



Foraging behaviour in leaf-cutting ants *Acromyrmex octospinosus*

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An ASAB teaching resource for A-Level and Advanced Higher students of Biology and Psychology
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Introduction

The main purpose of this teaching resource is to develop students' statistical skills in preparation for the AQA A2 Investigative Skills Assessment (ISA). It may also be used for any other course that requires students to calculate a test statistic from the following three tests; chi-square, Spearman rank correlation and standard error and 95% confidence limits. The resource was designed mainly to give students the opportunity to select the appropriate statistical test to apply to a certain set of data.

In addition, the resource includes questions aimed at aspects of 'How Science Works' and application of knowledge which many students find challenging. Within this area students need to be able to "analyse, interpret, explain and evaluate experimental methodology, results and the impact of their own and others' experimental and investigatory activities in a variety of ways" (AQA GCE Biology Specification for AS exams 2009 onwards and A2 exams 2010 onwards, version 1.3).



The resource has three main elements:

- Teachers' notes with general background information on leaf-cutting ants and suggested answers for all tasks.
- An investigation into the foraging preferences of leaf-cutting ants. This task is designed for students to collect data by observing DVD film footage of the ants' behaviour. Students will need to analyse the data using the chi-square test and interpret the outcome in relation to their test statistic. This task also includes 'How Science Works' and application of knowledge questions for students to consider aspects of the experimental design and behaviour of leaf-cutting ants.
- Two extension activities focusing on the two other statistical analyses:
 - 'Are females better parents than males?' Students will need to apply the standard error and 95% confidence limits to data collected in an investigation on the behaviour of burying beetles.
 - 'Is there a relationship between the forewing lengths of moths?' Students will need to apply the Spearman rank correlation test to data collected about the left and right forewing lengths in large yellow underwing moths.

Background information on leaf-cutting ants

Habitat and social organisation

Leaf-cutting ants are native to tropical habitats of South and Central America and also southern United States. They form large and very complex social organisations. In a developed colony, the leaf-cutting ants are divided (mostly by size) into different castes with each performing a different function. In many established colonies there are four different castes – minims, minors, mediae and majors.

- Minims are the smallest workers and care for the fungal gardens;
- Minors are slightly larger than the minims and are found in large numbers protecting and defending the foraging ants;
- Mediae are the foragers and these ants are responsible for cutting the leaf fragments and transporting them back to the colony;
- Majors are the largest ants and act as soldiers that defend the colony from intruders.

Communication between the different castes is vital for the overall functioning of the colony. The majority of this communication is carried out by means of pheromones. For example, alarm pheromones released from a distressed ant will cause all the workers in the vicinity to become alert and aggressive. Other pheromones are used to produce foraging trails to and from a particular food source. Workers returning from a successful foraging expedition produce a pheromone from the tip of the abdomen which is touched on the ground at intervals leaving a trail which subsequent foragers follow and, in turn, reinforce the trail as they return with food. When the interest in the food ceases the pheromone will eventually vapourise (Powell 1991).



Nutrition

The success of leaf-cutting ants is due to a remarkable symbiotic relationship they share with a species of fungus. The ants cut leaves in order to produce a substrate on which they cultivate the fungus in special chambers within the nest, known as 'fungal gardens'. Here the cut leaf sections are cut into even smaller pieces and are covered in droplets by the worker ants. The droplets contain enzyme complexes which help speed the breakdown of the substrate. Tufts of fungal mycelium are placed on to the medium to grow and in return they produce hyphal swellings called gongylidia which are eaten by the ants. If ants are removed from their fungal gardens, they soon die of starvation.

The fungus produces detoxification enzymes that neutralise any harmful chemicals, such as tannins, which would otherwise harm the ants. In exchange the ants secrete a chemical which kills any foreign fungal spores but has no effect on the fungus it cultivates. If the ants are removed from the fungus, it is quickly overrun by competing species of fungi and other organisms; it is therefore unable to compete on its own without the attention of the ants.

The leaf-cutting ants are also sensitive to chemicals emitted from the fungus in response to different plant material. If a particular plant material is toxic to the fungus the foragers learn to no longer collect it.

Vision

In general ants have either very small eyes or even none at all in the case of species that rely on tactile and chemosensory orientation. Like other insects, male and female ants have three ocelli (simple eyes) in addition to the more important compound eyes. The compound eyes, which are composed of many facets each of which acts like a simple eye, comprise a lens and set of photoreceptors (Gronenberg 2008).

Although colour vision has been behaviourally shown for a few ant species, the majority are not as advanced as other insects. Bees and wasps have three different photo sensitive pigments while ants appear to have only two. A study into the leaf-cutting ant *Atta sexdens* showed that they only possess a single photo sensitive pigment, which would render leaf-cutting ants colour blind (Martinoya *et al.* 1975).

This may prompt a group discussion based on the outcome of the statistical test and the knowledge of the leaf-cutting ant's habitat.

References

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I thoroughly enjoyed working on this project and I hope teaching colleagues around the UK find it useful.

