Functional Ecology:

Ecologically relevant radiation exposure triggers elevated metabolic rate and nectar consumption in bumblebees

Burrows, Jessica E.*[1]; Copplestone, David[1]; Raines, Katherine E.[1], Beresford, Nicholas A.[2]; Tinsley, Matthew C.[1]

Supplementary Materials

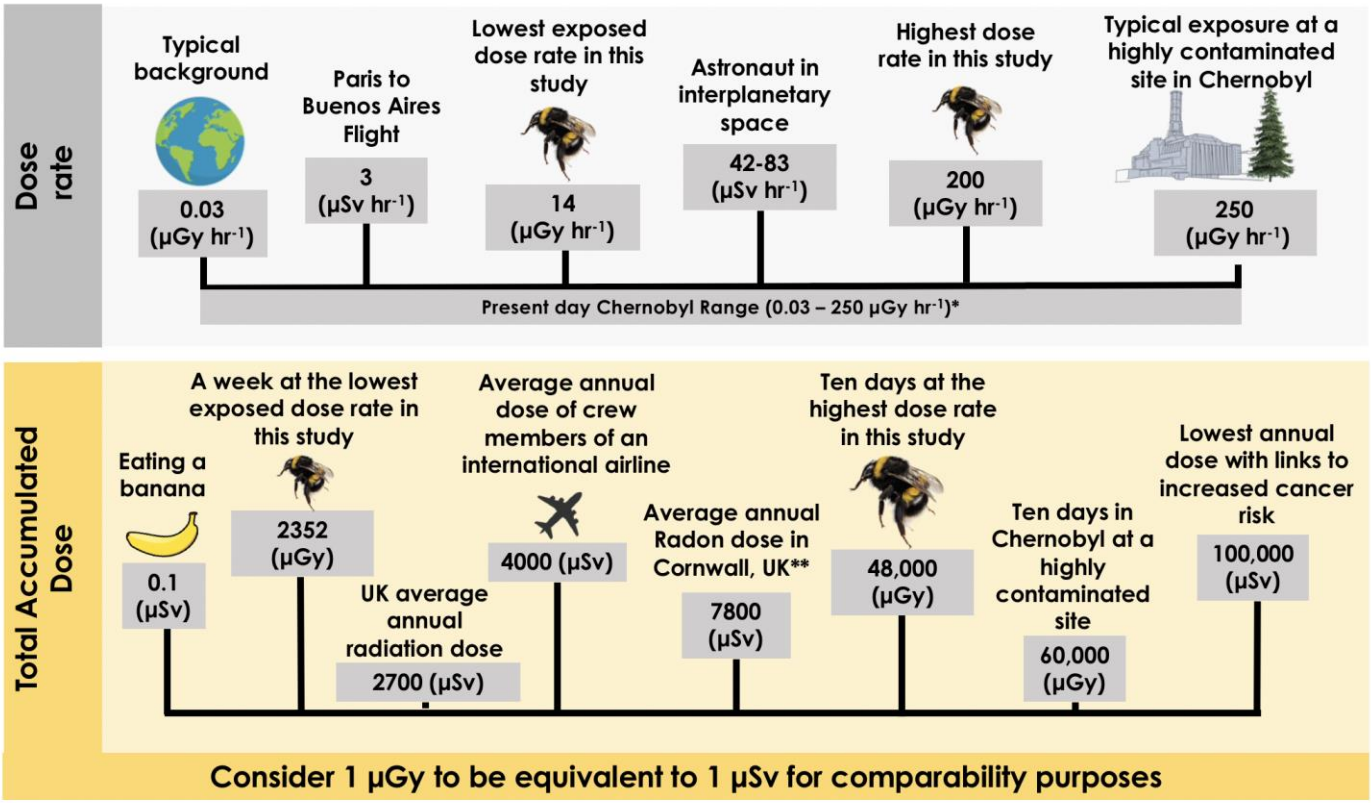
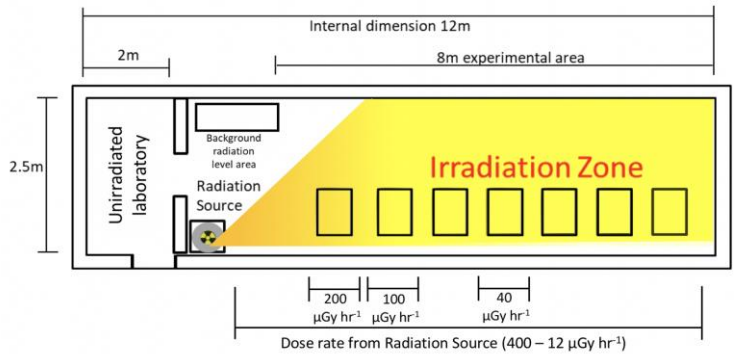
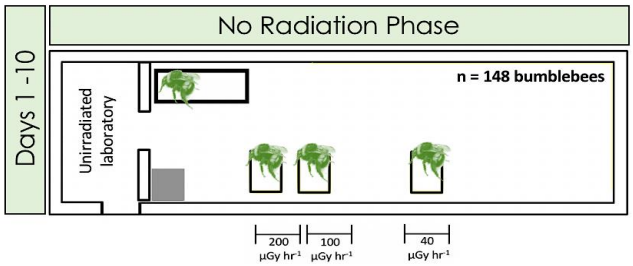


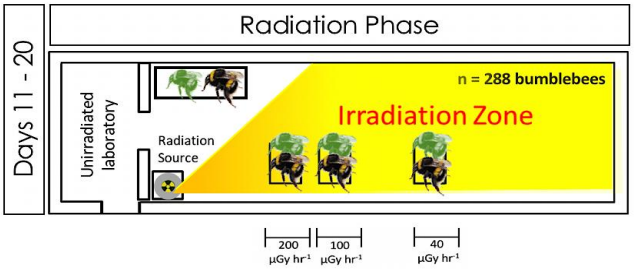
Figure S1: A diagram of various dose rates experienced in our natural environment in comparison with dose rates used in experiment 1 and 2 (ANSTO., 2022 ; Beresford *et al*., 2020 ; Chancellor *et al.*, 2018)



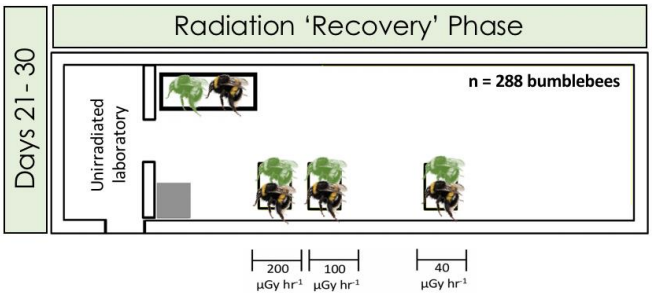
Experiment 1



- Radiation source is shielded.
- Nectar consumption was measured every 2 days.

