia as hatcheries sought to start fry production. However, consumer acceptance of the unknown fish was low, causing broodfish prices to fall below that of normal tilapia as hatcheries realised that there was an undeveloped market for the new fish.

The introduction of MT-treated tilapia into Thailand is not marketing difficulties to meet such since the appearance of the fish is similar to that produced with conventional methods. Indeed, MT-treated fish are more likely to have the lighter colour and heavier rounded body favoured by the market in both the Philippines (Torres and Navera, 1985; Escover et al., 1985) and in Thailand. Furthermore, the demand for tilapia fry is expected to increase on the basis of a continuing expansion of extensive and semi-intensive culture systems, particularly in Northeast Thailand (Fedoruk, 1986). In other tilapia producing countries in Asia, e.g. Taiwan, Malaysia and the Philippines, hormone treated fish have been readily accepted.

In Taiwan, tilapia culture has overtaken both carp and milkfish production to be the single-most important species of cultured fish (Kuo, 1984). Most fish are sex-reversed and high yields are obtained using a combination of integration with livestock and pelleted feeds (Liao and Chen, 1983). Tilapia are often raised in Thailand in polyculture with carps for which there is a well-developed market. Consequently, price-cost relationships for carps affect the demand for tilapia fry. Thus, the economics of fry production are likely to be quite different in Thailand compared to Taiwan and the Philippines. In Taiwan the production of sex-reversed tilapia fry is a specialised business, whereas in Thailand hatcheries commonly produce both carp and tilapia. The profitability of carp production can therefore be an important factor in the economic analysis of tilapia fry production.

Table 6.1 Number of tilapia and carp seed produced annually per unit weight of broodstock raised (seed kg-1) and unit area of nursery pond (seed m-2) in 3 hatcheries in Northeast Thailand

Site	Seed per kg broodstockb		Seed per m² nu	ursery pond
	Tilapia Carps ^c		Tilapia	Carpsc
Ubon	750	2850	12.5	285
Surin	222	1724	50.0	319
Udorn	333	2500	27.5	239

a Spawning seasonal

b Total broodfish on-farm (i.e. including fish held but not spawned)

^c Carps; mixture of common carp (<u>Cyprinus carpio</u>), Silver barb (<u>Puntius gonionotus</u>), Rohu (<u>Labeo rohita</u>) and small numbers of Chinese carp (Udorn & Surin only)

d Adapted after Little, Skladany and Rode (1987).

The present status of tilapia culture in Thailand

Nile tilapia is the single-most important freshwater cultured fish by volume and fourth by value in Thailand (18,366 t/year, estimated value Baht 22 million (\$0.88 million); Department of Fisheries, 1988). Between 1982 and 1986 production more than doubled and, on the national level, exceeded the volume and value of all the common freshwater carps (common carp, silver barb, Indian major carps and Chinese carps) combined. In Northeast Thailand, carps are relatively more popular, but tilapia still constitute an estimated 28% of the total volume of 5,800 t of cultured freshwater fish/yr in 1986 (Department of Fisheries, 1988). Fry production from all Government fishery stations nationwide was estimated at 20 million fish (Office of Agricultural Economics, 1986) but large numbers of even-sized fry are produced by large, irrigated commercial hatcheries located in Central Thailand, some of which are exported to Northeast Thailand. Tilapia fry are also produced in smallscale hatcheries within the Northeast but these operations tend to be small and without running water; carp fry are currently produced in much greater numbers as their breeding is more efficient under these conditions (Table 6.1; Little, Skladany and Rode, 1987). In general, current methods of tilapia fry production in Thailand have low productivity and produce seed of age and size unsuitable for sex-reversal. In both Taiwan and the Philippines, earthen pond spawning of tilapia remains the most common form of fry production by both small and largerscale farmers. The management of earthen ponds for tilapia fry



predation losses decrease in decrease in cannibalsm losses Fry unsuitable for sex Fry suitable for sex reversal reversal N d d Let potential for Seperation of breeding 8 conditioning In same system Breeding B conditioning Lakes, reservoirs, ponds Ponds/hapas/tanks Ponds / tanks Aquaria, small tanks System Ponds Hapas tanks Ponds (e) Swim-up fry production(1) continual
(2) batch (c) Mixed age / size fry production (f) Removal of eggs B yolk-sac fry commercial scale (g) Removal of just-spawned eggs laboratory scale (d) Even-sized fry production (b) Byproduct of culture Method (a) Natural system

Strategies used for the production of Oreochromis spp. fry

production is usually intensive (see Fig. 6.1) and provides fry of a uniform quality suitable for hormonal sex-reversal.

Northeast and Central Thailand are the most important areas of freshwater fish culture in the country. The Department of Fisheries has put most effort recently into aquaculture promotion in Northeast Thailand but Central Thailand, because of extensive infrastructure, well developed farms and irrigation networks, has greater potential for short-term gains in fish production (Asian Development Bank, 1985). An economic analysis of improved methods of tilapia fry production is warranted for these two areas of importance.

This chapter attempts to place the technical results obtained in the series of experiments described above in an economic context. An economic analysis of the production methods developed on the AIT campus is presented, together with an extrapolation of the effect of their introduction into fry production farms in two main fry production areas of Thailand.

Fig. 6.2

The location of the hatcheries modelled for production of MT-treated tilapia in Central and Northeast Thailand

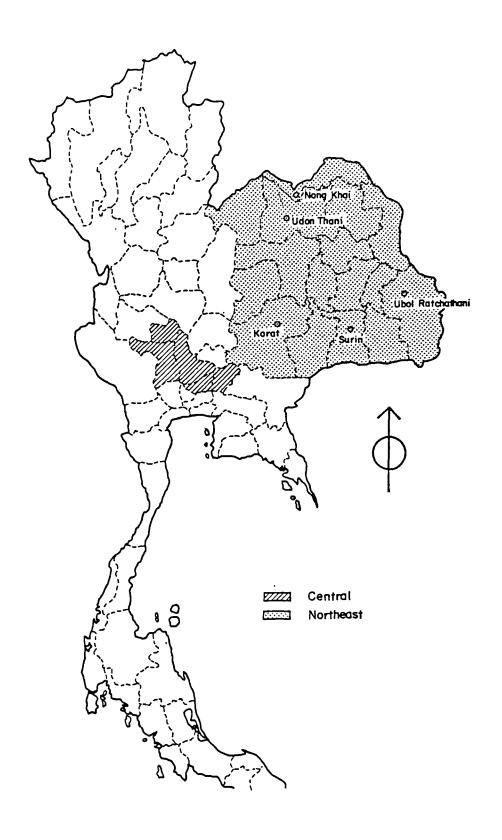


Table 6.2 Production of the common, herbivorous fish species in Central and Northeast Thailand (DOF, 1988)

G	Percentage		
Species	Central	Northeast	
Oreochromis niloticus Cyprinus carpio Puntius gonionotus Chinese carps Labeo rohita Cirrhina mrigala	60.4 < 1.0 29.0 7.3 3.1 0.0	30.9 26.2 35.4 1.2 4.4 1.8	
Total amount (t)	7612	5369	

6.2 Methods and assumptions of analysis

6.2.1 The strategies

6.2.1.1 The production of swim-up fry for hormone treatment

The technical data used were obtained primarily from on-campus experimentation. Price-cost relationships and production constraints obtained from selected farmers were added to construct a model for the commercial application of MT-treated fish production for a start-up operation based in an irrigated area of Central Thailand and the introduction of these methods into five existing fry production, mainly carp, operations in Northeast Thailand (Fig. 6.2).

The modelling of a start-up operation in Central Thailand is based on a combination of factors. Tilapias are the most important single fish species, comprising over 60% of the total production of herbivorous fish in this part of Thailand, whereas, in the Northeast the carps are predominant (Table 6.2). Fish seed production tends to be more specialised, in terms of species and stage of production i.e. spawning and nursing are often separate operations. Fry production in the private sector is usually a family business; the extended specialisation family structure has tended towards production by different members of the same family and joint marketing. It is assumed therefore that hormone treated tilapia fry would not substitute for carp, but rather would be produced as a start-up operation on new land by members of an extended family already involved in the hatchery business. Current

Table 6.3 Comparison of size and productivity of five hatcheries in Northeast Thailand

	Udorn	Nongkhai	Surin	Ubon	Korat
Area (ha)	2.9	2.1	0.9	0.2	0.5
Irrigation	no	yes	no	yes	no
Production (No. of seed x 1000)	2800	3318	2000	300	833
Productivity (seed x 1000/m²)	966	1565	2273	1167	1602
Net Revenue (Bt/1000 seed)	30	43	24	121	45

Source: Little et al., 1986

production practices tend to produce even size fry in production cycles of 30-60 days (Siripat et al., 1984), but daily removal of swim-up fry and separate nursery (Fig. 6.1) is common around Bangkok (Sensrimahachai, 1979).

In Northeast Thailand, hatcheries are generally dominated by carps and production and marketing is less specialised. Typically low yields of uneven-sized fry of tilapia are produced (Little et al., 1987). The addition or substitution of hormone treated fry production into such hatcheries was therefore modelled.

The period of production was limited to six months of the year because the purchase of fry is mainly limited to the wet season. Additional investment may be reduced by using present facilities and a reduction in the scale of production.

Integration of intensive tilapia fry production into the current seed operations of five different hatcheries in Northeast Thailand producing mainly carp was simulated. All the farms were treated as case studies and were not necessarily typical of other hatcheries in the same region (Table 6.3). Nevertheless, although the farms were all small-scale, there was still a large degree of variation in terms of production, and hence returns to land and labour. Land area varied by a factor of more than ten; production per unit area by more than two times, and net revenue per 1000 seed produced by up to four times. In addition, irrigation was not present on every farm.

Experimentation on campus provided a range of both production and cost strategies for mass-fry production. The unit size of

Table 6.4 Nomenclature of economic models for Northeast and Central Thailand using experimental treatments as basis for analysis

Economic strategy	Experimental treatment	Number of production units modelled		Opportunity cost considered
		Central **	Northeast*	Constacted
T11-T34	T11-T34	8	8	yes
н11-н35	Н11-Н35	18	18	yes
P12-P23	P12-P23	2	2	yes
h11a-h35a	н11-н35	_	9	yes
h11b-h35b	н11-н35	-	9	no
}	!			

Period of production (months/year)

^{** 12;} start-up operation

^{* 6:} small-scale carp operations

each of the main methods, pond, hapa-in-pond and tank was designed to make maximum use of the economies of scale within the constraints of capital, land and labour of typical fry producers.

The three basic methods compared represent a synthesis of adapted commercial methods and scaled-up laboratory-based technology (Table 6.4).

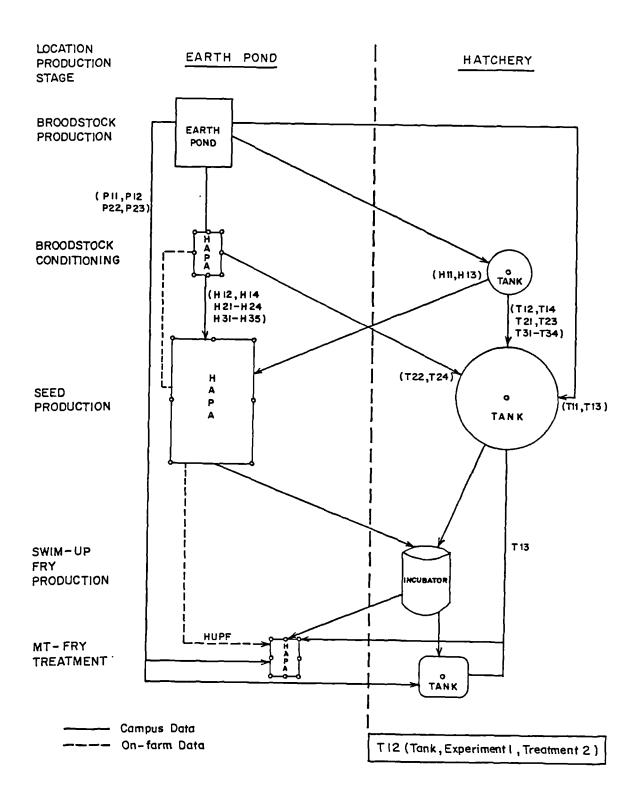
The major difference in variable costs between a start-up operation in Central Thailand, an area dominated by irrigated rice monoculture (Edwards et al., 1983), and on-going hatcheries in Northeast Thailand is the opportunity cost of land, in particular pond space. Whereas land rental, based on the gross margin of rice production, is low in Central Thailand for all of the treatments, it is of major importance in Northeast Thailand carp hatcheries in which the opportunity cost includes the lost profit from carp fry production.

However, if no opportunity cost were assumed for carp pond substitution (h11b-h35b) production is additive to present carp fry production. Most of the farms surveyed in a study of small hatcheries in Northeast Thailand (Little et al., 1987) had a high ratio of broodstock to nursery pond area, and broodstock were not maintained intensively. Seed production from tilapias held in hapas suspended in ponds used for spent carp broodstock is therefore unlikely to have a negative impact on carp fry production.

Feed and broodstock costs are higher in Northeast Thailand than in Central Thailand. Broodfish, even of small size, have a high

Fig. 6.3

Schema showing the strategies for producing MT-treated tilapia fry in Central Thailand



opportunity cost in Northeast Thailand. Pelleted feed costs are, on average, 15% higher and opportunity cost of labour 30% lower in Northeast than Central Thailand.

A framework of possible strategies to produce swim-up fry, and consequently, MT-treated fry is given in Fig. 6.3. Methods can be based on earthen ponds or concrete tank systems or a combination of both.

All strategies were designed as suitable for family-run operations because successful fish seed producers in Thailand are typically small-scale operators employing some off-farm labour. In a study of six small-scale hatchery operations in Northeast Thailand, net farm income was found to be very high from Bt 35,173/ha to Bt 242,256/ha (Little et al., 1987), especially when compared to average cash income levels in Northeast Thailand of Bt 2,551/ha (Office of Agricultural Economics, 1988). The strategies and optimum size of systems analyzed were therefore selected with regard to the labour constraints of a family-run business. Thus, of the two major methods used for obtaining swim-up fry from earthen ponds, the daily harvesting of fry from the pond margin, as practised in Taiwan and the Philippines, was used as the strategy more likely to be compatible with the type of labour available on a family-run farm.

The earthen pond strategy which use small sized broodfish (P23) may have undesirable consequences for the quality of the fry produced which is ultimately reflected by reduced value. The relationship between heritability of increased reproductive

output, and reduced body growth in progeny is not yet known for tilapia. It is possible that hormone treatment would ameliorate this negative effect and this is assumed for the purpose of this analysis.

Assumptions regarding production costs are given in Appendix 3. A recirculation system using biological filtration in a tropical environment is assumed to require a good quality roof to reduce eutrophication which might otherwise occur affecting the efficiency of the filter and the maintenance of water quality.

