

The effect of light levels on the predation of the peppered moth

The peppered moth has cryptic coloration that gives it camouflage from bird predators. I got interested in this animal because it was used as an example in our biology lessons and our found one when I was staying in the country in the centre of France. Biologists have been interested in this moth species because of the appearance of a genetically determined black (melanic) variety. The frequency of this melanic variety was seen to increase in polluted industrial regions of England from the middle of the 19th century. It was hypothesized that this is because of the effectiveness of the camouflage of the melanic form against polluted backgrounds (soot deposit and lack of lichen covering surfaces).

Bernard Kettlewell (1955 and 1956) carried out some famous field and laboratory studies that seemed to support the idea of bird predators being the selective agent but further studies suggest that the position of the moth on the support may also influence how well they are spotted by the birds (Leibert & Brakefield, 1987). If the moths are resting on the underside of a branch, in the shade, the predation is not the same as when they are resting in a more exposed situation.

The aim of this experiment is to model the effect of adjusting the light level on predation using a simulation of natural selection on peppered moths (*Biston betularia*) with human predators.

Method

The pepper moth simulator was uploaded at the site <http://www.techapps.net/interactives/mothproject.htm> and operated on line. The “predators” were 15 students (12 female and 3 male) from a second year IB biology class. The sound from the simulation was switched off as it could have affected other “predators” in the room.

Macbooks of same model and the same age were used by all the “predators”. The screen was adjusted to **maximum brilliance** and angled for optimal viewing and the simulation was set to fit full screen. The background room lighting was kept constant.

The simulation was started on **Light Forest** and repeated twice. Each run took one minute to complete.

The percentage speckled moths in the population, at the end of the predation period, were recorded in real time on a shared spread sheet in Google documents.

The simulation was then repeated with the **Dark Forest** three times.

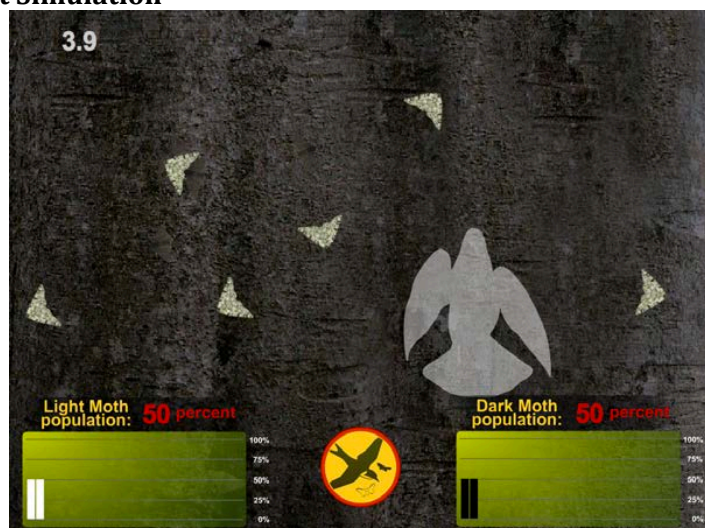
The screen light intensity was reduced to **half strength** and the simulation was repeated for Light Forest and Dark Forest again using three trials.

The whole simulation was repeated again after reducing the screen light intensity to **quarter strength**.

Finally, as a control, the simulation was run once for each background returned to full light intensity.

This investigation did not present any ethical, safety or environmental issues that needed addressing.

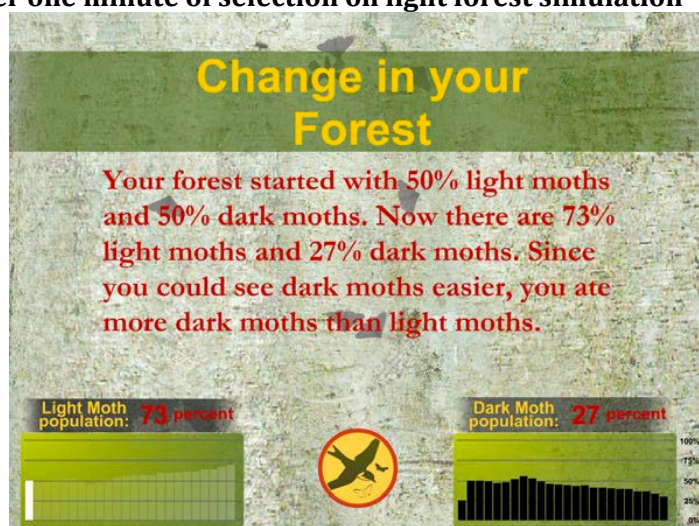
Screen shots from the simulation Dark Forest Simulation



