Ladybirds as teaching aids: 1 Collecting and culturing

M. E. N. Majerus, P. W. E. Kearns, H. Ireland, and H. Forge

Abstract

Ladybirds are popular insects with children. As a result, they have the potential to be useful as teaching aids for a range of biological subjects. One of the major drawbacks in the past has been the lack of information on the methods for obtaining ladybirds in the field and for caring for and breeding ladybirds in the laboratory. We here describe methods we have used for finding and collecting ladybirds during the Cambridge Ladybird Survey, and the methods we have developed for culturing ladybirds over the past nine years. The techniques described cover most of the British species.

Introduction

Ladybirds are one of the most attractive and popular groups of insects. They attract attention and mention wherever they occur. Because they find favour with children they are potentially useful as teaching material. We describe here methods for collecting ladybirds from the wild and breeding and rearing them in the laboratory. We confine ourselves to British ladybirds, although the culturing techniques we describe have already proved successful with a variety of foreign species. In a forthcoming paper we also describe ways in which ladybirds may be used as illustrative material for teaching a range of biological subjects and suggest areas where there is scope for original project work (Majerus *et al.*, 1989).

One of the main reasons why ladybirds have not been used widely in teaching is because of difficulties in culturing them in the laboratory. Several factors contribute to these difficulties. Providing an adequate supply of live food is time consuming and often problematic. Both the adults and larvae are highly cannibalistic and, if food is short, high mortality results. There have also been reports that breeding several consecutive generations results in a considerable reduction in fertility and fitness of stocks of many species (Hodek, 1973), which has made ladybirds unattractive as long-term research material. We have developed techniques which overcome these problems to a substantial extent. Our basic regime was developed for maintaining and breeding large cultures of the 2-spot ladybird (*Adalia 2-punctata*). However, the techniques work well with many other species, although in some cases modifications of the basic regime are required.

In addition, the lack of literature on the basic biology and ecology of British ladybirds has almost certainly contributed to their lack of use in teaching. We hope that this description of some of our techniques, coupled with the publication of a book on British ladybirds (Majerus and Kearns, 1989), will encourage the use of ladybirds in teaching, as well as stimulating original research (see Majerus *et al.*, 1989).

Ladybirds belong to the beetle family Coccinellidae. There are 42 British coccinellids, but a number of these are relatively small and not strongly patterned, so they would not normally be recognized as ladybirds. Consequently, there are 24 species of ladybird in Britain. Although many exhibit considerable variation in colour pattern, most species are easy to identify. For the few which are more problematic, a field key to the 24 species is available (Majerus and Kearns, 1989). (Full keys to the British Coccinellidae are given in Pope, 1953, and Majerus and Kearns, 1989.)

Collecting ladybirds

Ladybirds can be found in almost any part of the British Isles. At least a couple of species should occur within a short distance of most schools. The greatest range of species occurs in the south-east of England, with the number of species declining slightly to the west and north. Nevertheless, over half the British species may be found even in the Highlands of Scotland.

Some species of ladybird occur in a wide range of habitats, others are fairly habitat specific. Table 1

Table 1 Habitat preferences of British ladybirds

24-spot ladybird	Grassland, meadowland
13-spot ladybird	Marshes
Adonis' ladybird	Diverse, usually in dry areas
Water ladybird	Reed and reed-mace beds
Larch ladybird	Conifers
16-spot ladybird	Grassland, meadowland
2-spot ladybird	Diverse
10-spot ladybird	Diverse, usually on trees or shrubs
7-spot ladybird	Diverse
5-spot ladybird	River shingles
11-spot ladybird	Diverse, most common near coasts
Scarce 7-spot ladybird	Near nests of wood ants of the genus *Formica*
Hieroglyphic ladybird	Heather
Cream-streaked ladybird	Scots pine
Orange ladybird	Sycamore, mature woodland
18-spot ladybird	Mature Scots pine
22-spot ladybird	Grassland, meadowland
Cream-spot ladybird	Deciduous woodland, hedgerows
14-spot ladybird	Diverse
Striped ladybird	Scots pine
Eyed ladybird	Scots pine
Kidney-spot ladybird	Sallows, willows, poplars, birch, ash
Heather ladybird	Heather
Pine ladybird	Conifers

Note. This table indicates the most likely habitats in which species may be found, but almost all ladybirds can fly, so they may be found on other plants, or in other habitats from time to time.

gives a rough indication of the habitat preferences of British ladybirds. These preferences are sometimes related to specific host plant species, sometimes to general vegetation types, and sometimes to types of terrain or to climatic factors.