Other assumptions inherent in modelling the substitution of fry production strategies in Northeast Thailand carp hatcheries are the same as for a full start-up operation in Central Thailand, with the following differences:

(a) Additional strategies in Northeast Thailand hatcheries Hapa-in-pond production was also modelled using a reduced number of hapa units (9) over the same range of management treatments in only one pond (h11a-h35a). These options sought to reduce the cost of hapa strategies; the flexibility of an extra pond for temporary use, as in the start-up operation in Central Thailand, was not required as the current carp fry production necessitates a larger number of ponds (Little et al., 1986). The same strategies with fewer hapas were also modelled for a zero pond opportunity cost (h11b-h35b). This assumed that spent carp broodstock could be stocked in ponds with tilapia spawning hapas with no detrimental effect on carp

fry production. No pond opportunity cost was therefore included in these strategies since the effect on present carp production by tilapia fry production was assumed to be negligible.

In addition, the fry production and costs from a hatchery currently producing swim-up fry in large hapas in Northeast Thailand were included for comparative reasons (HUPF). This farm, using broodstock obtained from AIT, harvests swim-up fry after natural brooding from females in large nylon hapas identical to those used in the AIT trials.

(b) Period of production

The production and marketing of fish fry in Northeast Thailand is seasonal. Most fish are stocked in the early part of the monsoon season in the region and the season for marketing fry peaks over the 3 month period of the early rains (Department of Fisheries, 1988). Carp fry production is limited by ripeness and maturity of broodstock, and by market demand.

6.2.1.2 Hormone treatment to produce MT-fry

Tank system

The shallow concrete tank used for hormone treatment is based on the design used by commercial farms in Taiwan. Optimisation of system design and tank number was made on the basis of pumping capacity; a 24-tank system of 144,000 fry a month capacity can be supplied from a single submersible pump. The design incorporates a biological filter which maintains water quality to a level adequate for a 30-day culture period, after which the filter must be cleaned and water refilled (du Feu,

1987), in contrast to the commercial system in which water was recirculated through a large fish pond.

The use of shallow tanks for the AIT model also required an estimation of land, construction fill and roofing costs. Commercial farms in Taiwan were observed to use both covered and uncovered tanks. Experience at AIT indicated that direct exposure to sunlight considerably increased growth of epiphytic algae on the tank sides which are continually cropped by the fish (du Feu, 1987). A simple thatch shade was used to moderate exposure to sunlight and rain.

Hapa system

The sex-reversal of swim-up fry in nylon hapas has been proposed by both Buddle (1984) and Guerrero et al. (1985) on the basis of reduced cost and flexibility of use.

The hapa-in-pond treatment method required extra pond area for placing hapas, separate from that required for holding or spawning broodstock (see above), but no extra construction fill or roofing.

Five hapas (3 m \times 1.8 m \times 0.5 m) are required for a monthly output of 144,000 fry.

Tank and hapa systems

Any system for hormone treatment of swim-up fry requires a capacity to absorb fry numbers in excess of average monthly output since fry production shows variation with time. The number of treatment units required per strategy was calculated on the basis of a 25% excess over the mean monthly output. The quality of MT-treated fry is assumed to be as high in hapas as

in tanks.

Theoretically, as fry are more isolated from disease organisms, and water quality is more homogeneous in a closed recirculated water system, results should be more predictable. Several authors have reported poor or variable survival during intensive nursing and/or sex-reversal of Oreochromis spp. in clear water, intensive systems. Buddle (1984) and Rothbard et al. (1983) reported low survival during sex-reversal in tanks in Israel (mean 50%; range 17-96%), principally due to high levels of parasitic infection. New et al. (1984) improved survival (S%) of O. spiluris (S%=54%, 3 feeds daily) with frequent (5 times per day) or continuous feeding (S%=60-75%). Survival in hapas has generally been better. Berger and Rothbard (1987) reported survival rates of between 65-85% for red tilapia fed hormone treated and untreated diets in cages. Fry in similar cages but feeding only on natural feed suffered 50% mortality. Nacario (1987) recorded survival rates between 56-83% over a range of stocking densities and Guerrerro et al. (1985) obtained survival in excess of 90% in both cages and tanks. As hapa systems appear to be superior to clear water tank systems in terms of survival and average final growth, but more prone to occasional near total losses (e.g. net failure, water quality, excessive bird predation) than tanks, survival estimated at 60 % for both systems.

High levels of natural food may make sex reversal more unpredictable in both tanks and cages. Feeding rates were higher for tanks compared to hapas, but hormone dose and costs

were lower, based on data from AIT trials (6.3.2). The required hormone level was calculated at 60 mg/kg feed and 40 mg/kg feed for hapas and tanks respectively. These levels of hormone consistently produced a high percentage of males. In both tank and hapa systems, labour was costed for feeding 5 times per day; the larger number of units required to hormone treat equivalent numbers of fry thus required a much higher labour input for tank system.

The following were assumed to be necessary management practices for successful results in systems with or without natural feed:

(a) frequent feeding (>5 times/day), (b) the use of a diet of high appetency and (c) a high fry stocking density (12 fry/1).

6.2.1.3 The use of MT-fry for grow-out

The purchase of fry by farmers for grow-out is normally a major cost in aquaculture but in Central Thailand relatively few farmers breed their own fish (1.7% in Pathum Thani, Edwards et al., 1983); the cost of fry amounted to an estimated 15% of production costs on a farm producing mainly tilapia and carps in Supanburi, Central Thailand (Siripat et al., 1984). Culture of the Nile tilapia has enabled the farmer to minimise costs by reuse of his own seed; tilapia are thus in many ways more comparable to rice or maize as a crop than are the other common species of cultured fish which do not breed easily in the pond and require regular purchase of seed. This factor might have a negative effect on the demand for tilapia fry and the economics

of fry production, particularly with regard to the price of the new (MT-treated) product. On the other hand, grow-out producers gain from using MT-treated fry through a reduction in yield variation because relatively more fish are produced at a size which is favoured by the market. MT-treated fish have an intrinsically faster individual growth rate (Macintosh et al., 1989) and control of breeding in grow-out monocultures produced a more uniform marketable crop of fish. The stocking of MT-fish alone or in a polyculture with freshwater carp is therefore more predictable in terms of individual fish size achievable.

6.2.2 Analysis

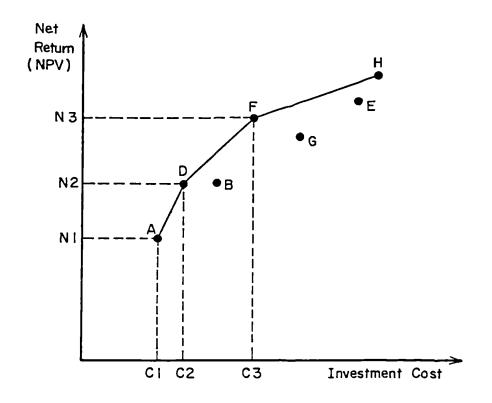
6.2.2.1 Framework of methodology

The various methods of swim-up fry production for MT treatment require different levels of capital investment. In this analysis, capital requirements were dependent on the seed production system, the requirement for incubation facilities and the level of swim-up fry production which determined the requirements for hormone treatment. Therefore, investment appraisal was used for this economic analysis. This procedure represents the actual flow of costs and returns as induced by the investment profile for the capital items in the different strategies. Ignoring the 'time value of money' would tend to favour strategies with high initial investment costs, low subsequent investment costs and a constant flow of revenues. Investment criteria such as the financial Internal Rate of Return (IRR) or Net Present Value (NPV) were computed (Kuyvenhoven and Mennes, 1985). However, the use of these criteria may have limitations as strictly speaking they are only valid for one investment alternative i.e. the IRR is compared to the bank rate of interest and a positive NPV indicates that the investment is profitable. Dominance analysis was applied to overcome this difficulty which allowed a comparison of alternatives over a range of investment levels. This technique has previously been used to derive input recommendations for farmers concerning different fertiliser levels in maize production (Perrin et al., 1976). Dominance

Fig. 6.4

Dominance analysis using a Net Present Value curve

Source: Perrin et al., 1976



analysis is based on the principle that higher output requires higher input. Thus, strategies (or levels of input) are ranked in ascending order of their input cost and (graphically) related to their net output/net returns. Hence, a strategy is dominated if, at a given level of cost, there is at least one other strategy with higher net output. Consequently, the 'frontier line of strategies' (Fig. 6.4) shows the undominated alternatives from which a decision maker can choose by comparing additional costs for additional net returns. For example, a decision maker who selected Strategy D rather than A (Fig. 6.4) invites increased costs (from C1 to C2) and gains increased returns (N2-N1). In applying this technique to investment appraisal, a comparison can be made of different fry production strategies on the basis of their NPV and total discounted costs. Therefore, discounted cash flow (NPV) and total discounted costs are used instead of net return and variable costs respectively.

Profitability was estimated over an investment period of ten years for all alternative strategies even though some capital items (ponds, tanks etc.) have a longer service life. The fry production sector in Thailand is extremely dynamic and analysis over a longer time perspective would make most assumptions highly uncertain.

6.2.2.2 Determination of costs

Costs were classified as either fixed, e.g. expenditures on construction, machinery and equipment, or variable, e.g. costs

Table 6.5 Definition of main categories of fixed costs associated with the 5 main components of the hatchery production of swim-up fry in Central Thailand model.

1. Spawning

The costs of the production of fertilised eggs by broodfish kept in mixed-sex groups at low density.

2. Conditioning

The costs associated with the maintenance of mature broodfish in single sex groups on a high plane of nutrition at high density.

3. Harvest and Incubation

The costs associated with removal of eggs and/or fry from the spawning arena and subsequent incubation if required.

4. Swim-up

The fixed cost requirements of holding fry after hatching but before total yolk sac absorption.

5. Pond Construction and Fill

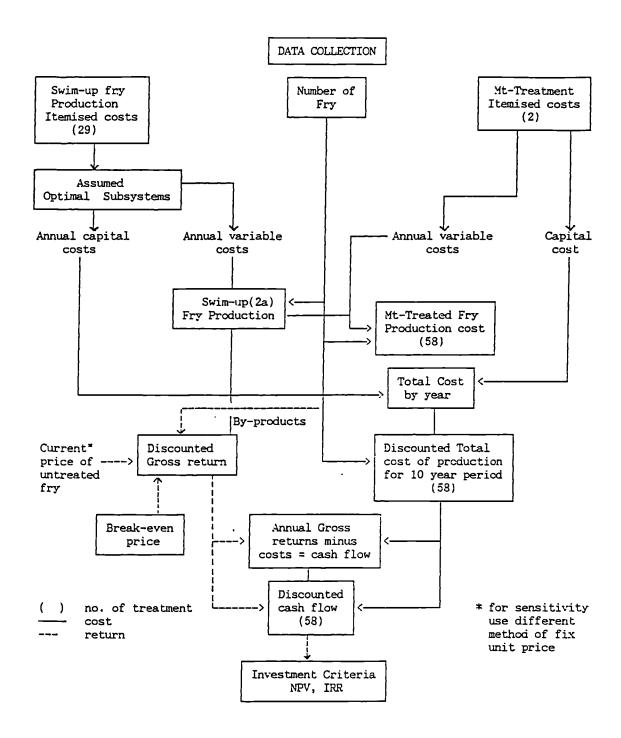
The costs associated with site preparation for each strategy; including extraction of fill, and/or excavation and construction of a pond.

Table 6.6 Major categories of variable cost for swim-up fry production.

1.	Land	
2.	Feed	
3.	Broodstock	initial weight extra weight
4.	Labour	feeding harvest/cleaning
5.	Electricity	<pre>broodstock conditioning seed production incubation/swim-up</pre>
6.	Pond Maintenance	pumping water sludge removal fertilizer piscicide general maintenance

Fig. 6.5

Summary of methodology for the estimation of Net Present Value (NPV) for MT-treated fry production in Central Thailand



directly associated with fry output such as feed, labour, power, and broodstock. Land costs, as a special category of variable cost, were regarded as an opportunity cost, equal to the potential profit forgone for its use.

The fixed costs associated with the production of swim-up fry suitable for sex-reversal were separated into five categories (Table 6.5) for ease of economic analysis. The cost of sex-reversal was subsequently calculated for treatment in tanks and hapas.

All costs used were based on current (1988) prices and were obtained from various sources (AIT purchasing department and directly from merchants and suppliers) except where otherwise stated.

Full annual budgets for 29 strategies for swim-up fry production in a start-up operation in Central Thailand were calculated. The main categories of variable cost for all treatments are given in Table 6.6. The production period was assumed to be 12 months as current tilapia fry production and marketing is year round in this area (Edwards et al., 1983). A schema for the methodology of investment analysis is given (Fig. 6.5). Annual variable costs of MT-fry were computed in two steps based on the variable costs of swim-up fry production being a major cost in subsequent MT-fry production. This cost together with capital and opportunity costs were added on an annual basis, to give a total annual cost which was then discounted at the lending rate of medium term loans (12%).

The assumptions inherent in modelling the substitution of fry production strategies in Northeast Thailand carp hatcheries are the same as a full start-up operation in Central Thailand with the following exceptions:

(a) Additional strategies in Northeast Thailand

An opportunity cost was not included for strategies (h11b-h35b) as it was assumed that the effect of tilapia fry production on overall carp production was negligible.

(b) Period of production

A fry production season of 6 months was used as a basis for estimation of production and costs for substitution of tilapia fry production strategies into Northeast Thailand hatcheries. All variable costs except for pond 'maintenance costs' were calculated for the 6 month production period only. It was assumed that farmers sold off remaining broodstock after the production cycle and bought in new stock every year from a neighbouring grow-out farmer. Pond costs were calculated over a 12 month basis at Bt $6.6/m^2$.

(c) Hormone treatment

Costs for hormone treatment were based on sex-reversal in hapas only.

(d) Estimation of opportunity cost of substitution

The effect on costs of the substitution of tilapia for carp was calculated as the marginal cost of tilapia fry production which includes both opportunity and other variable costs.

The opportunity cost of fry production was calculated as the amount of profit lost in the carp system due to the

introduction of the new tilapia production system. The area of substitution is of three types: (1) pond area used for swim-up fry production, (2) pond area used for MT-treatment in hapas, and (3) residual land used for tanks, dykes, roads, etc.

The first two categories have an opportunity cost at a level relative to the intensity and efficiency of current carp production. The residual land area was assumed to have a low opportunity cost, as 'non-productive' land. Typically 'residual land' will be higher rice terrace with a mixture of home enterprises which tend to be value added but rarely cash orientated.

(e) Fixed costs

Fixed costs were calculated on the basis of the additional investment cost required for the different tilapia production strategies within carp hatcheries. It was assumed that concrete tanks or nylon hapas, if currently used for carp production, were unsuitable for tilapia breeding.