Light Forest Simulation



Results after one minute of selection on light forest simulation



Data

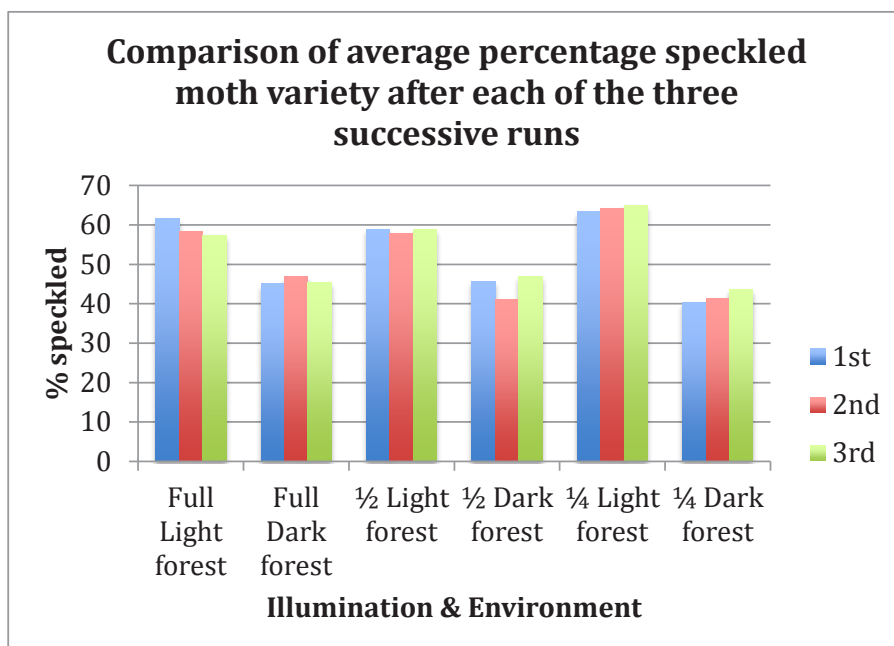
The table shows the percentage speckled variety in the population after the one minute hunt. It was considered unnecessary to record the percentage melanic form as this would show exactly the same variation in reverse (% speckled + % melanic = 100%). The simulation recorded percentages to the nearest whole percentage.

Percentage speckled variety after the 1 minute hunt ±1%																				
Full screen lighting						Half screen lighting						Quarter screen lighting						Return to full screen light		
Light forest			Dark forest			Light forest			Dark forest			Light forest			Dark forest					
1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	Light forest control	Dark forest control	
81	78	66	49	34	25	60	62	45	47	30	35	83	72	80	40	41	42	70	59	
58	62	47	52	53	51	61	48	38	55	44	45	25	28	28	57	58	61	18	58	
32	39	31	45	43	46	45	42	53	33	40	38	67	78	63	23	19	32	73	53	
81	53	59	43	57	39	64	77	68	43	41	43	78	68	79	39	38	39	65	50	
35	46	29	51	42	43	27	20	12	43	36	39	64	80	70	35	38	41	26	53	
67	58	24	42	51	62	59	53	49	47	43	47	46	56	55	39	44	47	43	44	
72	67	74	53	61	44	67	75	67	48	40	44	85	59	73	45	48	34	77	37	
50	40	62	47	30	45	52	51	54	56	41	56	48	50	46	50	45	44	47	50	
80	56	83	45	34	51	73	68	75	48	41	46	65	77	61	43	41	36	71	45	
54	64	56	52	51	42	64	52	56	52	54	56	71	62	78	47	40	44	56	42	
67	66	60	44	45	47	62	65	69	50	48	47	65	69	71	44	46	55	62	51	
66	65	66	37	52	51	66	74	85	35	40	46	51	56	60	49	51	48	50	49	
67	65	72	30	45	55	65	45	70	48	45	69	56	77	55	28	34	52	56	44	
47	65	69	46	62	34	47	74	64	38	36	52	68	70	81	28	38	47	67	45	
82	73	60	39	42	46	71	61	79	41	35	41	78	62	74	37	37	32	63	53	
Average of each run	63	60	57	45	47	45	59	58	59	46	41	47	63	64	65	40	41	44		
Overall average	60			46			59			44			64			42			56	49
Overall St deviation	15			8			15			7			14			9			17	6

4

Was there any sign of learning by the predator?

Averages were calculated from each of the predator's first, second and third runs to see if there was an improvement in the predation i.e. were the predators learning and developing a search image for the moths.