Collecting techniques

The most inexpensive way to find ladybirds is simply to use your eyes. Because most ladybirds are brightly coloured they are easy to see. However, some types of vegetation are easier to search than others, and of course some species of plant are more often used as host plants than others. Some of the plants which are most profitable to search by eye are listed in table 2. Other plants play host to ladybirds, but are difficult to search because of the nature of the leaves. These may be searched by either beating or sweep-netting. These techniques are also useful for finding the less brightly coloured species such as the brown coloured Larch ladybird (*Aphidecta obliterata*) and smaller species such as the 16-spot ladybird (*Micraspis 16-punctata*) and the 24-spot ladybird (*Subcoccinella 24-punctata*).

Table 2 Plants which are most profitable to search by eye for ladybirds

Nettles (*Urtica* sp.)
Thistles (e.g. *Cirsium* sp.)
Knapweed (*Centanrea* sp.)
Fat Hen (*Chenopodium album*)
Roses (*Rosa* sp.)
Willows and sallows (*Salix* sp.)
Limes (*Tilia* sp.)
Scots pine (*Pinus sylvestris*)
Birch (*Betula* sp.)
Cultivated peas and beans (*Lathyrus* sp. and *Vicia* sp.)
Larch (*Larix decidua*)
Douglas fir (*Pseudotsuga meriziesii*)
Reed-mace (*Typha* sp.)

Figure 1 Using a beating tray.

A beating tray, consisting of a piece of white material spread over a framework attached to a handle, is used for searching trees and shrubs (figure 1). It is held below a branch which is given a few sharp taps with a stout stick to dislodge any hidden ladybirds; these should then fall into the beating tray. This technique may be used with any trees but is particularly useful for finding ladybirds on oak where the irregular shape and clumping of the leaves make searching difficult. Gorse is also difficult to search except by beating because of the tightness of the vegetation, and the difficulty of moving the spiny stems by hand. Beating is also useful when searching for the Scots pine specialists. Some of these species, such as the black and red Pine ladybird (*Exochomus 4-pustulatus*), the large Eyed ladybird (*Anatis ocellata*), and the Striped ladybird (*Mysia oblongoguttata*) are fairly conspicuous. However, others, such as the 18-spot (*Myrrha 18-guttata*) and the Cream-streaked ladybird (*Harmonia 4-punctata*), are very difficult to find by eye (Majerus, 1985, 1988). Both of these latter species, although warningly coloured when moving on the pine needles, are camouflaged when at rest on the reddish brown pine buds (Majerus, 1985).

Beating trays may be obtained from entomological dealers, or are fairly easily made. An old umbrella held upside-down under the branch being beaten, is a useful substitute.

Sweep-netting is a technique used for searching low-growing vegetation. The sweep-net comprises a deep net-bag made of strong white material on a rigid ring set on a sturdy handle. It is used by walking slowly, sweeping the net with strong side-to-side

strokes, through the vegetation in front of you. It is particularly useful for finding the 16-spot, 24-spot, and 22-spot (*Psyllobora 22-punctata*) in grasslands and meadowlands, and for searching heather for the Heather ladybird (*Chilocorus 2-pustulatus*) and the Hieroglyphic ladybird (*Coccinella hieroglyphica*).

Sweep nets again may be bought from entomological dealers, but can also be made fairly inexpensively.

Ladybirds are fairly easy to handle. With a little practice it is quite easy to pick them up by hand, particularly if a moistened finger is used. However, one must be careful not to injure them, and a small paint brush is perhaps the best tool for picking them up (figure 2).

