

What happened to the holly leaf miner? Studying real food chains

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A practical ecological investigation suitable for 14–16 year-olds that is easy to resource, even in an urban environment

As part of the London Outdoor Science Project (see website) we took an observational stroll around West Ham Park in the London Borough of Newham to consider what types of urban fieldwork could be incorporated into lessons for the nearby schools. We wanted practical ideas that could be organised easily and swiftly for a class, would involve limited specialist equipment and demand little expert knowledge. We also wanted a practical experiment that would support the teaching of the new English key stage 4 science curriculum (for 14–16 year-olds), which a number of schools will be starting in May 2006, coinciding with improved weather conditions.

Holly trees (*Ilex aquifolium*) and their widespread resident, the holly leaf miner (*Phytomyza ilicis*) meet all of these needs. They are often found in urban parks and in many school grounds and lend themselves well to food chain and population studies (Collins, 1984).

The larvae of the Agromyzid leaf-miner fly (*P. ilicis*) burrow in the mesophyll of holly leaves (*I. aquifolium*) and produce characteristic white patches known as mines beneath the surface (Figure



Figure 1 Characteristic patch beneath the surface of the leaf caused by the larva of a holly leaf miner.

ABSTRACT

Investigation of the holly leaf miner's life cycle is a simple but under-used way of bringing to life the teaching of interdependence of organisms, whilst raising awareness of possibilities for fieldwork in an urban environment. This article describes an investigation devised for the new key stage 4 (for 14–16 year-olds) science curriculum in England. The life cycle of the leaf miner is investigated and food chain and population studies carried out including opportunities for the pupils to develop stronger evaluation skills. Practical resources to support the investigation are included and are available online.

1). The larvae (miners) are parasitised by a number of parasitic wasps and fed on by birds, such as blue tits (*Parus caeruleus*). Evidence of parasitism and predation are easily observable and thus provide the opportunity to study living organisms in the field, gain experience of ecological interactions in depth, and collect substantial data, which pupils can analyse and discuss (Metcalf, Marcal and Gaston, 2000).

Life cycle of the leaf miner

The tiny adult leaf-miner flies lay their eggs at the base of the midrib on the underside of the holly leaf in June. On hatching, the larva enters the midrib, slowly eating its way forward until the months of September to November when it enters the mesophyll. Further feeding produces the meandering mines that reach maximum size in March. Larvae



Figure 2 Large emergence hole left by escaping adult miner fly. The opening of the 'emergence plate' has left a triangular flap of leaf cuticle.

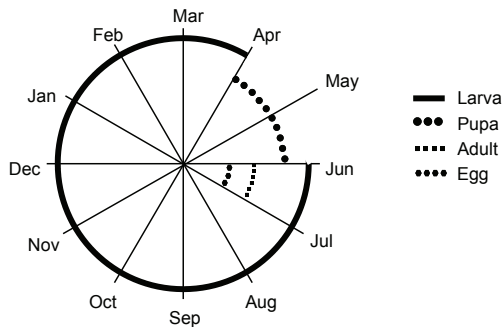


Figure 3 Life cycle of the Agromyzid leaf-mining fly, *Phytomyza ilicis* (Lewis and Taylor, 1968).

pupate within the cuticle during the last larval stage from March to May, the cuticle being retained as a protective covering (puparium) over the pupa. Before it pupates the larva prepares a thin triangular area on the leaf cuticle against which will fit the hinged 'emergence plate' of the puparium. The adult fly escapes by pressing against the 'emergence plate' in late May–June, leaving a large emergence hole (over 1 mm) (see Figure 2). The cycle continues as the adult fly lays her eggs (Figure 3).

The food chain of the holly tree

The holly leaf (producer) is consumed by the herbivorous leaf-miner larvae (first consumers), which are in turn preyed upon by the blue tit (*P. caeruleus*, a secondary consumer). If a mine has evidence of a V-shaped tear and no remains of a miner are found inside, this would indicate that the leaf has been opened by a beak (Figure 4).