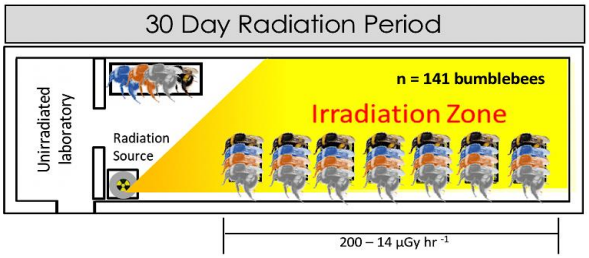


- Further 140 bees are added and then radiation source unshielded.
- Nectar consumption was measured every 2 days.
- Metabolic rate and activity were measured on days 7 and 9.



- Radiation source is shielded again.
- Nectar consumption was measured every 2 days.
- Metabolic rate and activity were measured on days 17 and 19.

Experiment 2



- Nectar consumption and bumblebee mass was measured every 2 days for 30 days.
- Four bees were placed at each dose rate and assigned a feeder of either 20%, 30%, 40% or 50% sucrose.

Figure S2: A diagram of the radiation facility at the University of Stirling**.** The top image represents the radiation facility with dimensions. The subsequent diagrams represent the two experiments and their design. For experiment 1: black boxes represent shelving units on which bumblebees were placed. Green bumblebees

represent those that entered the experiment on day 1 and black bumblebees represent those that entered in the radiation phase. For experiment 2: the different coloured bees represent each of the four sucrose feeding treatments of 20%, 30%, 40% and 50%.

Experiment 1: The effect of radiation on bumblebee nectar consumption

Table S1. Parameter estimates for models investigating the effect of the position of a bumblebee in the experimental facility for 10 days prior to radiation exposure (pre-radiation phase) on consumption of 40% sucrose nectar solution. Future dose rates were 200, 100, 40 μGy hr^{-1} and controls. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 148 bumblebees during these observations. Table S1a describes the minimal model used. Table S1b contains terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	0.55	0.01	-	-
Days within the experiment	-0.01	1.37×10^{-3}	21.19	4.17×10^{-6}
Mass of bee at start of experiment (g)	0.02	0.01	4.93	0.03
Average temperature during the days when the nectar measurements were made (°C)	0.28	0.07	14.38	1.50×10^{-4}
Average humidity during the days when the nectar measurements were made (%)	0.01	2.42×10^{-3}	11.18	8.26×10^{-4}
b. Terms removed from model in reverse order of deletion				
Future dose rate (μGy hr^{-1})	-1.16×10^{-4}	9.81×10^{-5}	1.44	0.23
Access to a second low nectar concentration feeder	1.23×10^{-2}	1.38×10^{-2}	0.82	0.36
Age of bee at start of experiment (days)	3.54×10^{-3}	6.05×10^{-3}	0.35	0.55
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	4.98×10^{-2}	4.01×10^{-2}	1.56	0.21
Future dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	8.30×10^{-5}	8.91×10^{-5}	0.91	0.34
Future dose rate (μGy hr^{-1}) by mass of bee (g)	-1.30×10^{-4}	9.70×10^{-5}	1.89	0.17
Future dose rate (μGy hr^{-1}) by days in the phase	-5.35×10^{-6}	1.80×10^{-5}	0.09	0.77

Table S2. Parameter estimates for models investigating the effect of radiation dose rate on bumblebee nectar consumption (of 40% sucrose solution) during the 10-day radiation phase of the experiment. Dose rates were 200, 100, 40 μGy hr^{-1} and controls. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 288 bumblebees during these observations. Table S2a describes the minimal model used. Table S2b contains terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	13.30	0.43	-	-
Days within the experiment	2.20	0.06	23.66	1.15×10^{-6}
Dose rate (μGy hr^{-1})	0.10	0.15	39.74	2.90×10^{-10}
Mass of bee at start of experiment (g)	7.34	1.47	16.94	3.85×10^{-5}
Average temperature during the days when the nectar measurements were made (°C)	118.00	2.16	9.07	2.00×10^{-3}
Average humidity during the days when the nectar measurements were made (%)	2.38	0.03	7.36	7.00×10^{-3}
Dose rate (μGy hr^{-1}) by days within the experiment	0.02	0.01	38.25	6.22×10^{-10}
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	45.500	10.02	3.97	0.05
b. Terms removed from model in reverse order of deletion				
Access to a second low nectar concentration feeder	-1.97×10^{-2}	1.48×10^{-2}	1.78	0.18
Age of bee at start of experiment (days)	-1.10×10^{-3}	6.33×10^{-3}	0.03	0.86
Dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	5.65×10^{-5}	8.62×10^{-5}	0.45	0.50
Dose rate (μGy hr^{-1}) by mass of bee (g)	-3.84×10^{-5}	1.00×10^{-4}	0.14	0.70
Dose rate (μGy hr^{-1}) by average temperature during the days when the nectar measurements were made (°C)	3.28×10^{-3}	1.40×10^{-3}	0.55	0.19
Dose rate (μGy hr^{-1}) by average humidity during the days when the nectar measurements were made (%)	-6.61×10^{-6}	4.32×10^{-5}	0.02	0.89