Storage

The most suitable containers for storing ladybirds when collecting are Perspex boxes with tight fitting lids. A variety of other containers may be used, but clear containers are better than opaque ones, and care should be taken with glass as condensation is often a problem in glass jars and the ladybirds may get stuck or even drown in the water droplets. There is no need to make air holes in containers as the air in the container will be sufficient for a considerable number of ladybirds for many hours. However, care should be taken to avoid placing the containers housing ladybirds in direct sunlight, even for a few minutes, as the air in the Perspex boxes will heat up rapidly and kill the ladybirds. Because of their cannibalistic tendencies, it is important not to overcrowd the ladybirds. If ladybirds are going to be kept in the collecting containers for several hours it is worth adding some food for them. This is usually easy to do as the ladybirds are generally found in the wild near suitable food, but care should be taken if plant material is added to the boxes with the food insects, as this will tend to increase condensation in airtight boxes. This may be countered by placing a layer of tissue paper or filter paper in the bottom of the container to absorb any moisture.

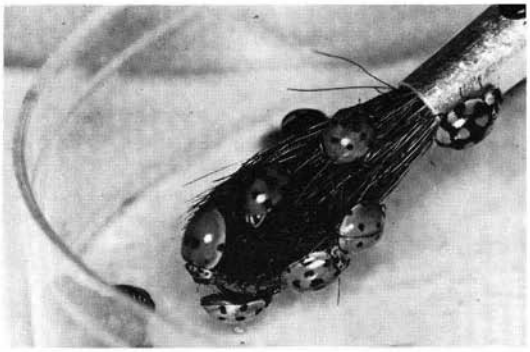

Figure 2 Picking up 10-spot ladybirds (*Adalia 10-punctata*) with a paint brush.

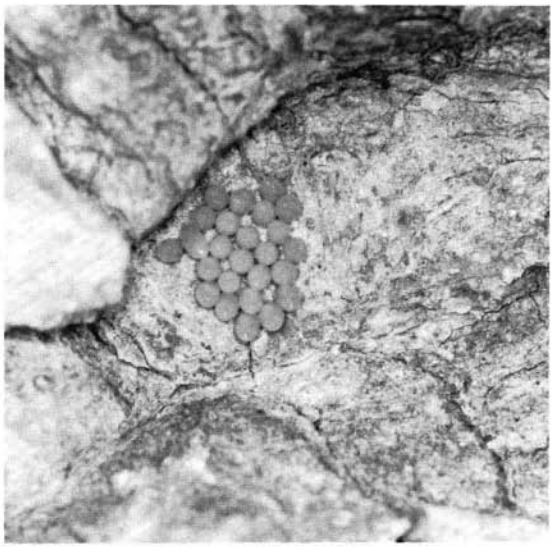

Figure 3 Batch of eggs of the Eyed ladybird (*Anatis ocellata*) on bark.

Collecting other stages

The earlier stages of the ladybird life cycle can also be collected in the wild during the summer months. Larvae of some species can be collected by beating, but, in general, searching by eye is the most efficient and successful way of finding all the early stages. Ladybird eggs are cylindrical, tapering towards the top, and are usually yellow. They are laid on either side of leaves, on stems or on the bark of trees, usually in batches of between 10 and 50 (figure 3). They should be sought on likely host plants, particularly in the vicinity of aphid colonies. The eggs themselves should not be handled in any way; they are extremely delicate and easily damaged. If a batch of eggs is found, the leaf or stem to which the eggs are attached should be collected. Eggs laid on bark can be collected by carefully slicing a thin slither of the bark under the eggs from the tree with a scalpel or sharp knife.

Ladybird larvae are elongate in shape, with relatively long legs and usually of a greyish or brownish colour with an array of white, yellow, or orange spots on the abdominal segments (figure 4). Larvae of the predatory species are almost invariably found with colonies of their food, aphids or other sap-sucking insects. They should not be handled, but should be picked up by gently sliding the bristles of a small paint brush under their legs. The larvae will cling to the brush and can then be transferred to a container.