6.2.2.3 Determination of revenues

Discounted gross return, calculated on the current price of untreated fry (Bt 0.1/fry), was used as a basis for estimation of discounted net return. Yield and return was estimated by break-even analysis for each dominant treatment.

Annual gross revenues (returns) for the entire investment period considered were calculated by multiplying the quantity of fry produced per year with the current price of untreated

fry. Subsequently, the current fry price was varied, thus representing several levels of 'break-even price' from the viewpoint of the grow-out producer and the fry producer.

Table 6.7 Discounted fixed costs (investment period 10 years) by category for 29 swim-up fry production strategies in Central Thailand

Strongo	Percentage of total costs by major category					
Stragegy	Spawning	Condi- tioning	Harvest & incubation	Swim-up	Pond construction	Total (Baht)
Tankl.1	73.9 49.1	0.0 31.4	17.7 12.1	5.6 3.7	2.8 3.7	233759 359187
Tank1.2	95.8	0.0	0.6	0.0	3.6	184302
Tank1.3 Tank1.4	49.1	31.4	12.1	3.7	3.7	359187
Tank1.4	49.5	31.4	12.3	2.6	3.8	352846
Tank2.2	66.7	7.8	16.5	5.0	4.0	262159
Tank2.3	49.5	31.8	12.3	2.6	3.8	352846
Tank2.4	66.7	7.8	16.5	5.0	4.0	262159
Tank3.1	49.3	31.6	12.1	3.7	3.3	357326
Tank3.2	49.3	31.6	12.1	3.7	3.3	357326
Tank3.3	49.3	31.6	12.1	3.7	3.3	357326
Tank3.4	49.3	31.6	12.1	3.7	3.3	357326
Hapal.1	18.5	43.3	27.0	4.1	7.1	620591
Hapa1.2	31.0	5.5	45.1	6.9	11.6	371046
Hapa1.3	29.0	43.3	27.0	4.1	7.1	620591
Hapal.4	29.0	5.5	45.1	6.9	11.6	371046
Hapa2.1	29.0	11.7	42.0	6.5	10.8	395821
Hapa2.2	29.0	11.7	42.0	6.5	10.8	395821
Hapa2.3	29.0	11.7	42.0	6.5	10.8	395821
Hapa2.4	29.0	11.7	42.0	6.5	10.8	395821
Hapa3.1	29.0	11.7	42.0	6.5	10.8	395821
Hapa3.2	27.3	16.4	38.5	6.1	11.7	420816
Hapa3.3	25.7	20.7	36.3	5.7	11.6	446339
Hapa3.4	29.0	11.7	42.0	6.5	10.8	395821
Hapa3.5	25.7	20.7	36.3	5.7	11.6	446339
Pond1.2a	0.0	8.7	8.6	0.0	82.7	73506
Pond1.3a	0.0	8.7	8.6	0.0	82.7	73506
Pond2.2a	0.0	8.7	8.6	0.0	82.7	73506
Pond2.3a	0.0	8.7	8.6	0.0	82.7	73506

Table 6.8 Initial fixed costs by major category for all fry production strategies in the Central Thailand model

Tankl.1 78.3 0.0 14.3 4 Tankl.2 51.4 32.5 9.3 2 Tankl.3 96.1 0.0 1.0 0 Tankl.4 51.4 32.5 9.3 2	m-up Pond construction .3 3.1 .8 4.0 .0 3.7 .8 4.0 .8 4.0	Total (Baht) 215559 335737 179651 335737
Tankl.1 78.3 0.0 14.3 4 Tankl.2 51.4 32.5 9.3 2 Tankl.3 96.1 0.0 1.0 0 Tankl.4 51.4 32.5 9.3 2	construction 3.1 8 4.0 0 3.7 8 4.0 8 4.0 4.0	215559 335737 179651
Tankl.2 51.4 32.5 9.3 2 Tankl.3 96.1 0.0 1.0 0 Tankl.4 51.4 32.5 9.3 2	4.0 3.7 8 4.0 4.0	335737 179651
Tankl.3 96.1 0.0 1.0 0.0 Tankl.4 51.4 32.5 9.3 2	3.7 4.0 4.0	179651
Tankl.4 51.4 32.5 9.3 2	4.0 4.0	
, ,	.8 4.0	1 225727 1
m 1		1 222121
Tank2.1 51.3 32.7 9.3 2		335989
Tank2.2 75.2 2.4 13.7 4	4.6	229014
Tank2.3 51.3 32.7 9.3 2	4.0	335989
Tank2.4 75.2 2.4 13.7 4	4.6	229014
Tank3.1 51.6 32.7 9.4 2	3.6	333876
Tank3.2 51.6 32.7 9.4 2	3.6	333876
Tank3.3 51.6 32.7 9.4 2	3.6	333876
Tank3.4 51.6 32.7 9.4 2	3.6	333876
	.6 9.3	472198
Hapal.2 12.7 5.1 56.3 8	17.3	248538
Hapa1.3 6.6 49.8 29.6 4	.6 9.3	472198
Hapal.4 12.7 5.1 56.3 8	17.3	248538
Hapa2.1 12.7 5.1 56.1 8	17.4	247428
Hapa2.2 12.7 5.1 56.1 8	17.4	247428
Hapa2.3 12.7 5.1 56.1 8	17.4	247428
Hapa2.4 12.7 5.1 56.1 8	17.4	247428
	17.4	247428
1	19.2	255656
	19.5	264413
	17.4	247428
Hapa3.5 11.9 9.5 50.8 8	19.5	264413
Pond1.2a 0.0 3.1 3.0 0	.0 j 93.9	56617
Pond1.3a 0.0 3.1 3.0 0	.0 93.9	56617
Pond2.2a 0.0 3.1 3.0 0	.0 93.9	56617
Pond2.3a 0.0 3.1 3.0 0	93.9	56617

Table 6.9 Initial fixed costs for (1) spawning category by item for tank strategies (T31-T34) and (2) incubation category by item for hapa strategies (H31-H35) in Central Thailand

1	1	١
١	Τ	. ,

Component	Cost (Baht)	*
Roof Concrete base Pipework Rings and bricks Pump	75,600 32,628 19,435 40,348 4,300	43.9 18.9 11.2 23.4 2.5
Total Cost	172,311	100.0

(2)

Component	Cost (Baht)	*
Roof	33,750	25.5
Hatchery fittings	9,220	7.0
Concrete base	14,013	10.6
Pipework	6,902	5.2
Filter	60,000	45.2
Pump	8,600	6.5
Total cost	132,485	100.0

Table 6.10 Annual variable costs by category for all fry production strategies in the Central Thailand model

Stratog				Percei	ntage by catego	ory .	
Strategy	Land rental	Feed	Brood- stock	Labour	Electricity	Pond maintenance	Total (Baht)
Tank1.1	1.2	27.3	0.0	31.8	39.7	0.0	36264
Tank1.2	2.0	30.2	0.0	23.6	44.2	0.0	48874
Tank1.3	1.0	35.3	0.0	39.0	24.8	0.0	29074
Tank1.4	2.2	26.2	0.0	25.0	46.7	0.0	46256
Tank2.1	2.0	30.9	0.0	28.8	38.4	0.0	56274
Tank2.2	1.3	33.6	0.0	32.1	28.6	4.5	50390
Tank2.3	2.2	30.7	0.0	28.7	38.4	0.0	56315
Tank2.4	1.2	33.4	0.0	32.2	28.7	4.5	50184
Tank3.1	2.6	23.3	0.0	27.8	46.3	0.0	46697
Tank3.2	2.6	25.1	0.0	29.9	42.5	0.0	50882
Tank3.3	2.6	27.3	0.0	31.3	38.8	0.0	55680
Tank3.4	1.9	29.4	0.0	32.8	36.0	0.0	60068
Hapal.1	3.8	21.6	2.1	32.3	26.9	13.3	130662
Hapa1.2	4.3	24.6	0.9	35.0	16.7	18.4	120283
Hapa1.3	3.9	20.8	2.1	32.5	27.2	13.5	129552
Hapal.4	4.2	24.1	1.0	35.3	16.8	18.6	119373
Hapa2.1	2.5	20.3	0.0	35.3	19.9	22.0	100757
Hapa2.2	2.4	21.0	0.0	37.4	18.7	20.6	107697
Hapa2.3	2.3	21.4	0.0	39.3	17.6	19.4	114308
Hapa2.4	2.2	21.0	0.0	41.5	16.8	18.5	119511
НараЗ.1	4.4	17.2	0.2	38.5	18.8	20.8	106559
Нара3.2	4.2	19.8	0.1	36.4	17.8	21.7	112990
НараЗ.З	3.9	22.1	0.0	34.6	16.9	22.6	118840
НараЗ.4	4.7	16.5	0.4	38.7	18.9	20.9	106266
Hapa3.5	4.2	15.8	3.0	35.9	. 17.6	23.5	114406
Pond1.2a	14.7	5.2	0.0	26.5	0.0	53.6	47645
Pondl.3a	13.6	5.8	0.0	17.6	0.0	63.0	40546
Pond2.2a	16.2	4.6	0.0	22.1	0.0	57.1	43905
Pond2.3a	24.6	4.2	0.0.	19.9	0.0	51.3	48844

- 6.3 Determination of Production Costs
- 6.3.1 Swim-up Fry
- 6.3.1.1 Central Thailand farms

Tanks

Tanks were used only for spawning fish (T11, T13, T22, T24), and for conditioning fish (H11, H13) or for both (T12, T14, T21, T23, T31-4).

The separation of spawning and conditioning was an expensive procedure. The extra fixed costs required for conditioning fish prior to spawning were considerable; in Treatments replacing broodstock (T12, T14), these costs were over 30% of total fixed costs. In non-replacement treatments, fish spawning costs were the most important category (Table 6.7). Harvest and incubation costs never exceeded 20% of total fixed costs for any tank treatment, and pond construction never more than 5%.

Initial construction costs (yr 0) were by far the largest component of total costs (Table 6.8). Subsequent costs were relatively minor - the most important being for replacement of submersible pumps every 5 years. The largest single component of initial costs of the spawning phase was for roof construction (approximately 44%) (Table 6.9).

The use of nylon hapas for conditioning fish (T22, T24) reduced total capital costs by over 25% and variable costs by approximately 10% compared with tank conditioning (T21, T23).

Differences in broodstock and feed costs between treatments in T2 were relatively minor (Table 6.10), broodstock costs were nearly 7% lower in non-replacement treatments but feed costs were only slightly higher for tank conditioning than hapa conditioning and total replacement than partial replacement treatments respectively. Electricity was the variable cost most reduced (27%) by use of hapas rather than tanks for conditioning of broodstock. Stocking density (T31-T34) had no effect on capital requirements but variable costs, except for electricity, increased proportionally with fish density (Table 6.8).

Hapas

Nylon hapas were used for spawning (H11, H13) and conditioning only (T22, T24), or both (H12, H14, H21-H24, H31-H35).

The most important categories of capital cost for hapa systems, despite the need for replacement of nets for spawning and conditioning every 2 years, were harvest and incubation costs (Table 6.8). In particular, the cost of a slow-sand filter required for an 18 hapa system was high (Bt 60,000) (Table 6.9). Initial costs for roofing (25%) and a slow-sand filter (45%) were by far the most important costs within the incubation category.

Incubation and harvest costs made up over 40% of capital costs for most treatments that both spawned and conditioned fish in nets. Capital costs for treatments requiring a greater capacity for conditioning fish (H32, H33, H35) were more evenly spread between spawning, conditioning and incubation cost categories.

The ratio of spawning to conditioning costs was considerably higher for hapa production than for tanks (2.5:1 compared to approximately 1.5:1), suggesting that conditioning was a particularly expensive option in concrete tank а hatchery. Capital costs in hapa systems made up a significantly lower proportion of total costs than in tank systems (28-39% and 47-78% respectively). Labour was the most important variable cost for all hapa treatments (30-40% of variable costs). Feed and electricity costs were correspondingly lower, a proportion of total variable costs than in tank treatments.

Variable costs were relatively unaffected by conditioning in tanks or hapas (H11-H14). Variable cost for hapas increased proportionally with stocking density (H21-24), as in tanks. The number of replacement broodstock required affected total variable costs considerably (H31-35) in strategies conditioning broodfish for longer periods of time; the treatment requiring most broodstock (H33) had variable costs 12% in excess of the treatment using fewest broodstock (H34). However, the proportion of costs changed little with treatment.

Earthen ponds

Strategies for fry production in earthen ponds included the simple release of broodfish into earthen ponds (P12, P13, P21, P22) or their spawning and/or conditioning in nylon hapas (see above). All required initial capital costs, plus variable costs associated with pond maintenance (pumping, fertilising, sanitising, etc.). Capital costs were concentrated in the first

Discounted fixed and variable costs as a proportion of total discounted costs of undominated swim-up fry production strategies in Central Thailand

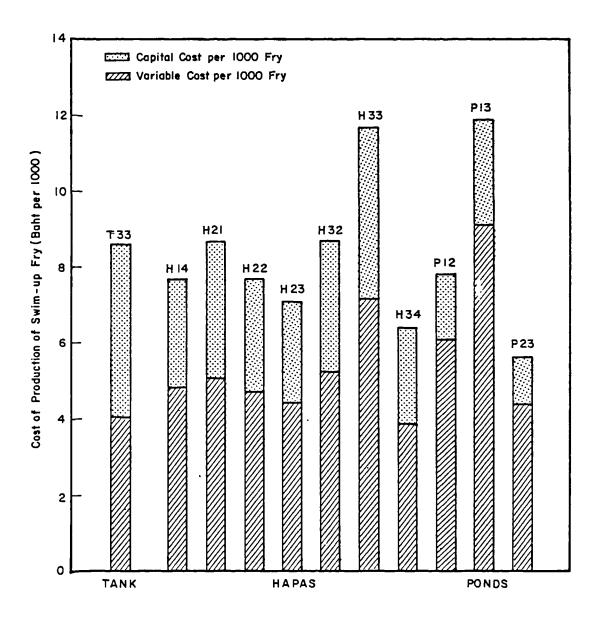


Table 6.11 Discounted total costs (investment period 10 years) and initial investment costs for 43 swim-up fry production strategies in Northeast Thailand

Strategy	initial investment	Discounted cash flow (10 years)	initial investment as % of total
Tank1.1	265205	236207	89
Tank1.2	399471	363094	91
Tank1.3	203013	195704	96
Tank1.4	399471	363094	91
Tank2.1	390273	350725	90
Tank2.2	308748	253835	82
Tank2.3	392838	351425	89
Tank2.4	306182	253135	83
Tank3.1	404653	363240	90
Tank3.2	407219	363940	89
Tank3.3	412350	365340	89
Tank3.4	402087	362540	90
Hapal.1	635971	470025	74
Hapa1.2	447100	242781	5-1
Hapal.3	646233	472825	73
Hapa1.4	439403	240681	55
Hapa2.1	459286	271758	59
Hapa2.2	466983	273858	59
Hapa2.3	474680	275958	58
Hapa2.4	474680	275958	58
Hapa3.1	424009	236481	56
Hapa3.2	448836	239971	53
Hapa3.3	462784	243777	53
Hapa3.4	441968	241381	55
Hapa3.5	478178	247977	52
Pond1.2a	28402	12381	14
Pond1.3a	23271	10981	47
Pond2.2a	25837	11681	45
Pond2.3a	46362 .	17281	37
h11a/h11b	383680	298699	78
h12a/h12b	271237	167070	62
h13a/h13b	388811	300099	77
h14a/h14b	268671	166370	62
h21a/h21b	281753	184983	66
h22a/h22b	284318	185683	65
h23a/h23b	289450	187083	65
h24a/h24b	289450	187083	65
h31a/h31b	260974	164270	63
h32a/h32b	275197	168150	61
h33a/h33b	282619	170175	60
h34a/h34b	268671	166370	62
h35a/h35b	290316	172275	59
HUPF	257255	88047	34

year of operation for all earthen pond strategies, and pond construction makes up by far the largest proportion of total costs in all treatments (82.7%). Other capital costs were limited to small accessory equipment items for holding fish and removing fry. The ratio of capital to variable costs was much lower in pond fry production than for hapas or tanks, mainly because less investment was required (Fig. 6.6). Overall costs of production were very low (P23) or moderately high compared to hapa and tank treatments respectively. Pond maintenance costs were the most important category of variable cost, and this was largely due to the cost of sediment removal.