Table S3. Parameter estimates for models investigating the effect of radiation dose rate on bumblebee nectar consumption (of 40% sucrose solution) during the 10-day radiation phase of the experiment excluding the top dose rate of 200 μGy hr^{-1}. This model is for dose rates of 100, 40 μGy hr^{-1} and controls. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 213 bumblebees during these observations. Table S3a describes the minimal model used. Table S3b contains terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	$4.93x10^{-1}$	$1.55x10^{-2}$	-	-
Dose rate (μGy hr^{-1})	$7.15x10^{-4}$	$2.32x10^{-4}$	12.27	$1.00x10^{-3}$
Mass of bee at start of experiment (g)	$3.36x10^{-2}$	$9.14x10^{-3}$	13.26	$2.00x10^{-3}$
Average humidity during the days when the nectar measurements were made (%)	$-1.06x10^{-2}$	$2.39x10^{-3}$	19.52	$9.93x10^{-6}$
b. Terms removed from model in reverse order of deletion				
Access to a second low nectar concentration feeder	$-1.76x10^{-2}$	$1.80x10^{-2}$	0.97	0.33
Days within the experiment	$-9.88x10^{-4}$	$1.32x10^{-3}$	0.52	0.47
Age of bee at start of experiment (days)	$-6.02x10^{-3}$	$7.56x10^{-3}$	0.66	0.42
Average temperature during the days when the nectar measurements were made (°C)	$6.59x10^{-2}$	$1.23x10^{-1}$	0.30	0.58
Dose rate (μGy hr^{-1}) by days within the experiment	$-1.57x10^{-4}$	$1.20x10^{-4}$	0.63	0.43
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	$3.11x10^{-2}$	$5.13x10^{-2}$	0.38	0.54
Dose rate (μGy hr^{-1}) by mass of bee (g)	$-1.23x10^{-4}$	$2.17x10^{-4}$	0.34	0.56
Dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	$-8.30x10^{-6}$	$3.34x10^{-5}$	0.07	0.79

Table S4. Parameter estimates for models investigating the effect of radiation dose rate on bumblebee nectar consumption (of 40% sucrose solution) during the 10-day radiation phase of the experiment at a doses rate of 40 μGy hr^{-1} and controls. Dose rates of 200 and 100 μGy hr^{-1} were removed. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 146 bumblebees during these observations. Table S4a describes the minimal model used. Table S4b contains terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	0.51	0.01	-	-
Days within the experiment	-3.00×10^{-3}	1.00×10^{-3}	4.93	0.03
Mass of bee at start of experiment (g)	0.03	0.01	6.51	0.01
Average humidity during the days when the nectar measurements were made (%)	-0.01	3.00×10^{-3}	4.23	3.00×10^{-3}
b. Terms removed from model in reverse order of deletion				
Average temperature during the days when the nectar measurements were made (°C)	-0.24	0.14	2.76	0.10
Dose rate (μGy hr^{-1})	0.03	0.02	1.51	0.22
Age of bee at start of experiment (days)	-6.00×10^{-3}	8.00×10^{-3}	0.57	0.45
Access to a second low nectar concentration feeder	-7.00×10^{-3}	0.02	0.13	0.72
Dose rate (μGy hr^{-1}) by days within the experiment	-0.03	0.02	2.81	0.09
Dose rate (μGy hr^{-1}) by mass of bee (g)	-0.02	0.02	0.49	0.48
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	0.03	0.07	0.18	0.67
Dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	-5.94×10^{-4}	3.55×10^{-3}	0.009	0.92

Table S5. Paired t-tests conducted to assess changes in bumblebee nectar consumption (40% sucrose) between day 10 at the end of the no radiation phase and two days later, after two days of exposure during the radiation phase. A total of 295 bees were measured on day 10 and 12.

Dose rate (μGy hr^{-1}) group	**Mean difference**	**df**	**T**	**P value**
200	-4.00×10^{-3}	74	-0.18	0.86
100	-0.03	73	-1.24	0.23
40	0.04	71	1.89	0.07
0.11	0.03	73	1.44	0.16

Table S6. Parameter estimates for models investigating the effect of radiation dose rate on bumblebee nectar consumption from the 5% nectar solution during the 10-day radiation phase of the experiment. Dose rates were 200, 100, 40 μGy hr^{-1} and controls. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 144 bumblebees during these observations. Table 6a describes the minimal model used. Table S6b contains terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	0.60	0.04	-	-
b. Terms removed from model in reverse order of deletion				
Days within the experiment	0.02	0.01	1.61	0.21
Average temperature during the days when the nectar measurements were made (°C)	-1.54	0.96	1.61	0.20
Mass of bee at start of experiment (g)	0.04	0.04	0.86	0.35
Dose rate (μGy hr^{-1})	-2.23×10^{-4}	3.98×10^{-4}	0.37	0.54
Average humidity during the days when the nectar measurements were made (%)	3.27×10^{-3}	1.33×10^{-2}	0.06	0.80
Age of bee at start of experiment (days)	-4.17×10^{-4}	0.04	0.01	0.94
Dose rate (μGy hr^{-1}) by mass of bee (g)	-5.77×10^{-4}	4.50×10^{-4}	1.98	0.16
Dose rate (μGy hr^{-1}) by average humidity during the days when the nectar measurements were made (%)	-1.55×10^{-4}	1.32×10^{-4}	1.96	0.16
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	0.33	0.25	2.26	0.13
Dose rate (μGy hr^{-1}) by days in the phase	9.90×10^{-5}	1.26×10^{-4}	0.76	0.38
Dose rate (μGy hr^{-1}) by average temperature during the days when the nectar measurements were made (°C)	-0.01	0.01	0.34	0.56
Dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	-3.9×10^{-7}	3.98×10^{-4}	0.01	0.90
Access to a second-high nectar concentration feeder	1.57×10^{-2}	4.48×10^{-2}	0.03	0.10