Ladybird larvae usually pupate on vegetation. They stop feeding, attach themselves at the tip of the abdomen to the substrate, and assume a hunched position before shedding the final larval skin. They may be found on almost any type or part of the vegetation in suitable places (figure 5). The pupa

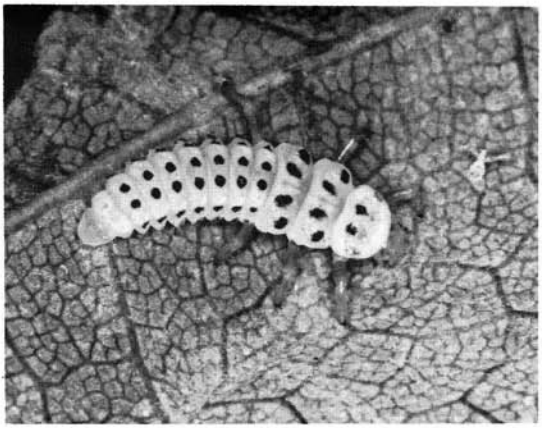

Figure 4 Larva of the orange ladybird (*Halyzia 16-guttata*).

should not be detached from the substrate, but should be collected by taking the piece of plant to which they are attached.

All British ladybirds pass the winter as adults. In the autumn, the ladybirds seek a suitable site to overwinter. Most become inactive and may stay in the same place throughout the winter without moving at all, although the three mildew-feeding species and the vegetarian 24-spot ladybird will become active and feed in mild spells. The various species of ladybird favour different types of site to overwinter. Table 3 gives a list of preferred overwintering sites. If ladybirds are found in the winter they may be collected without any detrimental effects as long as they are treated correctly afterwards. They may be kept in a warm environment, in which case they will quickly become fully active. However, they must be fed or they will die after a week or so because their activity will use up their energy reserves quickly. Alternatively, if food is not available, the ladybirds should be placed in a container with some peat, dry leaves, bark, egg boxes, or corrugated cardboard for them to rest on, and stored in an unheated outhouse, so that they are exposed to normal winter temperatures. Greenhouses or garden sheds in full sunlight are not suitable, as these will warm up too much on sunny winter days. Ladybirds being kept in this way should be disturbed as little as possible until the spring when live aphids become more readily available.

Table 3 Preferred overwintering sites of British ladybirds

24-spot ladybird	Low herbage
13-spot ladybird	Unknown
Adonis' ladybird	Plant litter, low on shrubs
Water ladybird	In reed mace or reed stems
Larch ladybird	Pine bark crevices
16-spot ladybird	Low herbage, grass tussocks, any low dry situations
2-spot ladybird	In or on bark, around window frames, in buildings
10-spot ladybird	Leaf litter
7-spot ladybird	Diverse, usually close to the ground
5-spot ladybird	Under stones, in litter
11-spot ladybird	Plant litter
Scarce 7-spot ladybird	On shrubs
Hieroglyphic ladybird	On heather, in litter under heather
Cream-streaked ladybird	Under bark or in bark crevices of pines
Orange ladybird	Leaf litter
18-spot ladybird	On pine trees, usually high up in foliage or in bark crevices
22-spot ladybird	Low herbage
Cream-spot ladybird	Leaf and plant litter
14-spot ladybird	Diverse, usually close to the ground
Striped ladybird	Unknown
Eyed ladybird	Unknown
Kidney-spot ladybird	Bark crevices of host trees
Heather ladybird	On heather, in litter under heather, on adjacent vegetation
Pine ladybird	On or under pines

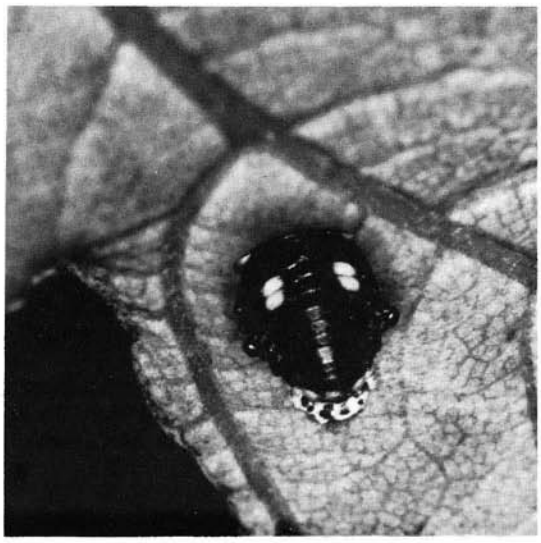

Figure 5 Pupa of the orange ladybird (*Halyzia 16-guttata*) on underside of sycamore leaf.