6.3.1.2 Northeast Thailand farms

The differences in costs between the Central and Northeast Thailand hatchery models were related mainly to the additional strategies simulated and are described in terms of capital and variable costs below:

(a) Initial capital costs

A smaller system using one pond and nine spawning nets (H1-4 a&b) lowered initial investment requirements by only 30-40% compared to a two-pond system (H11-H35), as incubation costs did not decline proportionally (Table 6.11).

The hapa system used commercially in Northeast Thailand (HUPF) was intermediate in terms of initial capital requirement (Bt 88047) between ponds (very low) and all other hapa strategies (Table 6.11). This was mainly because incubation and swim-up facilities were not required; however subsequent

Table 6.12 Annual variable costs by category for selected swim-up fry production strategies (P23, H12, T11, H34a, H34b, HUPF) in five hatcheries in Northeast Thailand

		Perc	sentage of	Percentage of total variable costs by category	le costs	by cate	gory
Strategy	llatchery	Feed	Labour	Electricity	Pond nain- tenance	Land	Total (BL)
P23	Ubon Korat	2.7	£*9		23.9	68.1	86,304
	odorn/surin/ Nongkhai	3.0	0.9	ı	26.7	64.3	77,018
1112	Ubon Korat	16.2	14.1	9.4	10.4	50.0	114,795
	Nongkhai	17.6	15.3	10.2	11.3	45.6	105,561
T11	Ubon Korat	33.3	23.8	41.5	1 1	1.4	17,334 17,334
	Nongkhai	33.3	23.8	41.5	ı	1.4	17,334
1134a	Ubon Korat	11.4	16.3 19.6	11.2	12.4 14.8	48.7	96,502 80,472
	odorn/surin/ Nongkhai	14.5	20.8	14.2	15.7	34.7	75,856
11346	Ubon Korat	20.9	29.9	20.5 20.5	22.6	6.1	52,715 52,715
	odorn/surin/ Nongkha i	20.9	29.9	20.5	22.6	6.1	52,715
HUPF	Ubon Korat	20.0	4.2	3.4 4.6	1.5	70.9 60.8	49,233 36,622
	Udorn/Surin/ Nongkhai	24.8	6.3	5.1	2.3	56.5	32,987

capital expenditure was higher.

(b) Annual variable costs for swim-up fry production

Variable costs, as compared by category, showed large proportional differences between systems. The five main categories of variable cost are compared in Table 6.12 for different systems and within different hatcheries in Northeast Thailand. Broodstock were not considered in this comparison although they were an initial cost, as they became an output or a source of net revenue on an annual basis. The systems compared were dominant strategies (see below), except for HUPF which was included as an example of a viable, intensive commercial system.

In general, ponds had a higher proportion of area-related costs (i.e. land rental and pond maintenance); feed, labour and electricity in contrast were a minor source of costs. Intensive tank systems (e.g. T11) showed the opposite trends, with hapa systems intermediate.

Hapa strategies, depending on whether tilapia production was considered an addition to carp production (h34b) or substitution for it (h34a), had variable costs evenly balanced between the main categories. The commercial 'pumped' hapa system (HUPF) using natural incubation and a long inter-harvest interval had a different cost profile to the other more intensive hapa systems. Land rental costs were a higher proportion of annual costs compared to other hapa systems, whilst electrical costs were proportionally lower despite considerable pumping.

Cost of hormone treatment for one million fry in (i) shallow concrete tanks (Area = 4 $\rm m^2$) and (ii) nylon hapas (Area = 5.4 $\rm m^2$) Table 6.13

Treatment		Variab	le Cost	(Baht)	
Treatment	1	nvestment Cost(%)			Tumber of uni	
(i) Tank	Construction Roof Pipes & Valves Rings & Bricks Equipment	45.7 4.9 25.7 30.0 19.4	Land ^c (m ²) Stock (1000) Alcohol (1) Feed (kg) Hormone (g) Electricity Labour Maintenance	50 12 108	14	2.5 12.1
	Total (Bt)	53069				59414
(ii) Hapa	Construction Equipment Hapas	18.3 67.0 14.7	Land ^c (m ²) Stock (1000) Alcohol (1) Hormone (g) Feed (kg) Electricity Labour Maintenance	50	125 173 51 14 233 90 108	8.2 4.8
	Total (Bt)	9506				31278

based on 30 day treatment period 1,036,000 fry

b

land cost in central Thailand

Variation in costs between sites was directly related to differences in the opportunity cost of pond use; these differences were least in intensive or 'additional' systems that were additional to rather than substitutive for carp activities (T11 and H34b).

6.3.2 Hormone treatment costs

The investment cost of hormone treatment in concrete tank systems, in terms of cost per fry, was higher than for treatment in hapas by a factor of more than five (Table 6.13). Total variable costs in hapas were almost half (53%) those required to treat the same number of fry in tanks. More land area, a greater amount of feed with a consequent increase in the requirement for alcohol, plus more labour were required for sex-reversal in tanks. However, the required dose of hormone was lower than for hapa treatment and the absolute amount of pure hormone used was approximately the same.

Feed costs for hormone treatment were 22% of variable costs in hapas and 16% in tanks; the cost of feed and alcohol was the largest part of these costs; the hormone itself was only 4.8% and 2.5% of variable costs in the two systems respectively.

Table 6.14 Annual swim-up fry production for strategies in Northeast and Central regions of Thailand

Strategy	Annual fry pro	duction (*1000)
	Northeast	Central
Tank1.1 Tank1.2 Tank1.3 Tank1.4 Tank2.1 Tank2.2 Tank2.3 Tank2.3 Tank3.3 Tank3.1 Tank3.2 Tank3.3 Tank3.4 Hapa1.2 Hapa1.3 Hapa2.1 Hapa2.1 Hapa2.3 Hapa3.2 Hapa3.2 Hapa3.2 Hapa3.4 Hapa3.2 Hapa3.5 Pond1.3a Hapa3.4 Hapa3.4 Hapa3.4 Hapa3.4 Hapa3.4 Hapa3.5 Pond2.3a Hapa3.4 Hapa3.4 Hapa3.5 Pond2.3a Hapa3.5 Pond2.3a Hapa3.6 Hapa Hapa3.6 Hapa Hapa3.7 Hapa3.6 Hapa	1628 1899 1997749 12534191 23301115 3310115 3310115 42777398 876769831969 649886477 8875779988 41838117447 4183811744990 4183811749904 318389510 3	3257 37577 3915897 5603230 667201782 667201782 1165496331 1161881299 1161881299 1161881299 11618811 11061930 114466829 11618811 11466829

6.4 Determination of revenues

Revenues were determined by the number of fry produced (yield) and their unit price.

6.4.1 Fry output

Fry production from the different experimental treatments was extrapolated to the commercial strategy using a factor for both scale and production period (Table 6.14). Thus, AIT data on fry production from one concrete tank for 106 days was extrapolated to a system of nine tanks operated for 360 days in Central Thailand.

6.4.2 Fry price

Since MT-treated fry are not yet produced commercially in Thailand, no market price exists. However, in order to calculate the potential net revenue of any hatchery aiming to produce MT-treated fry, a price must be set that is acceptable to the purchaser, i.e. the farmer who grows the fish to market size as well as the fry producer. In order to calculate the potential net revenue of any hatchery aiming to produce MT-fry, different possible price levels were assumed. Clearly, if the MT-fry treatment was going to be adopted by hatcheries, the price of fry must be acceptable for the hatchery producer as well as the grow-out farmer; i.e. it must be high enough to guarantee satisfactory profitability to stimulate the required additional investment. Conversely, the price must be low enough for the grow-out farmer to assure him satisfactory additional

profits. An equilibrium price will be reached in a free market system, at which producers and buyers of fry are equally satisfied (Snodgrass and Wallace, 1975).

In the absence of an existing market price for MT-fry, three price levels have been used for the determinations of revenue or returns and profitability.

6.4.2.1 Determination of different levels of fry price

(a) Current price

The current price of untreated fry (Bt 0.1 per fish, length 2-3 cm) at Government fishery stations and commercial fry suppliers was used to estimate net returns for different fry production strategies.

(b) Using marginal analysis

The increase in net return at current fry prices that the growout farmer could gain by stocking MT-treated fish (new
technology) compared to normal, untreated fish ('current'
practice) was estimated using marginal analysis. In other words
by how much could the fry price increase if the grow-out
production fully realized the potential of MT-treated fry.

Estimation of price using marginal analysis (b)

Data from an experiment on the AIT campus (AIT unpublished data) were used to model 'potential' net returns for a variety of fry costs and size related yield values. The net revenue obtained from growing, harvesting and marketing a crop of tilapia depends on several factors and is defined as the gross

Table 6.15 Mean harvested yields by size class of MT-treated and untreated <u>O. niloticus</u> stocked in 200 m² fertilised (septage) earthen ponds within the AIT experimental system for 5 months and fed fine rice bran

Size class (g)	Total harvest	ed yield (kg)
	untreated*	treated*
<10 <50 >50 <100 >100 <200 >200 <300 >300 <500	0.17 - 0.73 36.28	- 0.33 0.32 28.77 16.88
Total weight	37.19	46.30

^{*} mean of 3 replicates

Table 6.16 Marginal analysis of substitution of MT-treated fry for untreated fry and later marketing in an urban market in the Northeast of Thailand.

Fish raised in earthen ponds fertilised with tank slurry and fed fine rice bran.

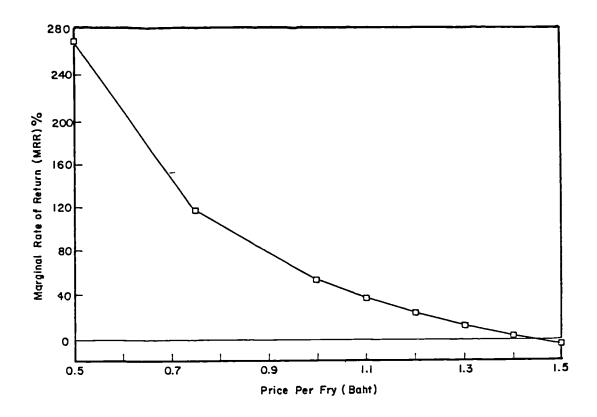
	Fry price (Bt/fry)	Total variable costs (Bt)	Net benefit (Bt)	Change in cost (Bt)	Change in benefit (Bt)	Marginal rate of return (%)
Untreated fry	0.1	260	479	-	-	<u>-</u>
MT-treated					_	
fry	0.1 0.5 0.75 1.0 1.1 1.2 1.3 1.4	260 239 392 445 466 487 509 530 551	756 677 624 571 550 529 507 486 465	74 124 174 194 214 234 254 274	277 198 148 92 71 50 29 7 -14	268 117 53 37 23 12 3 -5

Sensitivity analysis of the use of MT-treated fry compared to current use of untreated fry1 for 3 different market scenarios. Table 6.17

N The state of the		Siz	Size range of fish in g	fish in g		Marginal	Price of
market Scenario		d	Price (Baht per kg)	per kg)		rate of	fry
	<50 .	>50<100	>100<200	>50<100 >100<200 >200<300 >300<500	>300<500	%	(Dant/115ii)
Urban Northeast Thailand	4	12	91	81	20	53	1.0
Rural Northeast Thailand ²	20	20	20	20	20	45	0.75
Central Thailand	4	4	4	8	10	43	0.50

price of untreated fry Bt. 0.1 high value of small fish because of demand for restocking

The relationship between the marginal rate of return (MRR) and fry price after substitution of MT-treated fry for untreated fry by a grow-out farmer



return minus the variable production costs. Gross return is a function of yield and price of output. The latter is of primary importance, especially where there is a fish size preference in the market such that the 'size structure' of the harvested yield greatly affects gross returns. Thus, a higher yield of small fish may be of lower value than a smaller yield of large fish, or vice versa. It was therefore necessary to compute an average market price which was weighted according to the frequency of each size class. Variable costs included feed, labour and energy, as well as the cost of fry. If fixed costs were assumed equal, a partial budget analysis can be used for variations in the cost of fry. The MT-treatment increased yield and mean individual size of the harvested fish (Table 6.15). The improved yield distribution increased the value of the crop accordingly (Table 6.16, 6.17) and therefore the maximum increase in fry price (Fig. 6.7) corresponded to that profit margin.

However, at this point all the benefit would go to the fry producers, leaving no incentive for the grow-out farmers to buy MT-treated fry. Grow-out farmers are likely to accept a higher price only if the corresponding returns were sufficiently attractive.