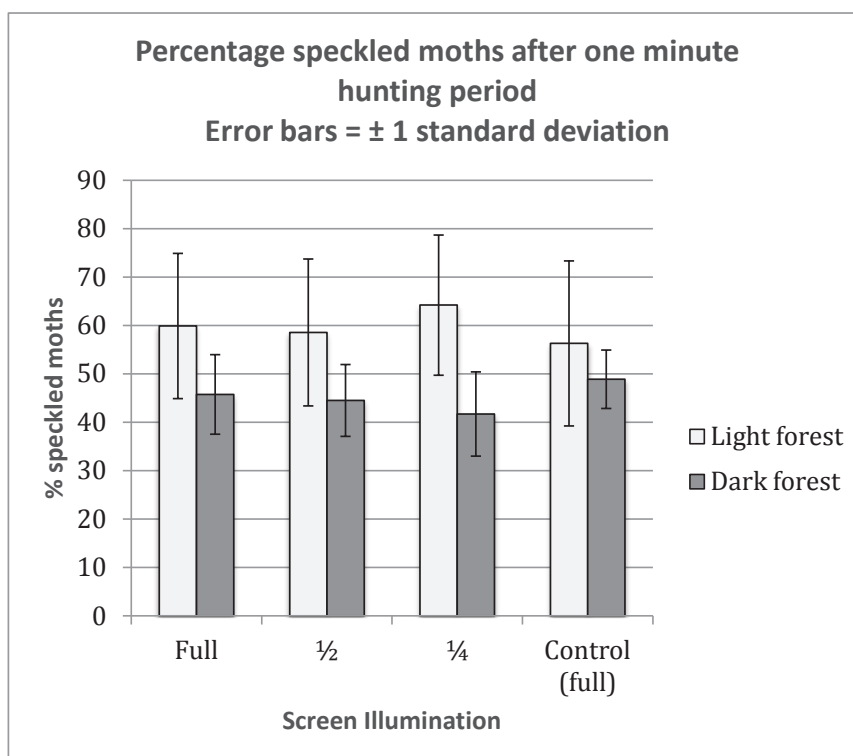


These averages do not seem to suggest that the predators were getting better at their predation during their three successive runs in the different environments. There is no apparent pattern.

Difference between the moth populations after the predation under different screen lighting conditions

This table summarizes the overall mean percentage speckled variety at the end of the one-minute hunt and their standard deviations $\pm 1\%$.

Illumination		Background	
		Light forest	Dark forest
Full screen	Mean	60	46
	Standard deviation	15	8
$\frac{1}{2}$ screen	Mean	59	44
	Standard deviation	15	7
$\frac{1}{4}$ screen	Mean	64	42
	Standard deviation	14	9
Control (return to full screen)	Mean	56	49
	Standard deviation	17	6



6

Light backgrounds appear to favor the speckled moth variety, the final percentages are consistently >50%. Similarly the Dark background selects against the speckled variety <50%. So the simulation agrees with the observations on real populations.

The overlap of the error bars of these data suggest that there is not a significant difference between the survival of the speckled forms it was decided to carry out t-tests between pairs of data sets to verify this.

t-test equation

$$t = \frac{|\bar{x}_1 - \bar{x}_2|}{\sqrt{\frac{s_1^2}{n_1} + \frac{s_2^2}{n_2}}}$$

Where:

\bar{x} = the mean

s = standard deviation

n = the sample size

Comparing the two backgrounds (light forest v dark forest) at different screen light intensities

Null Hypothesis (Ho) = There is no difference between the predation in the different simulated backgrounds (dark and light forest)

Alternative Hypothesis (Ha) = there is a difference in the predation of the peppered moth varieties in the different simulated environments

Screen lighting	full screen light intensity	half screen light intensity	quarter screen light intensity
t _{calc}	5.5435	5.5775	8.9304

For each of these comparisons the sample sizes are the same so the degrees of freedom and the t_{crit} are the same.

$$\text{Degrees of freedom } n_1 + n_2 - 2 = 45 + 45 - 2 = 88$$

t_{crit} for 88 degrees of freedom = 1.9873 (p= 0.05) for a two tailed test.

Therefore the null hypothesis is rejected in each case and the alternative hypothesis is accepted. There is a significant difference in the predation of the moths depending on the background. This is true for all the screen lighting levels. All of them are significantly different at p<0.001.

Therefore the simulation appears to be achieving its objective. The “predators are selecting for the speckled variety in the Light Forest (the values are >50% after one minute of predation) and against the speckled variety in the Dark Forest (the values are <50%).

It is interesting to observe that the difference gets greater as the screen lighting diminishes.