Culturing ladybirds

Many species of ladybird are easy to breed and rear in captivity. The method of culture will depend on the species of ladybird being maintained, the scale of the operation, and the time of year. The basic regime that we describe here was developed for maintaining large cultures (several thousand) of 2-spot ladybirds throughout the year.

Containers

We have used three basic types of containers in which to keep ladybird cultures. For most purposes, 9 cm diameter Petri dishes are the most suitable. Small

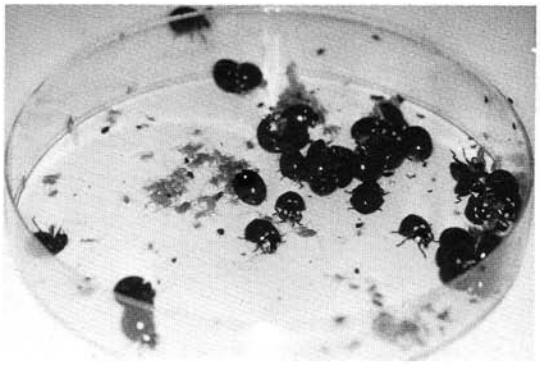

Figure 6 Breeding stocks of 7-spot ladybirds (*Coccinella 7-punctata*) in Petri dish.

populations of up to 25 2-spot ladybirds, or other species of a similar size, can be accommodated in a dish. Smaller species may be kept at a higher density, while larger species, such as 7-spot ladybirds (*Coccinella 7-punctata*) or Eyed ladybirds, need more space, and so must be kept at lower density (figure 6). If larger cultures are to be kept, then several Petri dishes can be used. Alternatively, larger containers, such as Perspex sandwich boxes can house up to a hundred 2-spots. The stocks are not so easy to feed and handle in such boxes, and egg cannibalism (adults eating the eggs) may become common. However, if non-breeding stocks are being kept, these larger containers can be very useful.

In some of our experimental work, we have kept the ladybirds in large purpose-built Perspex cages. These are 2 m × 1 m × 1 m with mesh netting covering the two ends to give an air flow and prevent both over-heating and the cages becoming wet with condensation. The ladybirds are maintained in these cages by providing them with aphids cultured on growing broad beans which are put in the cages in sand filled trays. The cages must be kept in a heated greenhouse, at least during the winter. In addition, maintaining a good balance of beans, aphids, and ladybirds requires some practice, and considerable attention must be given to the cages. This means that such a system is probably not suitable for school laboratories unless ladybirds are to be used on a very large scale.

Feeding

Most of the British ladybirds are carnivorous. The greatest problem in maintaining laboratory cultures is obtaining a constant and sufficient supply of food. Some carnivorous ladybirds seem to be fairly prey-specific and require their natural food to breed. However, many species will breed successfully when fed on species of aphid which are not necessarily part of their normal diet in the wild. We have found that over half the British species will breed when fed on the pea aphid *Acyrthosiphon pisum*.

Culturing aphids

Because we maintain large cultures of ladybirds throughout the year, we culture aphids on broad beans. The broad beans are grown in a greenhouse with a minimum day length of 18 hours and a minimum temperature of 21 °C. They are planted weekly and can be infected with aphids about three weeks after planting. Harvesting begins about one week later and continues for about another two weeks. Aphids can be harvested by brushing them from the leaves and stems into a box held below. The pea aphid (figure 7) is particularly suitable for harvesting because it drops readily from the plants when disturbed so that the plants do not get bruised or damaged when collecting aphids.