It has been hypothesised that a minimum rate of return of around 40% is required on additional input expenditures before farmers adopt new agronomic practices (Perrin et al., 1976). Use of this Marginal Rate of Return (MRR) allowed the computation of fry price expected to be acceptable to the

Table 6.18 Total annual value of revenue components for 29 swim-up fry production stratgies in Central Thailand (Baht)

Strategy	Swim-up fryd	Broodstock ^a	Recruitsc	Large fryd	Total
Tank1.1	65136	6774	-		71910
Tank1.2	75835	8179		1	84014
Tank1.3	11545	6912			18457
Tankl.4	79601	7741			87342
Tank2.1	103089	13581			116670
Tank2.2	133775	13638			147412
Tank2.3	120748	12819			133567
Tank2.4	124433	13242			137675
Tank3.1	122595	3969			126564
Tank3.2	135648	5101		j	140749
Tank3.3	164050	6161			170211
Tank3.4	108276	7293			115569
Hapai.1	252133	16197			268330
Hapa1.2	325087	18561			343648
Hapal.3	308781	15014			323795
Hapal.4	285128	14246			299374
Hapaz.1	233659	9619	Ì		243278
Hapa2.2	272618	13467	ļ		286085
Hapa2.3	312051	15601			327652
Hapa2.4	320698	18126			338824
Hapa3.1	236613	6042			242655
Hapa3.2	256282	112990			369272
Hapa3.3	200576	118805]]	319381
HapaJ.4	323980	106266	}		430247
HapaJ.5	283077	114406	Ì	Ì	397482
Pond1.2a	93056	8268	2113	3599	107036
Pond1.3a	53206	7747	3000	2664	66617
Pona2.2a	47632	8089 b	2954	989	59664
Pond2.3a	132179	7950, 6	3216	130	143476

<sup>a priced at Bt10/kg
b priced at Bt12/kg
c priced at Bt4/kg
d priced at Bt0.02/fish</sup>

grow-out farmer (Fig.6.7, B; Table 6.16). Calculation of the MRR for different price profiles of fish harvested from treatments using MT-treated rather than untreated fry gave positive returns (MRR>40%; Table 6.17).

Even if MT-fry were five times more expensive than untreated fry, it would still be worthwhile for the grow-out farmer to purchase them. The price of grow-out fish also has an affect on the prospective value of fry. In Northeast Thailand, small fish may have a very high value (per kg) for sale as large fingerlings (Table 6.17), but the use of MT-treated fry would still be advantageous even if the cost of fry were Bt 0.75. A market scenario in which fish prices were high (e.g. Urban Northeast Thailand, Table 6.17), but where there was a high premium for larger fish, gave the best incentive for the use of MT-treated fry, even when the price was a factor of ten greater than the current fry price.

(c) & (d) Break-even price

A third alternative for price setting was the determination of the unit cost of MT-fry at a point where the fry producer would obtain a reasonable return (c). This is distinct from the break-even price (d) which is the unit cost of production at a given level of output. Price (c) was calculated by fixing the IRR at 16% or 20% and determining the fry price by interpolation or extrapolation of prices at known IRR's. An IRR of 20% was chosen as this was well above the current rate of interest charged by commercial banks for medium term loans. The last alternative was to compute the break-even price

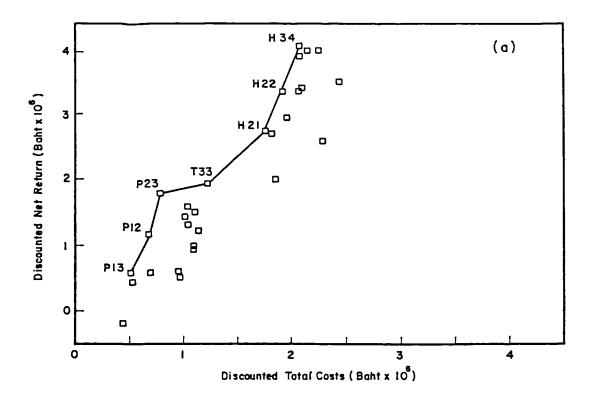
- (d) calculated as the price when the net present value (NPV)=0.
- 6.4.3 Value of by-products of fry production

The proportion of revenues from by-products of the hatchery system is given in Table 6.18. The overall contribution of by-products (i.e. recruits, fry too large for sex reversal, net broodstock production) to revenue, however, accounted for only between <1-11% of total revenue, suggesting that use of broodstock and/or capture of fry was intensive and efficient.

Discounted cash flow of 29 swim-up fry production strategies in Central Thailand after

(a) sex-reversal in hapas and(b) sex-reversal in tanks

(Undominated strategies are indicated)



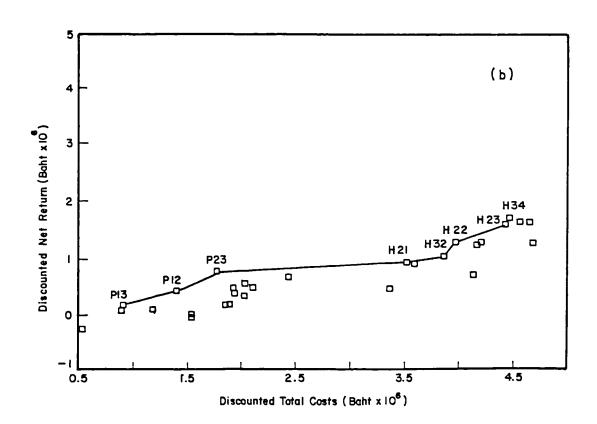


Table 6.19 Marginal analysis of investment in undominated strategies in Central Thailand of different discounted total cost (fry price Bt.0.1) after sex-reversal in tanks and hapas

Strategy	Discounted net return (Bt)	Discounted total costs (Bt)	Marginal net return (Bt)	Marginal costs (Bt)	Marginal ¹ rate of return(%)
TANK P13 P12 P23 H21 H32 H22 H23	201,464 478,264 834,464 1,025,504 1,112,341 1,350,974 1,665,498	905,957 1,397,955 1,775,768 3,522,475 3,855,926 3,969,660 4,425,937	276,800 356,200 191,040 86,837 238,633 314,524 121,058	491,998 377,814 1,746,707 333,451 113,734 456,277 44,557	56 94 11 26 210 69 272
Н34	1,786,556	4,470,494	121,000		
HAPA P13 P12 P23 T33 H21 H22 H34	598,637 1,202,948 1,823,016 1,971,340 2,789,891 3,408,310 4,193,981	508,784 637,271 787,217 1,217,966 1,758,088 1,912,324 2,063,069	604,311 620,068 148,324 818,551 618,419 785,671	164,487 113,946 430,749 540,122 154,236 150,745	367 544 34 152 401 521

Marginal net return

Marginal cost

Marginal cost

6.5 Results

6.5.1 Central Thailand farms

6.5.1.1 Calculation of Net Present Value (NPV)

At current fry prices (Bt 0.1 per fry) NPV's ranged from -0.3 to +1.8 million baht for hapa MT-treatment and from -0.3 to +4.5 million baht for tank MT-treatment (Fig.6.8). NPV's were related to discounted total costs for the purpose of selecting the most economic strategies by applying the concept of dominance analysis. Out of 21 strategies, eight were undominated if fry were tank-treated and seven if hapa-treated (Fig. 6.8). Pond production strategies tended to dominate at the lower level of production costs and the more intensive hapas-in-ponds at the higher level.

5.5.1.2 Marginal analysis of undominated treatments

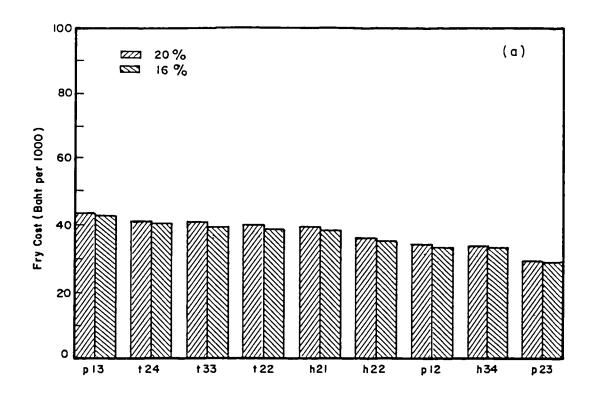
Marginal analysis of undominated treatments at a fry price of Bt 0.1 for sex reversal in tanks reveal marginal rates of return lower than that required for farmer adoption (i.e. < 40%) at levels of investment in excess of that required for the P23 strategy (Table 6.19).

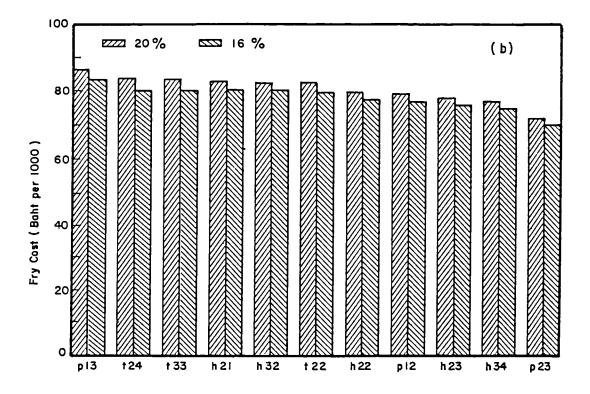
If P23 were eliminated from the NPV curve (Fig. 6.3; Table 6.19) for technical reasons (see 6.2.2.1), other strategies after MT-treatment in both hapas and tanks, the total discounted costs of which were below Bt 1.5 million, become undominated. Marginal analysis, however, indicated that investment at a level higher than that for a pond system (P12)

Sensitivity of IRR (16% and 20%) of fry price for the production of MT-treated tilapia after treatment in (a) hapas and (b) tanks

after swim-up fry production in a range of pond, hapa and tank methods (undominated strategies)

IRR = internal rate of return





was favourable (approx. 40%) at higher investment levels at the current fry price (Bt 0.1 per fry).

6.5.1.3 Sensitivity analysis

Break-even analysis of the individual undominated fry production strategies allowed calculation of fry prices which led to an IRR of 12% (discount rate) (d). This revealed that hormone treatment in tanks had a much higher break-even price in hapas (Fig.6.9). The break-even price for tank MT-treatment varied from under Bt 0.07 to over Bt 0.085 per fish, compared to under Bt 0.03 to over Bt 0.05 per fish for hapa treatment. The "commercial break-even prices" (c), i.e. when the fry producer earns an adequate return (IRR = 16%), followed a similar trend (Fig. 6.9) and were only slightly above the cost of production. Sensitivity analysis using a price that yielded an IRR of 20% indicated that a slight increase in fry price had a major effect on return. The fry price required to yield an internal rate of return of 16% after MT-treatment in tanks was around twice that required after MT-treatment in hapas (Bt 0.07-0.08 and Bt <0.03-0.045, respectively; Fig. 6.9) over a range of strategies. Clearly, lower cost treatments showed a particularly wide difference in the break-even fry price required (e.g. P23; Bt 0.029 in hapas, Bt 0.070 in tanks) and required lower prices of fry if an IRR of 20% were to be achieved. The differences reflected the much higher cost of hormone treatment in tanks compared to hapas.

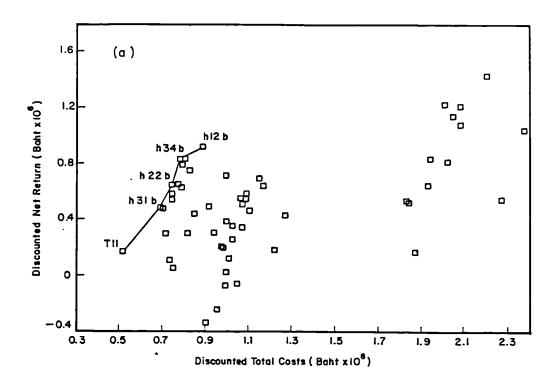
Table 6.20 Undominated strategies for producing swim-up fry over a range of discounted total cost in five hatcheries in Northeast Thailand

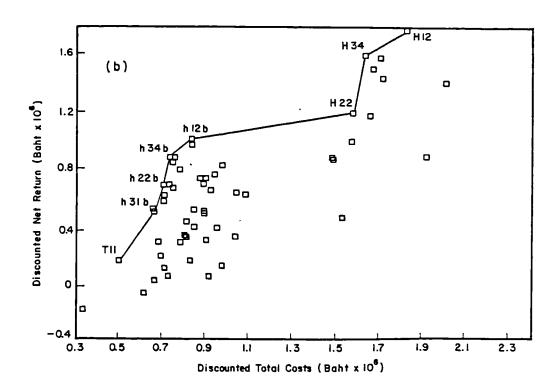
Investment range (Bt x 106)	Strategy	Udorn	Nongkhai	Surin	Korat	Ubon
> 1.5	H12 H34 H22	X X X	X X X	X X X	X X X	
> 0.7 < 1.5	h12b h34b h22b	X X X	X X X	X X X	X X X	X X X
> 0.5 < 0.7	h31b T11	X X	x x	X X	X X	X X

Discounted cash flow of 56 swim-up fry production strategies in hatcheries in Northeast Thailand after sex-reversal in hapas

- (a) mean for Nongkhai, Surin and Udorn
- (b) Ubon

(Undominated strategies are indicated)





6.5.2. Northeast Thailand farms

6.5.2.1 Calculation of Net Present Value (NPV)

Marginal analysis of discounted net returns of strategies in which the cost of fry was taken at the current price (Bt 0.1 per fish) revealed differences in undominated strategies between farms. Udon, Nongkhai and Surin farms, had opportunity costs for land (Table 6.20) that were very similar and a range of hapa systems plus the lowest cost tank treatment dominated at each of these site. On the smallest and most intensive farm, in Ubon, the strategies using large numbers of hapas in a pond area in excess of 3000 m² (total pond area) were not considered; only strategies in the lower end of the investment range were possible and the same range of tank and hapa strategies using only nine hapas (h1-3b) were undominated.

At all sites, nine hapa systems (h11b-h35b) in which no opportunity cost (see above) was accounted, dominated other tank and hapa alternatives within the same investment range (Bt 0.6-0.9 million). But if an opportunity cost were accounted with respect to hapas-in-ponds, some pond strategies became undominated as low-cost alternatives (Fig. 6.10).

6.5.2.2 Sensitivity analysis

Marginal rates of return were highly positive for increased investment over the whole range of strategies at the current price of untreated fry.

The commercial break-even price of MT-fry, i.e. the price for an IRR of 16 and 20%, respectively, for the five farms investigated varied between Bt 0.045-0.075 per fry (Appendix 4), depending on the production strategy. The break-even price of a single undominated tank strategy (T11) was higher than all others. The more intensive operations, at Korat and Ubon, had higher break-even prices than all the other operations, higher profitability of their present indicating the activities. The commercial break-even price of most of the tilapia production strategies was in the same range as the lowest farm-gate price for carp fry in the small-scale hatcheries investigated. The opportunity cost of substitution was based on farm incomes in which carp fry were sold at various prices, but always at more than Bt 0.06 and generally at more than Bt 0.1 each (Little et al., 1987).

6.6 Discussion

6.5.1 Comparison between hatchery systems

Net Present Value

In Central Thailand pond strategies are likely to be superior methods of swim-up fry production if investment cost were low i.e. not exceeding Bt 0.8 million (hormone treatment in hapas). If costs were not a constraint, the highest NPV was achievable with a hapa strategy (H34); this yielded a MRR of 272 % over the next lowest cost strategy (H23: at current fry price Bt 0.1/fry). The use of small broodstock and intensive harvest methods was most productive and had the highest NPV at any price; marginal analysis suggested additional investment gave attractive returns (MRR>40%). However, the actual price of MTfry for which there has been negative selection pressure was not possible to estimate at this point (see 6.2.1). Thus, the high fry output and NPV of strategy P23 may be linked to a poorer quality product that would eventually be balanced by a lower price and NPV. Elimination of this strategy changed the investment options in the range of low and medium investment range depending on fry price. The tank strategy that yielded the best seed production (T33) also resulted in the highest NPV but the Marginal Rate of Return compared to the next cheaper strategy was less than 40%. In the upper range of the NPV curve, investment was worthwhile for hapa systems (H34, H22) that had a high fry output for a comparatively low input in terms of costs for broodstock, labour etc.