Competing with the blue tits are a number of parasites. The most important of these is the wasp *Chrysocharis gemma* (another secondary consumer),



Figure 4 V-shaped tear caused by blue tit predation.

which attacks the fly larvae. The adult parasite inserts a single egg through the leaf cuticle into the body cavity of a miner-fly larva. Attacked larvae appear flaccid and pale, dirty yellow compared with the turgid, bright, shiny, whitish-lemon, healthy larvae. The parasite larva feeds within the fly larva, and eventually kills it. It then forms a shiny jet-black pupa that lies free inside the mine. Evidence that the adult parasite has emerged is a very small, neat, round hole found on either side of the mined leaf (Figure 5). Also leaving as evidence a small emergence hole, is another parasitic wasp, *Sphegigaster flavicornis*, which attacks the pupa (not the larva) of the miner fly. The adult parasite bores through the leaf cuticle



Figure 5 Small round emergence hole created by an adult parasitic wasp. Such holes may be found on either side of the leaf.

and the tough skin of the puparium with its ovipositor. It lays its eggs individually on the host pupa; the parasitic larva will bore into the pupa and feed and then pupate. The pupa of this parasite is black with a bluish tinge. Finally, yet another parasitic wasp, *Pleurotropis amyntas*, may become involved. It is less common than the other parasites but is unique as it may either live as a primary parasite in the pupa of the leaf miner (i.e. be a secondary consumer) or feed as a secondary parasite on the larvae of one of the other parasitic wasps (*S. flavicornis*) (i.e. be a tertiary consumer).

Although it is difficult to distinguish easily between the different parasites that feed on the miner, in particular in detecting whether *P. amyntas* is working at the fourth trophic level, these studies do provide opportunities to discuss contrasting food chains and feeding strategies (herbivorous, carnivorous, parasitic, hyper-parasitic).

The investigation

Three pupil worksheets to accompany these teacher guidance notes are reproduced here. Electronic worksheets, supporting *PowerPoint*, and photographs can be found at the London Outdoor Science Project website.

Preparation

Locating the holly tree colonised with mines prior to the lesson is crucial! Holly trees can generally be found in most urban parks. If you are planning to run this practical with more than one class annually, find different trees to use and record when the tree has been used for a practical. This will help you to gain worthwhile data (i.e. a tree with mines) and prevent 'over-harvesting' of a tree.

Observe and consider the immediate surroundings of the tree. These abiotic and biotic factors may later provide evidence to support interpretation by the pupils.

Equipment required

- polythene bags
- secateurs/scissors
- hand lenses
- recording sheets/clipboards
- gardening gloves

Introducing the investigation to the pupils

From their earlier studies the pupils will have prior knowledge of the essential characteristics of living organisms, food chains and food webs. Ask them to list three living organisms, their classification

and why they are assigned to different groups. This allows you to remind some pupils that plants, as living organisms, will be affected by the environment. Ask a few pupils to review a food web and the associated vocabulary on the board for the class. Using a photograph of a mined leaf (Figure 1), ask pupils what may be happening to the leaf. The life cycle of the leaf miner will need to be explained at this point. Links can be made with the caterpillar and butterfly, which pupils may have studied previously.

Key questions to emerge from discussion should include:

- What is the holly leaf miner dependent upon for its survival to mature into a fly?
- Do all holly leaf miners become flies? (Discussion of energy flow and what might happen to the pupa and the fly.)

This discussion should lead into the practical work, as evidence is now necessary to confirm possible life histories of the leaf miners and the numbers involved at each stage.

Preliminary fieldwork

Time permitting, instead of telling the pupils what they should look for as evidence of the life history of the holly leaf miner, allow the pupils to observe several mined leaves and to write down what they actually see. These leaves could be collected by the pupils or before the lesson. Using a hand lens, pupils should record any details they observe; be sure they check both sides of the leaf. Explain to them how to open the mine safely, for example by cutting around the mine and gently pulling the two epidermises apart. They should then record what they find. At this point the key may be introduced (Worksheet 2 and Figure 7). By comparing what they observe with the information on the key each group should be able to tell the class how their miner died (if at all). Photographs could be used on a projector to support this exercise (see website).