Table S7. Parameter estimates for models investigating the effect of previous radiation dose rate on bumblebee nectar consumption (40%) during the recovery phase of the experiment. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 288 bumblebees during these observations. Table S7a describes minimal model used. Table S7b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	404.00	16.10	-	-
Days within the experiment	15.00	3.00	24.44	7.66×10^{-7}
Prior dose rate received (μGy hr^{-1})	0.66	0.14	21.35	3.84×10^{-6}
Mass of bee at start of experiment (g)	28.00	9.75	8.24	4.00×10^{-3}
Average temperature for days leading up to nectar measurements at the data logger closest to the bumblebee (°C)	-53.50	67.70	0.63	0.43
Average humidity for days leading up to nectar measurements at the data logger closest to the bumblebee (%)	-15.50	3.68	17.70	2.59×10^{-5}
Prior dose rate received (μGy hr^{-1}) by days within the experiment	0.08	0.02	12.48	4.11×10^{-4}
Average temperature (°C) by humidity levels (%) for days leading up to nectar measurements at the data logger closest to the bumblebee	-61.50	30.20	4.17	0.04
b. Terms removed from model in reverse order of deletion				
Access to a second low nectar concentration feeder	-23.5	19.9	1.42	0.23
Age of bee at start of experiment at start of experiment (days)	3.93	8.39	0.23	0.63
Prior dose rate received (μGy hr^{-1}) by mass of bee (g)	0.04	0.14	0.09	0.77
Prior dose rate received (μGy hr^{-1}) by age of bee at start of experiment (days)	-0.02	0.12	0.03	0.86

Table 8. Parameter estimates for models investigating whether the effect of radiation on bumblebee nectar consumption (40%) changed across the radiation and recovery phases of the experiment. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 288 bumblebees during these observations. Table S8a describes minimal model used. Table S8b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	539.00	26.00	-	-
Days within the phase of the experiment	8.92	1.31	45.67	1.40×10^{-11}
Dose rate / Prior dose rate (μGy hr^{-1})	0.64	0.10	39.45	3.37×10^{-10}
Mass of bee at start of experiment (g)	31.70	7.33	18.25	1.94×10^{-5}
Average temperature during the days when the nectar measurements were made (°C)	121.00	55.40	4.77	0.03
Change from radiation to recovery phase	-108.00	12.00	91.93	$<2.2 \times 10^{-16}$
Average humidity during the days when the nectar measurements were made (%)	-8.82	1.68	27.31	1.74×10^{-7}
Dose rate / Prior dose rate (μGy hr^{-1}) by days within phase	0.12	0.02	58.95	1.62×10^{-14}
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	46.90	16.00	8.59	3.00×10^{-3}
Days within the phase by recovery phase	6.98	3.00	5.35	0.02
b. Terms removed from model in reverse order of deletion				
Access to a second low nectar concentration feeder	-25.40	14.70	3.04	0.08
Age of bee at start of experiment (days)	2.40	6.27	0.15	0.70
Dose rate / Prior dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	0.04	0.09	0.19	0.66
Dose rate / Prior dose rate (μGy hr^{-1}) by removal of radiation in the recovery phase	0.05	0.10	0.22	0.64
Dose rate / Prior dose rate (μGy hr^{-1}) by mass of bee (g)	-0.03	0.10	0.09	0.76

Experiment 1: The effect of radiation on bumblebee metabolic rate and activity

Table S9. Parameter estimates for models investigating the effect of radiation exposure (0.11 vs 200 μGy hr^{-1}) on mean bumblebee metabolic rate over a 5-minute observation period during the radiation phase of the experiment. The response variable (μmol min^{-1} CO_2) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 60 bumblebees for each time point. Table S9a describes minimal model used. Table S9b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	3.82	0.12	-	-
Day	-0.29	0.06	20.70	$5.36x10^{-6}$
Radiation exposure	0.34	0.15	4.80	0.03
Temperature of air drawn over bee (°C)	3.94	1.93	4.26	0.04
b. Terms removed from model in reverse order of deletion				
Time bee spent moving during measurement (s)	$1.00x10^{-3}$	$6.92x10^{-4}$	2.33	0.13
Humidity levels of air drawn over bee (%)	0.04	0.03	1.46	0.23
Age of bee at start of experiment at start of experiment (days)	-0.04	0.07	0.38	0.54
Access to a second low nectar concentration feeder	-0.08	0.16	0.30	0.59
Mass of bee at start of experiment (g)	-0.54	1.47	0.13	0.72
Radiation exposure by mass of bee (g)	-0.12	0.16	0.68	0.41
Radiation exposure by age of bee at start of experiment (days)	-0.10	0.16	0.42	0.52
Temperature levels (°C) by humidity levels (%) of air drawn over bee	0.27	0.51	0.28	0.60
Radiation exposure by days in experiment	0.03	0.08	0.11	0.75
Radiation exposure by humidity levels of air drawn over bee (%)	$-7.96x10^{-3}$	$7.73x10^{-2}$	0.014	0.91
Radiation exposure by temperature of air drawn over bee (°C)	$-3.84x10^{-2}$	$1.76x10^{-1}$	$4.0x10^{-3}$	0.99

Table S10. Parameter estimates for models investigating the effect of the previous radiation exposure (0.11 vs 200 μGy hr^{-1}) dose rate on mean bumblebee metabolic rate over a 5-minute observation period during the recovery phase of the experiment. The response variable (μmol min^{-1} CO_2) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g Model was linear mixed effects with normally distributed errors. Multiple measures were made on 60 bumblebees for each timepoint. Table S10a describes minimal model used. Table S10b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	3.90	0.07	-	-
Humidity levels of air drawn over bee (%)	-0.05	0.02	5.37	0.02
b. Terms removed from model in reverse order of deletion				
Mass of bee at start of experiment (g)	1.60	1.28	1.58	0.21
Prior radiation exposure	-0.17	0.14	1.66	0.20
Temperature levels of air drawn over bee (°C)	1.81	1.68	1.20	0.27
Time bee spent moving during measurement (s)	$3.62x10^{-4}$	$6.79x10^{-4}$	0.30	0.58
Access to a second low nectar concentration feeder	-0.06	0.14	0.17	0.68
Days within the experiment	-0.01	0.06	$7.0x10^{-3}$	0.93
Age of bee at start of experiment at start of experiment (days)	$4.0x10^{-3}$	0.06	$4.0x10^{-3}$	0.95
Prior radiation exposure by days within the experiment	-0.12	0.09	3.46	0.06
Temperature levels (°C) by humidity levels (%) of air drawn over bee	1.09	1.18	0.92	0.34
Prior radiation exposure by age of bee at start of experiment (days)	0.09	0.12	0.60	0.44
Prior radiation exposure by mass of bee (g)	-0.08	0.15	0.36	0.55