Richard Bottrill



Mark Scheme

SECTION A

Task 1

'Suggested' frequencies as observed by students

50 – 60	32	44	19	28
TOTAL	147	146	102	145

No marks for this task

Task 2

1. Clear statement of null hypothesis e.g. there is no significant difference between the expected number of cut fragments and the observed number of cut fragments for the different coloured rose petals; (1)
 2. Chi-square (χ^2) test; (1)
 3. Justification of test with a clear explanation of why specific test was chosen e.g. cut fragments measured as a frequency for 4 separate categories (colours) and this test determines whether there is a significant difference between the expected and observed frequencies; (1)
 4. Test statistic calculated correctly; (1)
 5. Correct interpretation of statistical test, in terms of acceptance or rejection of null hypothesis; (1)
- Interpretation involves comparing the test statistic with the critical value at $P = 0.05$ / 5% level of probability; (1)

[Total 6 marks]

SECTION B

1. To allow the ants to acclimatise to the new conditions in the tank / to allow a sufficient number of ants to enter the tank; (1)
2. To allow sample to be large enough to minimise the effect of anomalies; to allow the sample size to be sufficient for the correct use of the statistical test; (2)
3. Same mass of petals in each dish; each dish positioned at the same distance from entrance to the tank; same species of flower; (1)
4. Any two from:
Toughness of petal; affects ability to cut fragments.
Size of petals (smaller or larger petals); smaller petals would take less time to cut into sizes that can be carried away.
Chemical composition of petals; affects preference – e.g. if a certain petal colour has a higher sugar content than the others – ants may be responding to this rather than colour. (4)
5. Suitable suggestions might be:
The size of cut petal fragments is not the same each time; certain colour may have higher frequency of cut fragments but not necessarily greater mass taken.
Position of coloured petals within the tank; position may influence distribution of ants within the tank; would need to repeat experiment and rotate the dishes of coloured petals into different positions in order to control for position of petal colour. (2)

[Total 10 marks]

Mark Scheme

SECTION C

1. Mutualistic (symbiotic); (1)
2. Breakdown (hydrolyse) the plant material (allow decompose material); (1)
3. (a) Indigestible material / fungus unable to produce certain enzymes to breakdown polysaccharides e.g. cellulose; less (soluble) nutrients available; (2)
3. (b) Larger volume of plant material needs to be collected; (1)
4. Contaminants would compete with colony fungal species / may cause death of colony fungus / would reduce nutrient availability as colony fungus would be less productive; (1)
5. (a) Negative feedback response; (1)
5. (b) Prevents the collection of damaging / poisonous plant material; prevents damage / death to the mutualistic fungus; does not affect food supply to the ants caused by damage to the fungus; (2)
5. (c) Chemicals released by fungus / chemical communication between fungus and the ants; colonies with strong communication between fungus and ants more likely to survive (accept converse); (2)

6. (a) (Intraspecific) competition / competition for food source; (1)

6. (b)

Explanation:

Different plant material – odour produced by each colony would be (chemically) very different; ants would perceive each other as foreign (not belonging to the same colony) and attack; (2)

Evaluation:

Yes, environmental factors more important than genetic factors in causing aggressive behaviour as when provided with same plant material no aggression observed;

or

No, environmental factors may not be the most important factor – need to split a single colony and provide different plant material and observe behaviour when re-combined; (1)

[Total 15 marks]

Mark Scheme

Extension activity 1

Are females better parents than males?

1. Clear statement of null hypothesis e.g. there is no significant difference in the mean amount of time spent caring for offspring between the male and the female beetles;

①

2. Standard error and 95% confidence limits;

①

3. This test determines if there is a significant difference between the means of the two samples i.e. mean time spent caring for offspring between the males and females;

①

4. Test statistic calculated correctly;

Mean of males = 53.5 minutes,

①

Mean of females = 57.0 minutes

SD of males = 31.71 minutes,

SD of females = 33.27 minutes

SE of males = 20.07 (2d.p.),

SE of females = 21.05 (2d.p.)

53.5 ± 20.07 minutes (33.43 – 73.57 minutes)

57.0 ± 21.05 minutes (35.95 – 78.05 minutes)

SE bars overlap for the two samples.

5. Correct interpretation of statistical test, in terms of acceptance or rejection of null hypothesis;

Overlap of confidence limits used to determine if significant difference exists at $P = 0.05$ / 5% level of probability;

①

①

[Total 6 marks]

Extension activity 2

1. Clear statement of null hypothesis e.g. there is no significant relationship / correlation between the length of the left and right forewing;

①

2. Spearman rank correlation;

①

3. This test determines if there is an association / correlation between two factors e.g. whether there is a correlation between the length of the left forewing and the length of the right forewing;

①

4. Test statistic calculated correctly;

①

R value = 0.83 (2d.p.) R value greater than critical value.

5. Correct interpretation of statistical test, in terms of acceptance or rejection of null hypothesis;

①

Compares the test statistic with the critical value at $P = 0.05$ / 5% level of probability;

①

[Total 6 marks]