Ask the pupils: '*If we were going to investigate the main cause of death of people one hundred years ago, how would we do it?*' This question should move towards the necessity of taking samples of historic data because of time limitations. Methods of sampling information from books and the Internet can be discussed, leading to methods of sampling holly trees. Sampling methods will have been taught at an earlier stage so this may be an ideal opportunity for pupils to devise their own method to sample a holly tree. The differing sampling methods could be presented and the advantages and disadvantages of each method discussed.

- **Safety:** Caution should be exercised when collecting holly leaves and cutting the stems. Gardening gloves should be worn.

Fieldwork

One method of sampling we have found that works, using Worksheet 1 for recording, is as follows:

- To sample 600 leaves, split class into six groups.
- Each group should sample 10 randomly chosen branches.
- For each branch ignore the first few young leaves and record the number of leaves with and without mines for the next 10 leaves along the branch.
- Cut off the holly leaves that have mines and place in a plastic bag.
- Take notes of any problems or changes that had to be made during the practical.

Post-fieldwork analysis

On returning to the classroom the pupils should cut open the leaves (being careful not to cut into the mine). The top layer of the mine can then be lifted off and the inside of the mine examined. On determining what happened to each of the mined leaves and giving it a corresponding letter (using the key), the results can be recorded in Worksheet 2. These can be combined as a class set of data in Worksheet 3. A food chain with population numbers can then be drawn based on the results, as in Figure 6, and pyramids of numbers (based on living individuals or evidence of cause of death, rather than biomass) can be constructed.

With this real-life food chain in mind, a question sheet, matched to pupil ability, could ask for population prediction graphs to be drawn or

descriptions to be given if different scenarios were to occur. For example:

- If the parasite (*C. gemma*) decreased in frequency because of a sudden change in environmental conditions, what effect would this have on the leaf miner and blue tit population numbers?
- If the holly trees became diseased and prevented leaf-miner larvae from burrowing into the leaf: (a) what would be the effect on the other species in the food chain; (b) how might this affect the food web that this chain is part of?
- If numbers of the parasite (*P. amyntas*) increased rapidly, what would be the effect on: (a) the holly leaf miner; (b) the other parasites; (c) the blue tit?
- If sparrow hawks were introduced by Man into this environment, what effect would this have on the food chain?

Post-fieldwork evaluation

The pupils need to evaluate the methods they used when collecting the first-hand data and consider the validity and reliability of the data. They also need to analyse the data and discuss any weaknesses. If time is spent on discussing the chosen sampling technique to be used prior to the fieldwork, evaluating the method used should be easy and time made available to explore different techniques that they feel would provide fairer results. To support the construction of these ideas a table could be used to compare advantages and disadvantages of sampling methods. Pupils will need to think about their observational techniques, strengths and limitations of the 'trophic level' identification key and how this could be adapted for future pupils.

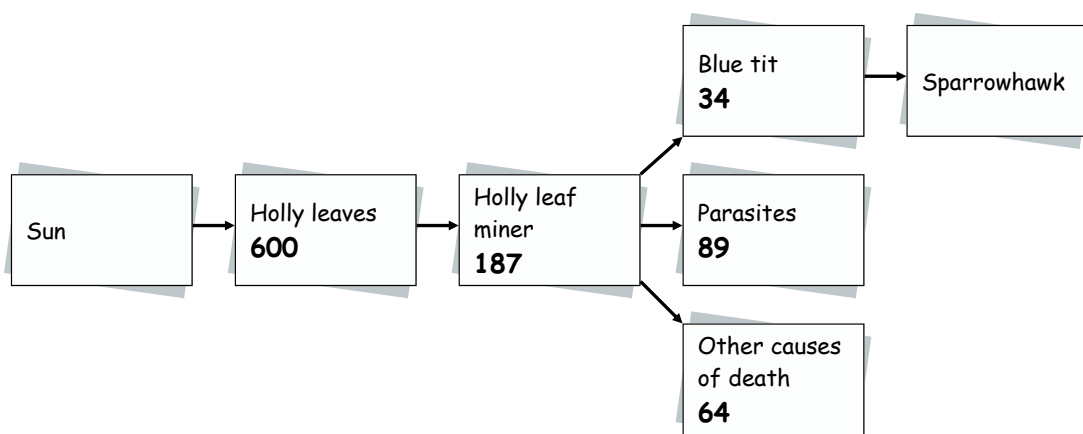


Figure 6 A holly leaf food chain.