The NPV curve identified quite different undominated strategies on Northeast Thailand farms. The lowest cost alternative produced seed in tanks (T11) and was one of the least productive tank strategies (Table 6.13). Although yield (and revenue) were lower, this was balanced by lower costs. Investment costs for swim-up fry production were lower for this treatment as only spawning tanks were required and variable costs were lower because fewer broodstock necessitated less feed, power etc. A lower output of fry considerably reduced hormone treatment costs compared to other strategies. The importance of opportunity cost on the selection of strategies compatible with carp fry production was demonstrated by the dominance of 'additive' hapa strategies in the cost range of Bt 0.5-1.0 million (Fig. 6.9). Undominated strategies (h12b, h34b, h22b) tended to have high fry output and lower costs. In the upper range of the NPV curve larger hapa systems using the same management dominated (H12, H34, H22). In Ubon, which was the smallest farm studied, investment was limited to strategies using a smaller pond area; thus strategies H12b, h34b, h22b, h31b and T11 were the most attractive.

Production costs

The analysis of investment costs for the three systems (i.e. ponds, tanks and hapas) suggested that roofing was a particularly important item for tanks and, to a lesser extent for hapa production. The overall production cost (capital cost per 1000 fry) was reduced by about Bt 1.2 per thousand fry (15-30%) for most treatments if a roof was not used.

The cost of pond maintenance, an important category of costs in ponds, was substantially increased by the use of septage. Septage is an excellent pond fertiliser except for its non-refractory nature which requires annual removal of voluminous sediments (Edwards et al., 1987). These costs could be reduced considerably by the use of chemical fertiliser and/or occasional removal of dry sediments by tractor.

Hormone treatment of fry stocked in hapas suspended in ponds compared to concrete tanks with a filtered and recirculated supply of water was a considerably cheaper option but the management limits of both of these options remain to be fully defined.

Frequent feeding and the use of a diet of high appetency are necessary management practices for successful sex-reversal which tended to increase costs of production; the required use of high stocking densities, however, could reduce production costs.

6.7.2 Comparison between production of hapa MT-treated tilapia fry in Central and Northeast Thailand

Net Present Value and investment in undominated strategies

Net Present Values for dominant strategies after MT-treatment
in hapas were higher on Central than in Northeast Thailand
farms by more than a factor of two. This was largely because
fry production was correspondingly greater since a 12 month
production period was possible in Central Thailand, compared to
a 6 month period in Northeast Thailand (Table 6.13). The NPV
curves indicated that a greater range of strategies was

dominant over the full range of potential investment in Central Thailand than in Northeast Thailand. Pond strategies dominated in the lower range of investment in Central Thailand but the high opportunity cost of land allowed more intensive methods (tanks and hapas) to dominate pond production in Northeast Thailand. The best strategies (<30% of all treatments) identified using dominance analysis indicated that investment in production of MT-treated fry could occur at levels ranging from around Bt 0.5-4.5 million, depending on the method of sex-reversal and strategy for producing swim-up fry.

Undominated strategies were not necessarily the treatments with highest fry production (e.g. T11). Based on current fry prices, marginal analysis showed that up to Bt 2 million additional investment was worthwhile on some Northeast Thailand farms. The additional initial investment required ranged from under Bt 20,000 (P23) to more than Bt 200,000 (H12) to begin production of sex-reversed fry in small-scale hatcheries in Northeast Thailand. This was within the range of capitalization of the farms' present activities (Bt 46,640 -BT 348,099 per farm; Little et al., 1987). The average initial capital requirements for large tilapia hatcheries (Area=10,000 m²) in the Philippines was in the same range (approx. Bt 360,000/farm; Yater and Smith, 1985). The initial investment costs for a start-up operation in Central Thailand ranged from less than Bt 60,000-BT 250,000 (P23 and H34, respectively).

Investment in aquaculture in this region was frequently high; Siripat et al., (1984) reported that capital investment was around Bt 560,000 for a large grow-out/fry production farm producing carps and tilapias in Supanburi, Central Thailand. Fedoruk and Srisuwantach (1984) found that initial fixed costs for cage culture of the marble goby (Oxyeleotris marmorata) ranged between Bt 26,000-BT 134,000 depending on size of operation, although this is a particularly high value fish in Thailand.

Break-even analysis indicated that the commercial break-even price of MT-treated fry was considerably lower in Central Thailand, because of the low opportunity cost of land and opportunity for year round production, than in any of the carp producing hatcheries surveyed in Northeast Thailand. A slight increase in fry price (<Bt 1.0/1000 fry) increased IRR from 16% to 20% in Central Thailand.

Similarly, on Northeast carp farms sensitivity analysis indicated considerable improvements in IRR for slight increases in fry cost. The lower cost of production from a start up operation in Central Thailand producing fry year round compared to the more seasonal production in Northeast Thailand hatcheries suggested that export of fry to Northeast from Central Thailand may be viable as was currently the case with untreated fry. At current freight rates, the cost of transport of MT-treated fry produced in Central Thailand to Northeast Thailand would add an estimated Bt 10-BT 20 per 1000 fry.

The "commercial break-even" price of MT-treated fry was lower by at least 25-70% than the current price of untreated tilapia fry over a range of undominated strategies in Central and Northeast Thailand.

Production costs

The substitution of intensive tilapia fry production for current production and revenue within Northeast carp fry hatcheries had a major affect on initial capital requirements for some of the systems, particularly those dependent on ponds (Table 6.10). The initial capital requirements for pond fry production were around 20% of those required for a full-start up operation in Central Thailand; in contrast, the initial capital costs of hapa production were reduced by only around 15-20% by substitution into a carp operation since pond capital costs were a much smaller proportion of total costs. Tank systems had similar costs at both sites - the only differences were due to the requirement for earth pond construction in the start-up operation for sex-reversal in hapas. The major difference in variable costs between a start-up operation in Central Thailand and an on-going business in Northeast Thailand was the opportunity cost of land, in particular pond space. Whereas land rental was only a minor proportion of variable costs in Central Thailand for all of the treatments (Table 6.9), it was of major importance in Northeast Thailand carp hatcheries (Table 6.11). However, if no opportunity cost were assumed for carp pond substitution (h11a-h35a) production would be additive to present carp fry production. Most of the farms surveyed in a study of small hatcheries in Northeast Thailand (Little et al., 1987) had a high ratio of broodstock to nursery pond area and broodstock were not maintained intensively.

Therefore, seed production from tilapia broodstock held in hapas suspended in ponds used for spent carp broodstock would be unlikely to have a negative impact on carp fry production. The cost of both feed and broodstock were higher in Northeast Thailand than in Central Thailand. Broodstock, even of small size, had a high opportunity cost in the Northeast Thailand and transport costs increased manufactured pelleted feed costs on average by more than 15%. However, the opportunity cost of labour was nearly 30% less in Northeast Thailand than in Central Thailand.

6.6.3 Adoption of new seed technologies by farmers

MT-treated fry have the potential to offer considerable technical and economic advantages to both fry producers and grow-out farmers but their adoption remains dependent on the latter buying the 'improved' seed in preference to obtaining fry from current sources.

The adoption of new seed by farmers is not dependent only on price and availability. Attempts to substitute improved seed for traditional varieties, which have not always been successful highlight the difficulties of technology-transfer. New seed usually require new inputs and improved management practices which may not be readily available to farmers, especially small-scale farmers.

An analogy with 'seed' in general is used because of the dearth of information on fish, or even livestock, seed adoption by farmers. Only small numbers of livestock are typically raised by farmers in Northeast Thailand and most of these are self-sustaining populations.

In general, little livestock seed is purchased but farmers do buy seed for the major arable crops. The introduction of new rice varieties in Northeast Thailand has had mixed results. It is still common for farmers to save seed of traditional rice varieties, partly because of their low cost and superior taste (Grandstaff and Grandstaff, 1986). However, a willingness by farmers to test new varieties and outlay cash for seed of cash crops has been reported for farmers in Northeast Thailand practising rice-fish culture.

Surintaraseree (1988) found a clear distinction between farmers growing fish for home consumption who tried to minimise cash outputs and those growing fish for sale in which stocking densities of fish (and hence costs) exceeded recommended levels.

The adoption of new varieties of seed by farmers as their availability increases is perhaps best illustrated by maize seed adoption which provides a comparison for the prospects of the introduction of MT-treated fry in Thailand. Both maize and tilapia are relatively recent introductions to Thailand and neither can be regarded as traditional systems in the same way as rice. The former is now grown as a cash crop on large areas of the better quality upland soils, which were previously forested, mostly for sale to distant markets. Tilapia, since its earliest introduction in the late 1940's has been promoted as a replacement for wild fish and as a source of protein for the rural poor, but it has since become important as a low cost fish for urban markets. The use of new varieties of maize in Thailand began with the Suwan varieties in the 1970's and subsequently with hybrid varieties in the 1980's; similarly Nile tilapia first introduced in 1966, became available to farmers in the early 1970's and the introduction of MT-treated fry was expected within the 1980's. The use of Suwan maize and Nile tilapia allows farmers to collect seed, which reduces costs and encourages transfer of seed from farmer-to-farmer. In general, the quality of seed declines in both over a few seasons, necessitating replacement if production is to be

maintained; contamination with other species and/or negative selection are the main causes of deterioration in tilapia stocks on farms. Despite the large numbers of fry that are produced during grow-out of normal fish, seed often needs to be purchased. In Northeast Thailand there are frequently shortages of fingerlings in rural areas and demand is high during the monsoon season. Highly seasonal rainfall causes fish ponds and rice fields to dry up, necessitating the repurchase of fry. Also predatory and competitive fish can effectively, if erratically, remove potential 'seed'.

Substitution of MT-fry for normal fry under these conditions is undoubtedly most dependent on price, and a comparison with maize is again useful. Both hybrid maize and MT-treated tilapia require repurchase of seed for each crop. Farmer adoption of hybrid maize has been relatively poor to date, probably because of its high cost relative to the improved Suwan varieties (three times). In addition, if hybrid maize yields are to improve over those of Suwan varieties, other inputs must also be increased. This is in marked contrast with MT-treated fry in which a higher value crop may be obtained for the same level of inputs (Table 6.14).

Another major deterrent to investment in higher cost maize seed, and probably also to fish seed, is the risk of environmentally induced loss. Mid-season drought is a common cause of maize failure, necessitating replanting late in the season; use of low cost seeds reduces risk compared to high cost hybrid seeds.

Similarly, fish farmers who risk partial or complete loss of their crop, especially by flooding, are probably least likely to invest in higher cost seeds. Small fish are acceptable for home consumption, which reduces the premium for MT-treated compared to normal fry.

Other possible deterrents to the adoption of new seeds include low availability and lack of assurance to farmers of the purity of the product (Grandstaff and Grandstaff, 1986). As fry, MTtreated and normal fish are not distinguishable, and similarly the slow acceptance of hybrid maize in some areas has been connected to the same problem (C.Simmonds pers. com., 1989). Nevertheless, MT-treated fry has several critical advantages over the use of hybrid maize, and the potential for its adoption by farmers, especially those raising fish as a cash crop as well as for domestic consumption, is high. Break-even analysis for MT-fry production suggested that the margin for profit was healthy for fry producers at price levels below that of the current technology. Also, many farmers are dissatisfied with the performance of the current tilapia technology, largely because of the free spawning and small size of fish at harvest, drawbacks that use of MT-treated fry can overcome. Moreover, the price of tilapia is relatively high and stable, especially as it is usually consumed locally and sold live at the farmgate or in nearby rural markets. In contrast, maize suffers large changes in price as an export crop in the international commodity market. Currently, freshwater fish in Northeast Thailand is almost a unique commodity and is contrary to almost

any other agricultural product in which the farm-gate price is lowest in the marketing chain.

6.6.4 Recommendations

- a. Models of fry production, in Central and Northeast Thailand suggest that ponds are the most profitable method of producing swim-up fry when the cost of land is low and/or the opportunity cost of carp fry production is low or non-existent. The relationship between improved fry production and a deterioration in quality of the fry, with or without hormone treatment, requires further study. The potential for more intensive conditioning of broodstock and reducing the spawning cycle to further improve productivity also warrant more investigation.
- b. The use of spawning hapas can increase productivity in terms of seed yield per unit area compared to simple release of broodstock into ponds and removal of fry by dipnetting. The spawning of broodfish at higher density in hapas may make tilapia fry production competitive with carp production under certain conditions. Whilst carp broodstock management remains extensive (=inefficient) hapa production can be a most versatile and compatible method of tilapia fry production within a hatchery producing both groups of fish. A decline in the profitability of carp fry production, or conversely an increase in the price of tilapia fry, could stimulate the use of hapas for tilapia fry production especially if they were managed to limit broodstock exchange to spawned and unripe fish model (H34).

The relationship between broodstock density and production needs further study, as does the management of water quality for sustained hapa production. The addition of tilapia fry production hapas into carp broodstock ponds without any apparent detrimental effect on the latter requires further confirmation.

c. The use of concrete tank systems (T1-T3) for tilapia fry production is only recommended, for situations when demand is relatively low, carp production is intensive and efficient, and/or land is limiting. Non-exchange of tank fish or exchange with selected fish conditioned in hapas gave the best returns in this study but exchange of spawned and unripe broodfish only might further improve returns. In areas where the market for fry is limited or the market for carp saturated, tank production of tilapia fry is the best option.