Table S11. Parameter estimates for models investigating changes in the effect of radiation dose rate (0.11 vs 200 μGy hr^{-1}) on mean bumblebee metabolic rate over a 5-minute observation period across the radiation and recovery phases of the experiment. The response variable (μmol min^{-1} CO_2) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 60 bumblebees during these observations. Table S11a describes minimal model used. Table S11b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	3.83	0.11	-	-
Days within the experiment	-0.29	0.06	16.50	4.88×10^{-5}
Radiation exposure	0.35	0.14	1.66	0.20
Phase of the experiment	0.03	0.15	1.69	0.19
Temperature of air drawn over bee (°C)	3.96	1.36	5.05	0.03
Radiation exposure by phase	-0.44	0.19	5.54	0.02
Days within experiment by phase	0.21	0.06	10.93	1.00×10^{-3}
b. Terms removed from model in reverse order of deletion				
Time bee spent moving during measurement (s)	7.15×10^{-4}	4.95×10^{-4}	2.14	0.14
Access to a second low nectar concentration feeder	-0.07	0.11	0.39	0.53
Mass of bee at start of experiment (g)	0.02	0.05	0.22	0.64
Humidity levels of air drawn over bee (%)	-0.01	0.02	0.26	0.61
Age of bee at start of experiment at start of experiment (days)	2.00×10^{-3}	0.05	0.01	0.91
Radiation exposure by days in experiment	-0.07	0.06	1.26	0.26
Radiation exposure by mass of bee (g)	0.07	0.11	0.43	0.51
Radiation exposure by age of bee at start of experiment (days)	-0.01	0.07	0.10	0.76
Temperature levels (°C) by humidity levels (%) of air drawn over bee	-0.02	0.47	3.0×10^{-3}	0.96
Phase by radiation exposure by days within the experiment	-0.16	0.13	1.70	0.19

Table S12. Parameter estimates for zero inflated model investigating the effect of radiation dose rate (0.11 vs 200 μGy hr^{-1}) on the time a bee spent active during the radiation phase of the experiment. The response variable (s) was not square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 gModel is zero inflated which assumed errors had a Gaussian distribution. Multiple measures were made on 60 bumblebees for each timepoint. Table 12a describes minimal model used. Table S12b describes terms removed from the model in reverse order of deletion during model simplification. Table S12c describes terms removed from the zero inflated part of the model in reverse order of deletion during model simplification. No terms were significant for the zero inflated part of the model.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	127.50	12.51	-	-
Radiation exposure	37.80	18.01	2.10	0.04
b. Terms removed from model in reverse order of deletion				
Days within the experiment	8.40	5.94	1.41	0.16
Age of bee at start of experiment (days)	-9.65	8.35	-1.16	0.25
Temperature levels of air drawn over bee (°C)	139.07	246.22	0.57	0.57
Mass of bee at start of experiment (g)	0.82	9.12	0.09	0.93
Radiation exposure by age of bee at start of experiment (days)	15.59	11.88	1.31	0.19
c. Terms removed from the Zero-inflated part of the model in reverse order of deletion				
Radiation exposure	0.55	0.46	1.19	0.24
Mass of bee at start of experiment (g)	-0.24	0.22	-1.05	0.30
Days within the experiment	-0.05	0.15	-0.34	0.74
Age of bee at start of experiment (days)	-0.02	0.22	-0.07	0.94
Temperature levels of air drawn over bee (°C)	0.90	6.82	0.13	0.90
Radiation exposure by age of bee at start of experiment (days)	0.23	0.31	0.72	0.47

Table S13. Parameter estimates for zero inflated model investigating the effect of radiation dose rate (0.11 vs 200 μGy hr^{-1}) on the time a bee spent active during the recovery phase of the experiment. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model investigating time a bee was active is zero inflated which assumed errors had a Gaussian distribution. Multiple measures were made on 60 bumblebees for each timepoint. Table S13a describes minimal model used. Table S13b describes terms removed from the model in reverse order of deletion during model simplification. Table S13c describes terms removed from the zero inflated part of the model in reverse order of deletion during model simplification. No terms were significant for the zero inflated part of the model.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	130.32	10.53	-	-
b. Terms removed from model in reverse order of deletion				
Temperature levels of air drawn over bee (°C)	260.49	276.96	0.94	0.35
Days within the experiment	2.27	7.65	0.30	0.77
Mass of bee at start of experiment (g)	-2.42	11.18	-0.22	0.83
Radiation exposure	4.61	21.21	0.22	0.83
Age of bee at start of experiment (days)	-1.44	9.03	-0.16	0.88
Radiation exposure by age of bee at start of experiment (days)	3.11	14.15	0.22	0.83
c. Terms removed from the Zero-inflated part of the model in reverse order of deletion				
Radiation exposure	0.04	0.42	0.91	0.36
Days within the experiment	-0.22	0.14	-1.51	0.13
Mass of bee at start of experiment (g)	-0.16	0.22	-0.74	0.46
Temperature levels of air drawn over bee (°C)	-2.37	5.49	-0.43	0.67
Age of bee at start of experiment (days)	0.01	0.19	0.05	0.96
Radiation exposure by age of bee at start of experiment (days)	-0.53	0.29	-1.81	0.07