Once harvesting has begun, it is important to maintain the level of infection carefully. If too many aphids are harvested on one day, the aphid density may take several days to recover to a level where more may be harvested. If too few are removed, the aphid cultures may become so dense that the beans are adversely affected. If maintaining the balance of infestation requires more aphids to be harvested than are needed for feeding on a given day, surplus aphids should be put in a deep freezer. They should not be wasted.

Our regime for culturing pea aphids on broad beans is summarized in table 4. Other species of aphid may be cultured on broad beans. The black bean aphid (*Aphis fabae*) cultures well, but is much smaller and is much more difficult to harvest as it does not drop from the plants so readily. It is also unacceptable to some species of ladybird, or causes a decrease in fecundity. The vetch aphid (*Megoura viciae*) will also culture readily on broad beans. However, this species should be avoided as it is poisonous to many species of ladybird (Dixon, 1958; Blackman, 1967). Occasionally, wild individuals of this species find their

Figure 7 Pea aphids (*Acyrthosiphon pisum*) on broad bean.

Table 4 Programme for producing a continuous supply of live pea aphids (*Acyrthosiphon pisum*) to feed to ladybirds

Day 1	Plant broad beans (A) in pots. We plant eight seeds in each 12 cm pot. A small amount of peat is placed in the bottom of the pot, with the seeds planted about 2.5 cm down in a potting compost. The pots are watered and then put in a dark place or covered with brown paper or cardboard to accelerate germination and early growth.
Day 3	Top water pots (A).
Day 5	Top water pots (A).
Day 7	Remove pots (A) to light if shoots are showing. Top water pots (A).
Day 8	Plant broad beans (B) in pots. Thereafter treat as (A). A new set of beans should be planted each seven days thereafter. Top water pots (A). Watering should be done daily thereafter.
Day 18	Stake broad beans (A). Use a 40 cm stake placed in the centre of the pots and loosely circle a plant-tie around the beans 10–15 cm above the soil.
Day 21	Isolate pots (A) from pots (B). Infect beans (A) with pea aphids. This should be done by placing about 20 aphids on an upper leaf of one of the beans in each pot. Aphids should only be moved with a soft fine paint brush as they are very easily injured.
Day 28	A second plant tie should be placed around the beans at a height of 25–30 cm above the soil.
Day 28 (approx.)	Aphids can begin to be harvested. The first aphids harvested should be used to infect the following set of beans. The surplus may be used for feeding ladybirds, or may be frozen. With a little practice one becomes experienced at knowing how many aphids to remove from the beans. If too many are removed the culture may take several days to recover to a density where they can be harvested again. If too few are collected, the population density becomes too great, and the beans will deteriorate very rapidly. Ideally, similar numbers of aphids should be harvested each day until the beans begin to die. Following this regime, the beans should maintain harvestable numbers of aphids for about a fortnight, by which time the beans from the next two plantings should be producing harvestable colonies of aphids. A constant supply of aphids is thus maintained. Any aphids surplus to the requirements of feeding ladybirds or infecting plants should be frozen.
Day 42 (approx.)	Remove all remaining aphids from beans (A). Discard beans and soil. Wash pots before re-using.

way on to our beans, and must be removed. Fortunately, the vetch aphid is easily recognized. It is larger than the pea aphid, darker green in colour, and has obvious black legs.

From time to time problems crop up with our culture regime because the beans or aphids are attacked by other organisms. Red spider mite and white fly both attack beans and every effort should be made to detect and deal with infestations of either of these pests at an early stage, or the complete culture program may grind to a halt. The aphids themselves are prone to both predators and parasites. The predators that most frequently get into our cultures are lacewings, hoverflies, and spiders. All should be removed as a matter of course whenever they are seen. Several species of hymenoptera parasitize pea aphids. Again it is essential to keep an eye open for these parasites or for aphids which have been parasitized; the latter, when killed by the parasite, remain stuck to the plant and change colour to pale grey, tan, or buff. Parasites or parasitized aphids may be dealt with *in situ* by squashing them, or they may be removed. Early detection of hymenopterous parasites usually prevents them from reaching a density which affects the aphid cultures too severely.

Other plants may be used to culture pea aphids and we have used peas, sweet peas, and runner beans although we do not find any of these as convenient as broad beans.