CHAPTER 7

CONCLUSIONS

The mass production of <u>Oreochromis niloticus</u> fry suitable for hormonal sex-reversal was found to be possible and economically viable using a variety of systems. The application of laboratory techniques such as early egg robbing from the brooding mother and artificial incubation was found to be possible for larger, commercial-scale units. The importance of broodfish conditioning and frequent harvest of seed was shown for intensive tank and hapa systems. The following specific conclusions can be made:

- (1) Regular disturbance and harvesting of seed after a short period of spawning opportunity (5-10 days) was found to increase seed production in concrete spawning tanks. Exchange of female broodfish, both partial and total, increased synchrony of breeding.
- (2) A change in conditioning and spawning environment had no effect on seed yield from spawning tanks and hapas (area =12.57 m² and 40m² respectively). Seed wet weight, seed clutch size and weight was greater in female fish spawned in tanks than hapas. Females conditioned in hapas however produced heavier seed clutches of larger absolute and relative size than tank conditioned fish.
- (3) Records of tagged females indicated considerable differences in the frequency of spawning; in hapas, the distribution was normal whereas in tanks it was skewed. The

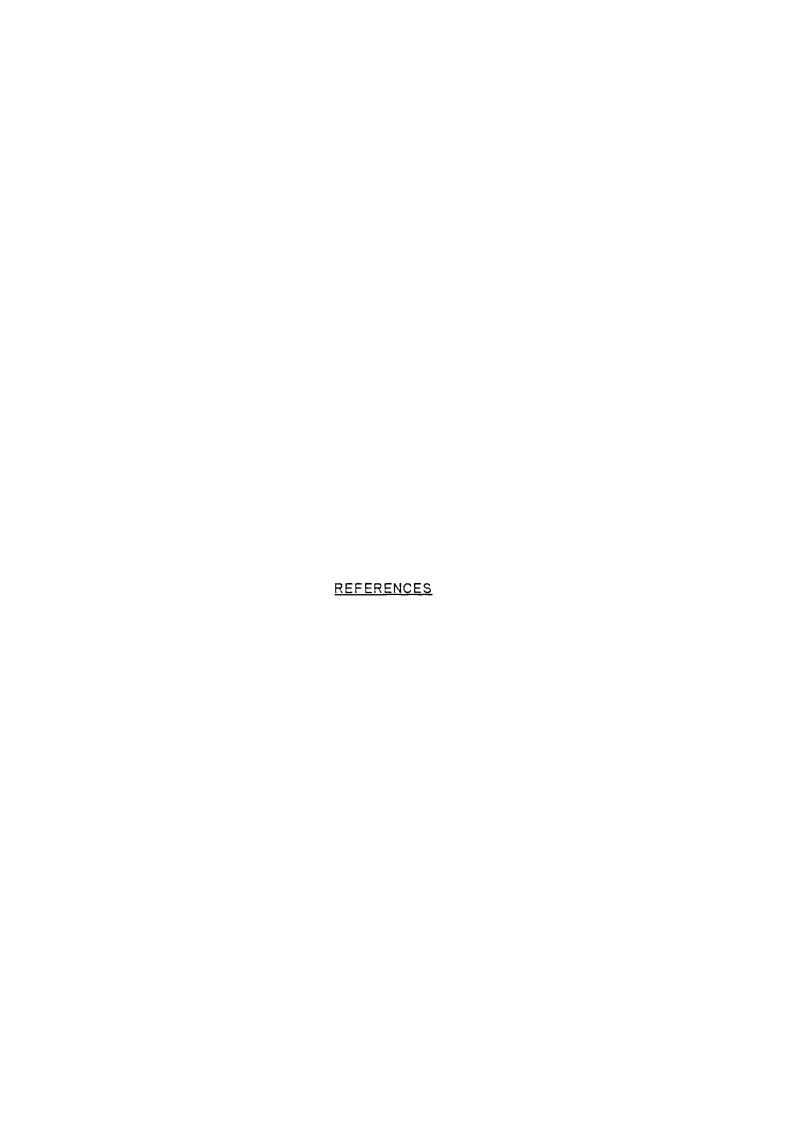
evidence suggests that hierarchy is important in the control of reproduction and exerts it's strongest effect in clear water, densely stocked tanks.

- (4) Selective female broodfish exchange optimised seed yield per unit weight of broodfish and seed production was not improved by conditioning females for periods longer than 10 days. Male broodfish exchange did not significantly improve (P > 0.05) seed yields.
- (5) Early nutrition of broodfish raised under different supplemental feeding regimes in fertilised earthen ponds had a significant effect on later spawning frequency in concrete tanks. However, this effect was confined to broodfish maintained at densities lower or higher than optimal for seed production.
- (6) Broodfish stocked over a range of densities for extended periods (201 days) showed greater variability of seed production in hapa than tank production systems. This was mainly due to periods of poor water quality in hapas; when water quality was high seed production was significantly higher in hapas than tanks over a range of broodfish densities.
- (7) The relationship between seed production and broodfish density over time suggested that both stocking biomass and number have an effect on fry output. Density of broodfish

showed an inverse relationship to clutch size in both tanks and hapas and synchrony of spawning in tanks.

- (8) Production of swim-up fry in large earthen ponds $(area=1740m^{2})$ was not significantly different (P>0.05) at two levels of harvest intensity.
- (9) The use of small broodfish produced double the yield of hormone treatable fry than a similar biomass of larger broodfish of the same cohort but the use of such fish by hatchery operators constitutes a form of negative selection and is therefore not to be advised.
- (10) A commercial scale incubation system, of capacity >100,000 eggs/seed was constructed from easily available materials and found to be suitable for hatching the seed harvested from both tanks and hapas. A mean survival of 75% of all harvested seed to swim-up fry was obtained over several trials.
- (11) A trend to intensification (fry/m²/day) from ponds to hapas to tanks was evident when yields of swim-up fry are compared. Productivity exceeded any in the published literature for comparable systems, largely because of the intensity of broodstock management and early and efficient harvest of seed. Broodfish productivities (fry/kg female/month) were also higher across the range of systems tested often by a factor of 1.5-3.

(12) Economic analysis suggested that for a start-up operation in Central Thailand fry production in earthen ponds can give the best return on levels of investment of less than Baht 0.8 million. Substitution of techniques into current carp fry production operations in Northeast Thailand indicated that more intensive methods (production in tanks and hapas) are more attractive over a range of investment levels. The break-even price of MT-treated fry after hormone treatment in nylon hapas was approximately half the cost of treatment in a recirculated water concrete tank system. The break-even price in Central Thailand was lower than the Northeast by a factor of around 1.5 but the break-even price for both areas was lower than the current price of untreated Oreochromis fry.



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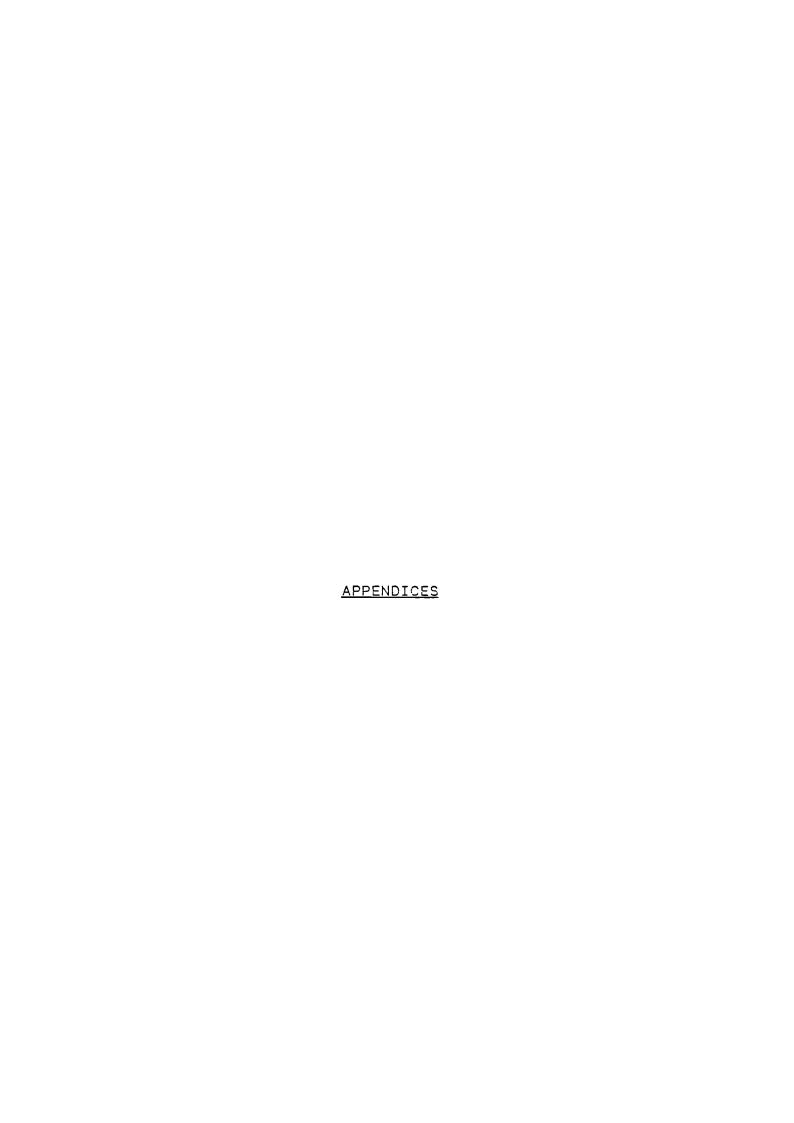
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Appendix 1

- 1.1
- Temperature of pond water: measured daily with a maximum/minimum thermometer suspended in the water column at a depth of 30 cm.
- 1.2

Dissolved oxygen of pond water: measured weekly at dawn by the azide modification of the iodometric (Winkler) titration method.

1.3

pH of pond water: measured weekly at dawn using a pH meter Pye model 290.

1.4

Ammonia: indophenol method described by Strickland and Parsons (1972) was used to determine ammonia in pond water weekly (Experiment 2 only).

1.5

Phytoplankton biomass: from samples taken at dawn in the midwater column phytoplankton biomass was estimated from its relationship to chlorophyll a. The chlorophyll a content was first determined by the extraction of pigment with 90% acetone solution, maceration and centrifugation of water samples. The absorbance of the clarified extract was measured by a Superscan UV-Visible Spectrophotometer model 3.; chlorophyll a content (mg/m²) was obtained from a calibration curve of chlorophyll a and absorbance. The phytoplankton biomass was subsequently estimated by multiplying the chlorophyll a content by a factor of 67 (Experiment 2 only).

1.6

Moisture content of fish carcasses and eggs: fish carcasses and whole ovaries dissected from the body were dried at 80° C to constant weight.

1.7

Crude lipid of fish carcasses and eggs: samples were ground up (after moisture content analysis) and the lipid extracted with diethyl ether for 6 hours in a Soxtec system HT2 (Tecator) 1045 extraction unit and 1044 service unit.

1.8

Crude protein of fish carcasses and eggs: total Kjeldahl nitrogen of fish carcasses and whole ovaries dissected from the body was estimated using a Kjeltec system (Tecator) 1028 distilling unit and digestion system 12 (Tecator) 1009 digestor. Crude protein was calculated from the relationship: Crude Protein = Total Kjeldahl Nitrogen x 6.25.

1.9

Ash determination of fish carcasses and eggs: carcasses and whole ovaries dissected from the body were ignited in a muffle furnace at 600-650 $^{\circ}\text{C}$ for 4 hours (AOAC 1980), following moisture analysis.

<u>Appendix 2</u>

Appendix 2.1 Total biomass of female broodfish in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^2) over experimental period (201 days, T2)

Appendix 2.2 Total biomass of male bropdfish in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^4) over experimental period (201 days, T3)

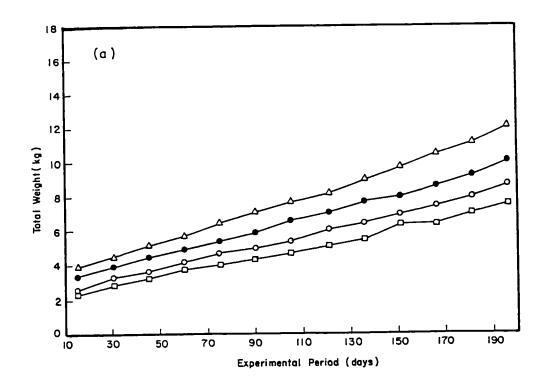
Appendix 2.3 Density of female broodstock in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^2) over experimental period (201 days)

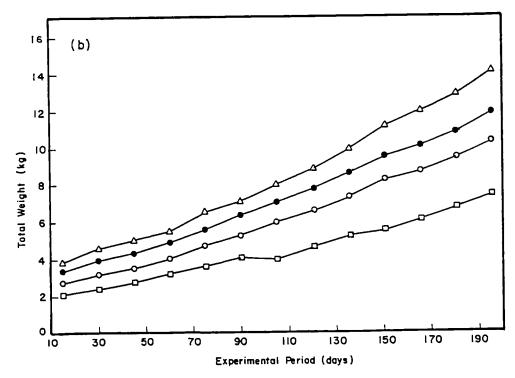
Appendix 2.4 Total ammonia levels (mg/l) in (a) hapa systems i.e. spawning hapas (square), conditioning hapas (cross) and outside hapas (diamond); (b) conditioning tanks (Area = 1.96 m^2 ; mean of all treatments, T3) and (Area = 40 m^2 , 5.4 m^2 and 1740 m^2 respectively H3); samples collected weekly at 09.00 a.m.)

Appendix 2.5 pH in (a) spawning hapas (square), conditioning hapas (triangle) and outside hapas (circle) and (b) spawning tanks over the experimental period (201 days) measured from samples collected weekly at 09.00 a.m. (T3 and H3)

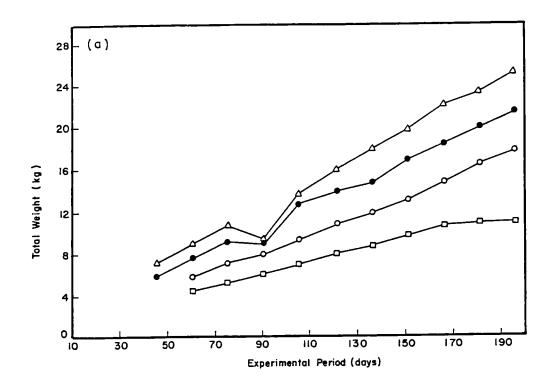
Appendix 2.6 Five-day mean maximum and minimum temperature in (a) spawning tanks (b) spawning hapas measured daily at 20 cm below the water surface (Mean of 2 readings daily, T3 and H3)

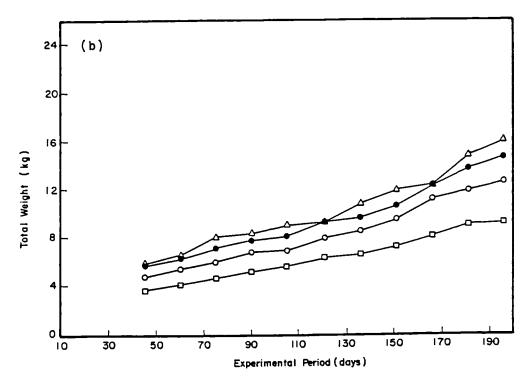
Appendix 2.7 Chlorophyll a levels in hapa systems i.e. spawning hapas (square), conditioning hapas (triangle) and outside hapas (circle) (Area = 40 m⁻, 5.4 m⁻ and 1740 m⁻ respectively, T3 and H3); samples collected weekly at 09.00 a.m.)