The methodology also needs to be considered carefully by the pupils. Do they feel that this is an accurate or effective method for exploring food chains? What limitations does it have? Are there anomalies that can be explained using the site observational data? In considering the life cycle of the holly leaf miner the pupils may want to consider how results could vary depending on the time of year when the sampling took place. If the tree had been sampled in previous years how might this affect the results, if at all?

Evaluating the collection technique gives tremendous scope for discussion, as not only can further techniques be designed and justified, but research on previous sampling methods can be considered. This could be a good practice in evaluation for 'Practical data analysis' (e.g. OCR A, Unit 5 A219 strand E).

Learning outcomes

By considering population numbers and food chains pupils can investigate competition, predation and parasitism. By going outside into their local environment – even to frequently overlooked

sites in heavily developed urban settings – all pupils are given an opportunity to investigate a real-life example of ecology. This will also link to their role as custodians and beneficiaries of the natural environment, citizenship, and education for sustainable development.

Curriculum links

English Science National Curriculum Key stage 4 (pupils aged 14–16):
5a organisms are interdependent and adapted to their environment (QCA, 2004)

Possible examination board link: OCR (Oxford Cambridge and RSA) *Twenty First Century Science: Science A Module B3: Life on Earth* (B3.4) Obj. 1–4.

		Branch number									
		1	2	3	4	5	6	7	8	9	10
Number of leaves without mines											
Number of mines											

Looking around this habitat what other plants are found near your holly tree?

Would they affect the holly tree you are studying? How?

Did you have any difficulties in doing this practical?

Start in this Column		What happened to the Holly Leaf Miner		Tally	
Tear or hole found on the surface of the mine, this can be on the upper or the lower surface	V shaped tear possibly a peck mark from a beak, no remains of miner inside the mine		Predation by blue tits	A	
	Small round exit hole found on mine	Large exit hole (>1mm)	On opening the mine a shiny light brown puparium is found	Successful adult leaf miner	B
		Small exit hole like a pin prick	On opening the mine a brown puparium is found with a black pupal skin next to it	Pupa killed by parasitic wasp	C
	On opening the mine a black pupal skin is found but no brown puparium		Larva killed by parasitic wasp	C	
No hole or tear present on the upper or lower surface of the mine	On opening the mine a puparium is found	Puparium shiny and light brown		Successful pupa of leaf miner	B
		Puparium dull and dark brown	Black pupal skin can be seen inside the puparium	Pupa killed by parasitic wasp	C
			No black pupal skin inside the puparium	Other death of pupa	D
	On opening the mine a puparium is not found	Living larva present, shiny and lemon colour		Successful larva of leaf miner	B
		Remains of dead larva present	Black pupal skin present, lying away from the remains	Larva killed by parasitic wasp	C
			No pupal skin, remains are dirty yellow colour	Larva killed by parasitic wasp	C
No pupal skin, remains are dirty greenish colour	Other death of larva		D		

Group Number / Tally Totals							
	1	2	3	4	5	6	Class Total
A Predation by blue tits							
B Successful larva, pupa or adult leaf miner							
C Larva or pupa killed by parasitic wasp							
D Other death of larva							
E Number of leaves without mines (from sheet 1)							
Total number of mines sampled (A + B + C + D)							
Total number of leaves and mines sampled (A + B + C + D + E)							

References

- Collins, M. (1984) *Urban ecology*. Cambridge: Cambridge University Press.
- Lewis, T. and Taylor, L. R. (1968) *Introduction to experimental ecology*. London: Academic Press.
- Metcalf, J. A., Marcal, J. and Gaston, K. (2000) The holly leaf miner as a study organism. *Journal of Biological Education*, **34**(2), 90–94.
- QCA (Qualifications and Curriculum Authority) (2004) *Science: the National Curriculum for England*. London: HMSO.

Website

London Outdoor Science Project:
www.field-studies-council.org/outdoorscience

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