Table S14. Parameter estimates for zero inflated model investigating the effect of radiation dose rate (0.11 vs 200 μGy hr^{-1}) on the distance a bee moved during the radiation phase of the experiment. To investigate distance a bee moved, all variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model is zero inflated which assumed errors had a Gaussian distribution. Multiple measures were made on 60 bumblebees for each timepoint. Table S14a describes minimal model used. Table S14b describes terms removed from the model in reverse order of deletion during model simplification. Table S14c describes terms removed from the zero inflated part of the model in reverse order of deletion during model simplification. No terms were significant for the zero inflated part of the model.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	100.78	9.79	-	-
Radiation exposure	25.21	14.29	1.76	0.05
b. Terms removed from model in reverse order of deletion				
Temperature levels of air drawn over bee (°C)	225.06	140.86	1.60	0.11
Age of bee at start of experiment (days)	-4.03	6.56	-0.62	0.54
Mass of bee at start of experiment (g)	3.35	7.01	0.48	0.63
Days within the experiment	0.05	6.73	7.0×10^{-3}	0.99
Radiation exposure by age of bee at start of experiment (days)	9.77	9.51	1.03	0.30
c. Terms removed from the Zero-inflated part of the model in reverse order of deletion				
Radiation exposure	0.72	0.45	1.58	0.12
Mass of bee at start of experiment (g)	-0.15	0.22	-0.67	0.50
Days within the experiment	-0.04	0.15	-0.28	0.78
Age of bee at start of experiment (days)	-0.04	0.22	-0.19	0.85
Temperature levels of air drawn over bee (°C)	0.59	6.57	0.09	0.93
Radiation exposure by age of bee at start of experiment (days)	0.22	0.31	0.72	0.47

Table S15. Parameter estimates for zero inflated model investigating whether the effect of radiation dose rate (0.11 vs 200 μGy hr^{-1}) on the time a bee spent active changed between the radiation and recovery phases of the experiment. Multiple measures were made on 60 bumblebees for each timepoint. Table S15a describes minimal model used. For time a bee spent active all variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.07°C, humidity is 3.41% and bumblebee mass of bee at start of the experiment 1.02 g. Model is zero inflated which assumed errors had a Gaussian distribution. Table S15b describes terms removed from the model in reverse order of deletion during model simplification. Table S15c describes terms removed from the zero inflated part of the model in reverse order of deletion during model simplification. No terms were significant for the zero inflated part of the model.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	138.21	7.01	-	-
b. Terms removed from model in reverse order of deletion				
Radiation exposure	21.87	13.92	1.57	0.12
Phase of the experiment (Recovery)	-15.43	13.87	-1.11	0.27
Temperature levels of air drawn over bee (°C)	246.11	155.10	1.59	0.11
Age of bee at start of experiment (days)	-5.45	6.19	-0.88	0.38
Days within the experiment	2.42	5.58	0.43	0.67
Mass of bee at start of experiment (g)	-1.64	7.08	-0.23	0.82
Radiation exposure) by age of bee at start of experiment (days)	9.38	9.21	1.02	0.31
Days within the experiment by phase (recovery)	-1.53	9.56	-0.16	0.87
Radiation exposure by phase (Recovery)	-1.16	9.51	-0.12	0.90
c. Terms removed from the Zero-inflated part of the model in reverse order of deletion				
Radiation exposure	0.45	0.31	1.47	0.14
Days within the experiment	-0.14	0.10	-1.30	0.19
Mass of bee at start of experiment (g)	-0.20	0.16	-1.25	0.21
Phase of the experiment (Recovery)	0.32	0.31	1.04	0.30
Temperature levels of air drawn over bee (°C)	-0.50	4.16	-0.12	0.91
Age of bee at start of experiment (days)	1.00×10^{-3}	0.14	0.01	0.99
Days within the experiment by phase (recovery)	-0.18	0.22	-0.86	0.39
Radiation exposure by age of bee at start of experiment (days)	-0.18	0.21	-0.87	0.39
Radiation exposure by phase (Recovery)	-0.14	0.64	-0.22	0.83

Experiment 2: The dose-rate threshold of the effect of radiation on bumblebee nectar consumption

Table S16. Parameter estimates for model investigating the effect of radiation dose rate on bumblebee nectar consumption (ml). Multiple measures were made on 141 bumblebees during these observations. Table S16a describes minimal model used. The response variable (ml) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.36°C, humidity is 2.68% and bumblebee mass of bee at start of the experiment 1.04 g. Model was linear mixed effects with normally distributed errors. Table S16b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	727.00	12.82	-	-
Concentration of nectar	-3.63	1.16	6.79	0.01
Days within the experiment	3.98	1.00	27.57	1.5×10^{-7}
Dose rate (μGy hr^{-1})	0.39	0.16	4.89	0.03
Average temperature during the days when the nectar measurements were made (°C)	-0.07	0.01	21.21	4.12×10^{-6}
Concentration of nectar by days within experiment	744.00	8.97	10.86	9.80×10^{-4}
Concentration of nectar by dose rate (μGy hr^{-1})	-2.36	0.84	4.39	0.04
Concentration of nectar by dose rate (μGy hr^{-1}) by days within the experiment	-4.88	1.97	6.03	0.01
b. Terms removed from model in reverse order of deletion				
Dose rate (μGy hr^{-1}) by days within experiment	-0.08	0.01	2.0×10^{-3}	0.96
Mass of bee at start of experiment (g)	-1.49	9.48	2.85	0.08
Age of bee at start of experiment at start of experiment (days)	-2.16	2.41	1.02	0.32
Average humidity during the days when the nectar measurements were made (%)	0.04	0.03	0.91	0.34
Dose rate (μGy hr^{-1}) by mass of bee (g)	0.39	0.16	4.74	0.29
Dose rate (μGy hr^{-1}) by age of bee at start of experiment (days)	-0.07	0.05	3.25	0.06
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	8.73×10^{-3}	6.49×10^{-3}	1.80	0.18
Dose rate (μGy hr^{-1}) by average temperature during the days when the nectar measurements were made (°C)	-1.9×10^{-4}	2.66×10^{-4}	0.53	0.47
Dose rate (μGy hr^{-1}) by average humidity during the days when the nectar measurements were made (%)	0.01	0.01	0.18	0.67

Table S17. A breakdown of parameter estimates for model S16 when dose rates are removed in increments to investigates a potential threshold effect of radiation dose rate on bumblebee nectar consumption (ml). For each line of the table, data from dose rates above the threshold stated were removed (in 10 μGy hr^{-1} increments) and the parameter estimate for the effect of radiation was recalculated.