Other species of aphid may be cultured on other plants. For example, the nettle aphid (*Microlophium carnosum*) can be cultured on stinging nettles (*Urtica dioica*), but the culture cycle is longer and aphid production less efficient than when using pea aphids on broad beans.

Collecting aphids from the wild

If ladybirds are to be cultured on a small scale or only for short periods a less elaborate aphid culture protocol may be used. Alternatively, aphids can be collected in the field. In fact, in the summer months we occasionally supplement our own supplies with aphids from the wild. But collecting aphids from the wild is not always straightforward. The plants upon which aphids are abundant vary from month to month throughout the summer. A myriad of factors such as the harshness of the previous winter, recent weather conditions, host plant quality, predator and parasite abundance, will affect aphid abundance. Irregularities in aphid cycles under the effects of these factors mean that plants which carry large aphid populations one year may not do so the next. In addition, not all aphids are suitable as food. Some reduce fecundity or cause larvae to develop more slowly than normal. Others are actually toxic to many species of ladybird. Generally, aphids being fed upon by wild ladybirds are suitable, but even then care must be taken, for some ladybirds are immune to some aphid toxins, while others are not. For example, the 7-spot ladybird will often be found feeding without any deleterious effects on vetch aphids which are poisonous to other species such as the 2-spot and the 10-spot ladybirds. Suitable aphids may be found on a wide range of both garden and wild plants and trees. Nettles are perhaps the most reliable source of large numbers but we have also harvested useful numbers of aphids from thistles, knapweeds, various umbellifers, roses, sweet peas, various beans, a range of fruit trees particularly flowering cherry and apple, lime, sycamore, maple, willows, sallows, and birch.

Some aphids, such as the nettle aphid, can be collected by brushing them off leaves and stems of the

host plants into a box held below. Others are more difficult to collect because they stick more tenaciously to the vegetation, or because they are small and easily squashed, even with a paint brush. For example, those from sallows and willows are usually both small and sticky. The best way to collect them is to take the leaf to which they are attached. This can then be placed directly into the ladybird container with the aphids still intact.

If aphids collected from the wild are used for feeding ladybirds, there is of course a danger of contaminating the stocks with wild ladybird larvae or with a host of other organisms. Young ladybird larvae, particularly in their first instar, are difficult to see. Their habit of riding 'piggy-back' on aphids with their mandibles embedded in their steed as they feed (figure 8), means that they will often be collected with the aphids. Wild larvae may of course confuse and disrupt experimental work if they are not detected, and they can also introduce disease into cultures.

It is almost impossible not to collect other organisms when harvesting wild aphids. Small hoverfly and lacewing larvae will often be introduced, and these are a nuisance as they also feed on aphids and so will compete with the ladybirds for aphids. Small spiders are even more of a problem, for these will spin webs in the Petri dishes which may then prove fatal to ladybirds and particularly to young ladybird larvae. It is best to remove all contaminating organisms from cultures as soon as they are detected.

Alternative foods

If it is not practical to obtain a regular supply of live aphids by culturing aphids or by harvesting them from wild populations, alternative foods may be used. The next best food to live aphids are dead ones, kept deep frozen until they are used. Aphids can be collected in the summer months when they are abundant in the wild, or when there is a surplus from laboratory cultures. The aphids should be collected into a sandwich box or similar container, which can then be put straight into the freezer until they are needed. Alternatively, aphids can be frozen in smaller quantities in Petri dishes. Ladybirds and their larvae will feed on deep frozen aphids readily, provided that the aphids were alive when frozen. Fecundity is slightly reduced, and larval development is slowed. In addition, there is often a small rise in larval mortality when frozen aphids are used. However, this can be kept to a minimum by ensuring that new frozen aphids are provided daily, that they are scattered over the bottom of the Petri dishes, and that the Petri dishes are cleaned out daily and changed regularly.

Aphids will keep for a couple of years in a freezer, provided that they are thawed and refrozen as little as possible. Repeated thawing and refreezing soon reduces frozen aphids to a glutinous mulch which is difficult if not impossible to use, and eventually becomes unacceptable to the ladybirds. To avoid this problem, it is sensible, when feeding, to remove from the freezer only the number of frozen aphids actually required.