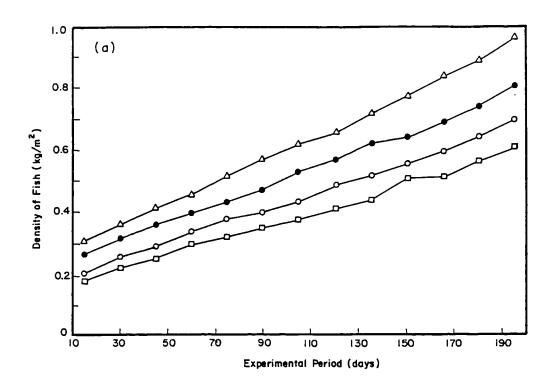


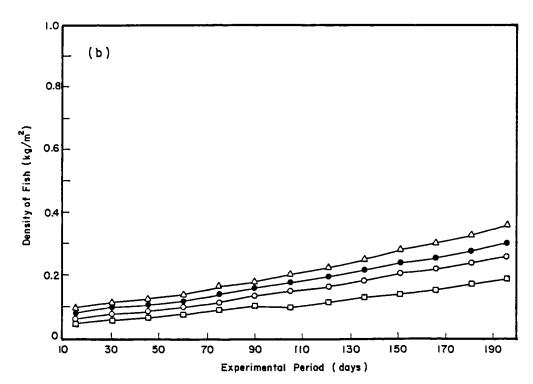
2.1 Total biomass of female broodfish in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^2) over experimental period (201 days, T3)



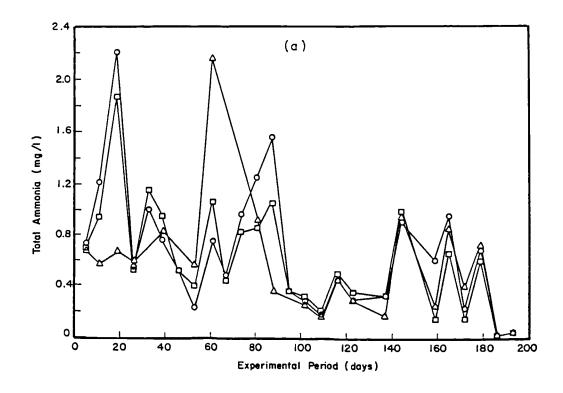


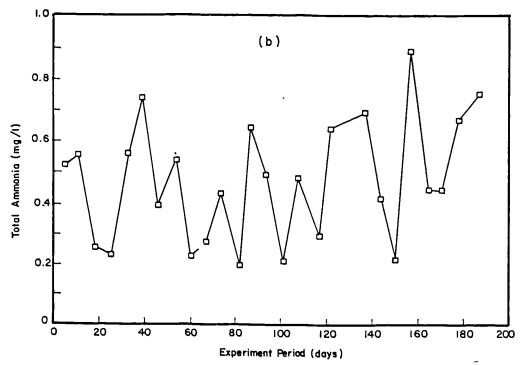
Total biomass of male broodfish in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^2) over experimental period (201 days, T3)



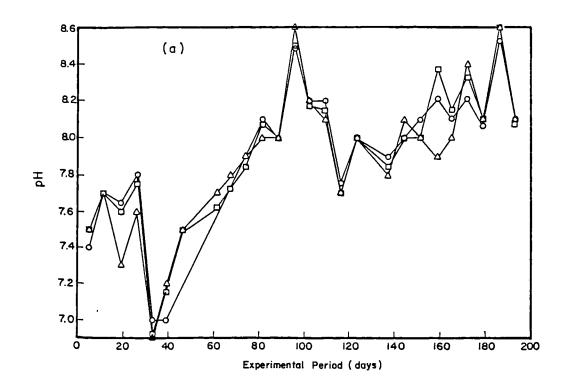


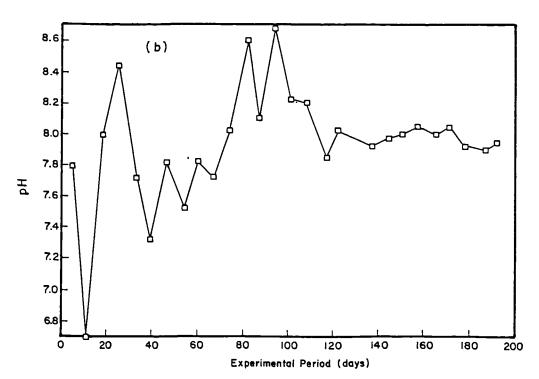
2.3 Density of female broodstock in (a) tanks (Area = 12.57 m^2) and (b) hapas (Area = 40 m^2) over experimental period (201 days)



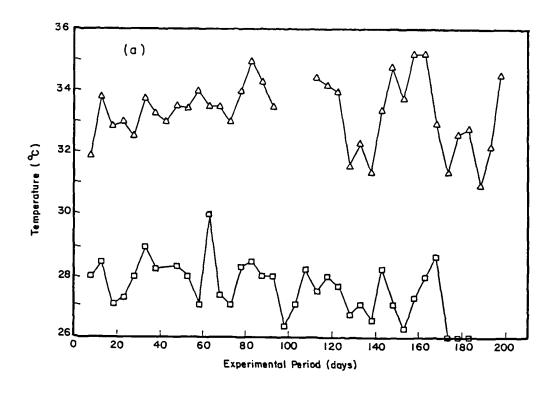


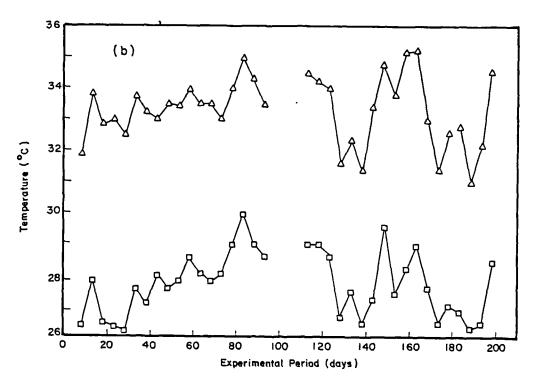
2.4 Total ammonia levels (mg/l) in (a) hapa systems i.e. spawning hapas (square), conditioning hapas (cross) and outside hapas (diamond); (b) conditioning tanks (Area = 1.96 $\rm m^2$; mean of all treatments, T3) and (Area = 40 $\rm m^2$, 5.4 $\rm m^2$ and 1740 $\rm m^2$ respectively H3); samples collected weekly at 09.00 a.m.)



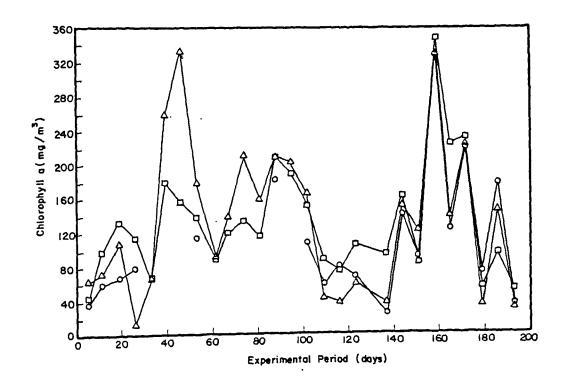


pH in (a) spawning hapas (square), conditioning hapas (cross) and outside hapas (circle) and (b) spawning tanks over the experimental period (201 days) measured from samples collected weekly at 09.00 a.m. (T3 and H3)





2.6 Five-day mean maximum and minimum temperature in (a) spawning tanks (b) spawning hapas measured daily at 20 cm below the water surface (Mean of 2 readings daily, T3 and H3)



Chlorophyll a levels in hapa systems i.e. spawning hapas (square), conditioning hapas (triangle) and outside hapas (circle) (Area = 40 m², 5.4 m² and 1740 m² respectively, T3 and H3); samples collected weekly at 09.00 a.m.)

<u>Appendix 3</u>

Notes to Determination of Costs

- Opportunity cost of land: Based on gross margin for rice production in Pathum Thani, Central Thailand. Variable costs per hectare Bt 10,000 (Gallo, 1988), at 4 t/ha yield, 2 crops per year, at Bt 3.5/kg (Cost: Bt 0.8/m²/yr)
- 2. Land Tax: For land used for agricultural purposes effectively zero (approximately Bt 1/rai/yr)
- Field cost of floating catfish pellet (30% crude protein): Bt 12/kg
- 4. Field cost of broodstock: Bt 10/kg
- 5. Extra Broodstock: The weight of extra broodstock added throughout the duration of the experiment to replace mortalities and/or escaped fish
- 6. Labour requirement for feeding fish estimated at 15 min/tank/day; Bt 70 per man-day, 1 working day = 8 hours
- 7. Labour requirement for harvesting seed estimated at 1 man-day per 4 tanks, 13 harvests in 125 days. Treatment 3 required approximately 12 minutes per day harvest time (3.2 man-days)
- 8. Electricity cost: Commercial rate Bt 2 per kw hour
- 9. Specifications of electrical pump used in spawning tank system. Capacity 0.12 m²/min submersible 2" Tsurumi; electrical rating 0.4 kw. Over a 24 hour period one pump supplies 8 tanks (4 m diameter); or Bt 20/day/tank over a 125 day period
- 10. Pump as (9) for conditioning tank system (16 tanks
 1.5 m diameter)
- 11. Pump as (9) for incubation system can supply up to 20 incubators (20 1 capacity) or one swim-up system
- 12. Cost of hatchery roof Bt 450/m. Zinc roofing on steel uprights, cost of labour and paint inclusive. Area over tanks only is covered (not filter and reservoir)
- 13. Fill sand used over whole hatchery site, 0.15 m depth, Bt 120/m
- 14. Gravel filled in hatchery areas not covered by

- 15. A reinforced concrete base of hatchery (concrete:sand:gravel 1:2:4) of 0.1 m thickness was used. To make 1 m of concrete, 325 kg of cement (Bt 553), 650 kg sand (Bt 60) and 1300 kg of gravel (Bt 200) are required (Edwards et al, 1988). Total cost of concrete is Bt 813/m Cost of steel reinforcement Bt 500/m concrete Cost of labour = 30% cost of concrete (Bt 244/m)
- 16. Pipes and valves in hatchery of local manufacture. Pipes are high grade PVC purchased in 4 m long units. Quantities as used in AIT hatchery. Renewal of pipes and valves required every 15 years. Cost of glue 1.3% of pipe cost, cost of labour 25% of material cost
- 17. Spawning and conditioning tank design (Fig. 2.3).
 Underground filter (Fig. 2.4) filled with heterogeneous waste plastic material. Cost Bt 8/kg unwashed.
- 18. Accessory equipment and incubation facilities (Figs. 5.2 & 5.2)
- 19. Design of slow sand filter (Fig. 5.1), 5 m² surface area. Total cost including labour and materials from AIT hatchery (Bt 12,000)
- 20. Design of swim-up system (Fig. 5.1)
- 21. Earthen pond construction costs based on a total of Bt 22,400 to construct a pond (102m x 22m x 1.5m), excavating 755 m of earth. Bulldozer excavation (Bt 4,000 /day) plus extra labour for preparing and finishing the pond is included; pond construction costs based on Bt 29.7/m. Even when pond is not required, fill is still removed for filling hatchery site to 1 m. When a pond is required for hatchery function, earth excavated is enough to construct a pond that will hold water to 1.5 m (i.e. both excavation and pond dyke building)
- 22. Physical contingencies budgeted at 5%.
- 23. Conditioning nets (3 m x 1.5 m x 0.9 m) are cleaned in the same time as small conditioning tanks
- 24. Maintenance cost of pond per unit area per year for an earthen pond receiving septic tank slurry is Bt6.6/m². Nylon cages estimated to require a pond water area 4 times the actual cage area, i.e. a net of 40 m² requires 160 m² of pond surface.

- 25. Purchase cost of conditioning hapa, nylon rope and bamboo poles is Bt 200 + Bt 15 + Bt 18, respectively
- 26. The cost of harvesting hapas is approximately the same as the cost of harvesting and cleaning tanks. Cost of cleaning hapas is Bt 14 per net; over a period of 20 harvests; hapas are cleaned 10 times
- 27. Incubation and swim-up system for an 18 hapa system, require 2 and 1 submersible pumps, respectively
- 28. Unit cost of large hapa with ropes bamboo poles is Bt 871. Duplicate set of nets required for cleaning purposes
- 29. Harvest and incubation equipment to service a 18 hapa system
- 30. Cleaning cost as (23). Harvesting cost dependent on density of fish i.e.

total cost x no.of fish in treatment total no. of fish in experiment

- 31. Annual output of eggs from 8 tank system based on a 360 day period
- 32. Residual broodstock sold off as table fish after each experimental period
- 33. Annual output of eggs from 16 hapa system based on a 335 day period (30 days per year pond cleaning).
 18 hapa system based on 2 x 1,740 m² pond hapa capacity. Two ponds required to allow continual production
- 34. In earthen ponds 3 x 116 day total cycle of production
- 35. Pumping costs in earthen ponds using 9 HP gasoline pump and 6" pipe.Oil and gasoline Bt 50 per 300 m, Bt 0.1724/m respectively. Water is pumped in and out of the pond. Assume that ponds are constructed proximal to an irrigation supply canal and that there is no requirement for constructing either a discharge or supply canal
- 36. Sludge removal in earthen ponds (Edwards et al., 1988) requires 55 man-days labour per 1,740 m² pond and Bt 370 worth of fuel. Total cost Bt 4,220 in 60 man-days (2 ponds 120 man-days)
- 37. Septic tank slurry required in earthen ponds: 2 m² per two pond system per day
- 38. General maintenance required year round in earthen

- ponds, 2 man-days per week
- 39. Potassium cyanide used in earthen ponds at cost Bt 49 per pond
- 40. In earthen ponds brood purchased before each cycle of 3 months and sold immediately after draining in earthen pond production

Appendix 4

Break-even price of MT-fry produced using undominated strategies on five small-scale hatcheries in Northeast Thailand

Break-even price at 2 different IRR's (Baht/1000 fry)											
HATCHERY		UBCN		NC	I (JDORN	SURIN		<u>+</u>	KORAT	
	1	16%	20%	16%	20%	16%	20%	16%	20%	t 169	% 20%
Strateg	У										
T11	73.	5 8	3.5	76.4	31.5	76.4	81.5	76.7	81.5	76.7	31.5
h31b '	59.	5 6	1.3	61.0	61.8	61.0	61.8	57.3	58.3	57.5	59.2
H22	[-		-	60.5	61.6	60.5	51.6	57.2	58.3	59.5	60.8
h22b	53.	3 5	5.4	50.8	52.5	50.8			56.3		
H34	-		-	50.8	51.5	50.8	51.5	50.8	52.5	53.5	54.3
H12	-		_	49.4	50.0	49.4	50.0	49.6	50.5	51.4	52.8
h34b	50.	3 5	1.3	46.2	47.4	46.2	47.4	46.2	47.3	46.8	48.0
h12b	48.	5 4	9.5	45.3	46.4	45.3	46.4	45.5	46.5	46.0	47.2