Dose rates	Estimate	SE	χ^2	P Value
192	$3.53x10^{-4}$	$1.61x10^{-4}$	2.16	0.03
180	$2.42x10^{-4}$	$1.83x10^{-4}$	1.85	0.18
170	$1.36x10^{-4}$	$2.08x10^{-4}$	0.32	0.57
160	$1.35x10^{-4}$	$2.06x10^{-4}$	0.31	0.56
150	$2.64x10^{-4}$	$2.34x10^{-4}$	4.51	0.03
140	$2.66x10^{-4}$	$2.51x10^{-4}$	1.41	0.25
130	$2.17x10^{-4}$	$3.08x10^{-4}$	0.59	0.44
120	$2.17x10^{-4}$	$3.09x10^{-4}$	3.80	0.05
110	$5.15x10^{-4}$	$3.54x10^{-4}$	2.92	0.08
100	$5.43x10^{-4}$	$3.74x10^{-4}$	1.63	0.20
90	$2.75x10^{-4}$	$4.37x10^{-4}$	0.58	0.44
80	$4.77x10^{-4}$	$5.08x10^{-4}$	2.11	0.15
70	$5.56x10^{-4}$	$6.22x10^{-4}$	4.51	0.03
60	$5.97x10^{-4}$	$6.79x10^{-4}$	0.57	0.45
50	$2.54x10^{-4}$	$7.70x10^{-4}$	0.09	0.76
40	$1.72x10^{-3}$	$1.10x10^{-3}$	4.29	0.04
30	$1.50x10^{-3}$	$1.33x10^{-3}$	0.23	0.63
20	$2.82x10^{-3}$	$2.11x10^{-3}$	0.35	0.56

Table S18. Parameter estimates for model investigating the effect of radiation dose rate on bumblebee (wet) mass. Multiple measures were made on 141 bumblebees during these observations. Table S18a describes minimal model used. The response variable (grams) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of temperature is 0.36°C, humidity is 2.68% and bumblebee mass of bee at start of the experiment 1.04 g. Model was linear mixed effects with normally distributed errors. Table S18b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	0.14	2.86×10^{-3}	-	-
Days within the experiment	-56.69×10^{-4}	1.22×10^{-4}	18.09	2.17×10^{-5}
Mass of bee at start of experiment (g)	2.61×10^{-2}	2.64×10^{-3}	71.99	$< 2.2 \times 10^{-16}$
b. Terms removed from model in reverse order of deletion				
Average humidity during the days when the nectar measurements were made (%)	8.34×10^{-4}	6.66×10^{-4}	2.96	0.09
Concentration of nectar	1.37×10^{-4}	3.39×10^{-4}	1.09	0.31
Dose rate (μGy hr^{-1})	2.68×10^{-5}	4.27×10^{-5}	0.53	0.47
Average temperature during the days when the nectar measurements were made (°C)	1.55×10^{-4}	2.75×10^{-3}	0.02	0.88
Dose (μGy hr^{-1}) by days within the experiment	2.82×10^{-6}	2.47×10^{-6}	1.34	0.25
Concentration of nectar by days within the experiment	9.17×10^{-6}	1.15×10^{-5}	0.82	0.38
Concentration of nectar by dose rate (μGy hr^{-1})	2.24×10^{-6}	3.78×10^{-6}	0.40	0.53
Average temperature (°C) by humidity (%) during the days when the nectar measurements were made	4.71×10^{-4}	1.20×10^{-3}	0.12	0.38
Dose rate (μGy hr^{-1}) by average humidity during the days when the nectar measurements were made (%)	-1.98×10^{-6}	6.92×10^{-6}	0.08	0.35
Dose rate (μGy/hr^{-1}) by mass of bee at start of experiment (g)	9.69×10^{-6}	4.69×10^{-5}	0.08	0.73
Dose rate (μGy hr^{-1}) by average temperature during the days when the nectar measurements were made (°C)	7.11×10^{-6}	4.88×10^{-5}	0.02	0.88

Table S19. Parameter estimates for model investigating the effect of radiation dose rate on bumblebee dry weight. The response variable (grams) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of bumblebee mass of bee at start of the experiment is 1.04 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 141 bumblebees during these observations. Table S19a describes minimal model used. Table S19b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	4.24×10^{-2}	5.51×10^{-4}	-	-
Concentration of nectar	1.56×10^{-4}	4.87×10^{-5}	19.77	9.93×10^{-6}
Dose rate (μGy hr^{-1})	1.13×10^{-5}	6.98×10^{-6}	2.11	0.11
Mass of bee at start of experiment (g)	7.22×10^{-3}	5.32×10^{-4}	470.57	$< 2.2 \times 10^{-16}$
Dose rate (μGy hr^{-1}) by mass of bee at start of experiment (g)	3.03×10^{-5}	7.02×10^{-6}	18.71	1.76×10^{-5}
b. Terms removed from model in reverse order of deletion				
Concentration of nectar by dose rate (μGy hr^{-1})	-1.89x 0^{-7}	5.96×10^{-7}	0.10	0.75

Table S20. Parameter estimates for model investigating the effect of radiation dose rate on bumblebee thorax temperature. The response variable (degrees centigrade) was square root transformed. All variables except dose rate and days were mean centered and scaled by the standard deviation. For this model one standard deviation of bumblebee mass of bee at start of the experiment 1.04 g. Model was linear mixed effects with normally distributed errors. Multiple measures were made on 141 bumblebees during these observations. Table S20a describes minimal model used. Table S20b describes terms removed from the model in reverse order of deletion during model simplification.