7

Comparing the predation against the same background at different light intensities

Ho = There is no difference between the predation in the same forest at different levels of illumination

Ha = There is a difference between the predation in the same forest at different levels of illumination

Once again the sample sizes are the same so the degrees of freedom and the t_{crit} values will be the same as above.

Light Forest background

Screen lighting	full screen light v half screen light	full screen light v quarter screen light
t_{calc}	0.4192	1.3871

Dark Forest background

Screen lighting	full screen light v half screen light	full screen light v quarter screen light
t_{calc}	0.7540	2.2673

t_{crit} for 88 degrees of freedom = 1.9873 ($p = 0.05$) for a two-tailed test.

The t_{calc} values are all below the critical values for the light background whatever the lighting level. Therefore the Null hypothesis is accepted and the Alternative hypothesis is rejected. The differences could be due to chance.

Therefore there is no significant difference due to the screen light levels used for the Light Forest backgrounds

However, for the Dark backgrounds t_{calc} is above the critical value when comparing the full lit Dark background with the quarter lit Dark Forest background. The Null hypothesis is rejected and the Alternative hypothesis is accepted.

The light intensity does seem to have an affect on the predation rates in the simulated Dark Forest environments but only when the light level is reduced to a quarter of its full intensity.

Comparing initial predation at full lighting v control (return to full lighting) at the end

Ho = There is no difference between the predation rates, in the same forest, at the beginning and at the end of the simulation

Ha = There is a difference between the predation rates, in the same forest, at the beginning and at the end of the simulation

The initial light forest predation v the control in the light forest

$$t_{\text{calc}} = 0.7293$$

The initial dark forest predation v the control in the dark forest

$$t_{\text{calc}} = 1.5810$$

For both comparisons the degrees of freedom and the critical values are the same

$$\text{Degrees of freedom} = (45 + 15) - 2 = 58$$

t_{crit} for 58 degrees of freedom = 2.0017 ($p=0.05$) for a two-tailed test.

Both of the calculated values are lower than the critical values therefore the alternative hypothesis is rejected and the null hypothesis is accepted. There is no significant difference between the initial runs and the control runs at the end.

This is reassuring as it means the predators had not changed their performance during the course of the experiment. Any differences are probably due to chance.

Discussion

The simulation succeeded in making a significant difference to the predation of the two types of moth on the different backgrounds. This was true whatever the light level used.

The human predators in this simulation showed no sign of learning that might have improved their score. This might not be true of the real predators (birds) which may learn to detect the moths more easily with practice over a longer period.

The difference in predation levels with the changes in screen lighting was less obvious. Only the lowest lighting level (a quarter screen light) showed a significant difference from full screen lighting and then only for the dark forest background. The “predators” observed that against the dark background the black form of the moth did become quite invisible. Though the t-test did not produce a significant difference of the light forest results, the calculated t value was higher when comparing full with quarter light than when full was compared with half light. A greater range of lighting levels should be tried to see if there is a trend.

The experiment used human predators on a simulation that is obviously different from the real situation. The natural predators of these moths are woodland bird species. Their vision is not the same as humans used in this experiment. Birds have vision that extends into the ultraviolet end of the spectrum. Work by Majerus, Brunton & Stalker (2000) revealed that the speckled moth varieties may be more visible to bird predators than was thought at first. The speckled variety (*typica*) is quite visible against a lichen background when the lichen is of the leafy type (foliose) but it is less visible when the lichen is of the flat (crustose) type. The melanic variety (*carbonaria*) is actually less visible under UV light when it is resting on a foliose background but it is much more visible against crustose lichens.

The simulation was designed to accommodate human visual characteristics (birds do not play video games) as a result this limits the conclusions that can be drawn. Perhaps a touch sensitive screen with real bird predators could be used or the simulation could have UV light sensitivity built into it.

The variation in the colours of the peppered moth are not as simple as speckled form and melanic form. The inheritance is polygenic so there are intermediate varieties (*insularia*) Intermediate colours are not simulated in this video and it could make a difference to the selection of the alleles that control the moths wing pigments. It would make the simulation a bit more complex but a simulated intermediary strain could be introduced into the game.

The standard deviations for these data showed a lot of variation. However, in general it can be seen that the standard deviations for the light forest are greater than those of the dark forest. In addition to this, the deviation of the average percentage light coloured variety from the starting percentage (50%) is greater for the light forest than the dark forest. This suggests that the selection was stronger in the light forest than the dark forest. The simulation may not be entirely unbiased.

Some of the “predators” noticed that the screens of the laptops were full of finger prints and in need of cleaning. It is possible that this may have introduced an uncontrolled variable into the experiment. The solution would be to clean the screens before the simulation starts.

References

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