a. Minimal Model				
Predictors	**Estimate**	**SE**	χ^2	**P Value**
(Intercept)	26.22	0.05	-	-
b. Terms removed from model in reverse order of deletion				
Dose rate (μGy hr^{-1})	1.57×10^{-3}	2.04×10^{-5}	2.54	0.11
Mass of bee (g)	0.06	0.04	2.28	0.13
Concentration of nectar	4.00×10^{-3}	3.00×10^{-3}	1.39	0.24
Days within the experiment	3.00×10^{-3}	0.01	0.47	0.50
Thorax width of bee (mm)	-1.00×10^{-3}	0.05	0.05	0.83
Dose rate (μGy hr^{-1}) by thorax width of bee (mm)	9.59×10^{-4}	7.15×10^{-4}	1.64	0.24
Dose rate (μGy hr^{-1}) by bee mass (g)	-1.00×10^{-3}	8.71×10^{-4}	2.23	0.14
Concentration of nectar by days in experiment	5.33×10^{-4}	4.11×10^{-4}	1.71	0.19
Concentration of nectar by dose rate (μGy hr^{-1})	-5.70×10^{-5}	5.69×10^{-5}	0.80	0.37
Days in experiment by dose rate (μGy hr^{-1})	5.46×10^{-5}	8.16×10^{-5}	0.46	0.50
Mass of bee (g) by thorax width (mm)	3.00×10^{-3}	0.04	0.01	0.95
Days in experiment by dose rate (μGy hr^{-1}) by concentration of nectar	5.46×10^{-6}	8.16×10^{-6}	0.97	0.49

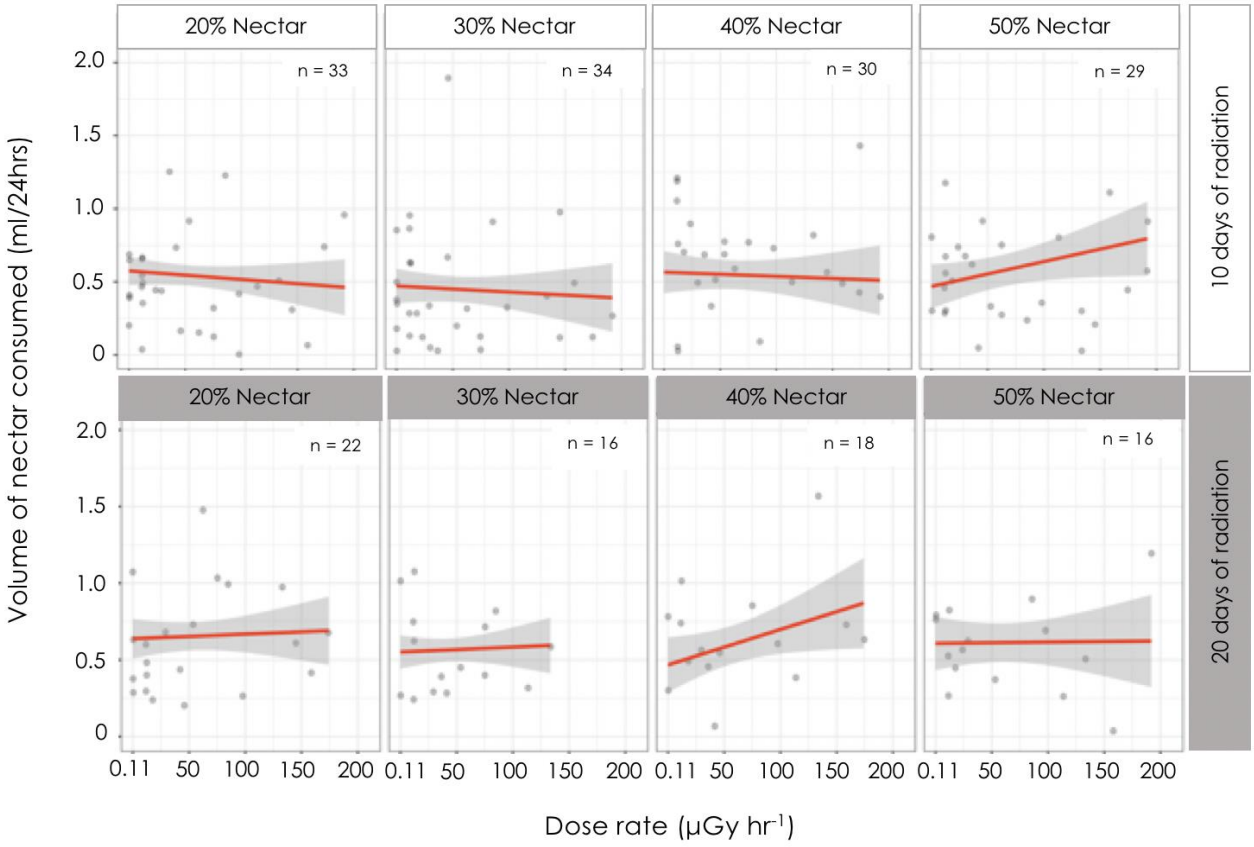


Figure S3 The volume of nectar consumed for bees provided with a nectar concentration of either 20%, 30%, 40% or 50% whilst exposed to a gradient of radiation exposure. The top four panels represent the model estimates for volume of nectar consumed on day 10, following 10 days of radiation exposure. The bottom four panels represent the model estimates for volume of nectar consumed at day 20 after a further 10 days of radiation exposure. Lines denote model fit. Points on each graph represent raw data values from each bee measured. The full model from which this was calculated was the minimal model presented in Table S16. However, for this figure nectar concentration was fitted as a fixed factor rather than a covariate to enable independent estimates of the radiation effect for each panel.

References

Australian Nuclear Science and Technology. (2022). *What is radiation?.* Available: https://www.ansto.gov.au/education/nuclear-facts/what-is-radiation. Last accessed 8th March 2022.

Beresford, N. A., Barnett, C. L., Gashchak, S., et al. (2020). Radionuclide transfer to wildlife at a "Reference site" in the Chernobyl Exclusion Zone and resultant radiation exposures. *Journal of Environmental Radioactivity*, *105661.* doi: 10.1016/j.jenvrad.2018.02.007.

Chancellor, J. C. *et al.* (2018) 'Limitations in predicting the space radiation health risk for exploration astronauts', *npj Microgravity*. Springer US, 8(3), pp. 1–11. doi: 10.1038/s41526-018-0043-2.

637