In the wild, if aphids are in short supply, carnivorous ladybirds will resort to eating a variety of other foods. They will feed on other ladybirds or eggs, larvae and pupae of ladybirds, or any other live insects they can catch. They will scavenge on the corpses of invertebrates and vertebrates, and will also feed on plant pollen, some mildews, and on honeydew. However, in most cases, these foods, while helping to sustain the ladybirds, cause rapid declines in fecundity. The range of alternative natural foods is reflected in the range of published alternative foods for use in the laboratory. These vary from crude preparations such as chopped banana, liver, honey, or maple syrup to quite complicated recipes. In our experience none of these alternatives will keep ladybird stocks in breeding condition, and if ladybird stocks which have been fed on aphids are switched to one of these alternatives, rate of oviposition declines fairly rapidly to a very low level, or stops completely. However, we have found some alternative foods, such as the artificial diet described in table 5, to be useful in a variety of situations. Firstly, they will help to keep

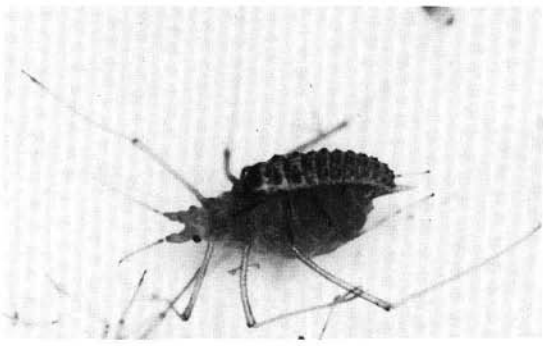

Figure 8 First-instar ladybird larva piggy-backing on pea aphid.

Figure 9 Orange ladybirds (*Halyzia 16-guttata*) feeding on artificial food.

Table 5 A recipe for an artificial food for ladybirds

Ingredients

6 grams yeast (dried, powdered, and ground)
9 grams commercially available powdered and desiccated liver
15 grams sugar
10 cm^3 maple syrup (pure)
2–3 Boots vitamin pills (without iron or copper) ground to a powder
2 grams agar
150 cm^3 water

Method

1 Grind the yeast, liver and sugar in a pestle and mortar or a Pyrex bowl.
2 Separately crush the vitamin pills, and put to one side.
3 Dissolve agar in 150 cm^3 water in a large 500 cm^3 bottle in a microwave oven, or in a small saucepan or Pyrex bowl on a hot ring or Bunsen. Pour into the receptacle containing the powdered ingredients and mix thoroughly. Reheat in microwave or oven, on a hot ring or Bunsen until the solids are dispersed evenly.
4 Allow to cool to approximately 50 °C. Add the crushed vitamin pills and maple syrup, and stir until cool enough to pour. Pour into Petri dishes or other containers, cover and allow to cool.
5 Store in a fridge.

This artificial food will keep for about a week in a fridge. After this time it may develop fungal and bacterial growths. Large quantities can be made and stored in a deep freeze, but the medium is never as good when it has been frozen and thawed. The artificial food is prepared by slicing it, in the Petri dish, into small blocks about 4 mm square. A block can then be lifted from the dish with forceps and placed into the ladybird dish. A block of this size is sufficient for about five ladybirds. The artificial medium should be replaced each day.

ladybirds alive for long periods when aphids are scarce. Secondly, they may be used to help regulate reproductive rate. When fed on a sufficient supply of aphids, ladybirds reproduce very rapidly, producing up to 40 eggs a day. Such high and rapid fecundity may become difficult to cope with. However, if an artificial food is used, the rate of egg-laying will decline and then stop, but can be restarted and increased again by reintroducing aphids into the diet when required. They can also be used in genetic breeding programs to keep ladybirds of one generation alive while their own progeny or even those of the following generation develop to the adult stage, so that matings to F_1 or F_2 progeny can be carried out.

The only crude alternative food we can recommend is chopped banana. The banana must be cut into very small pieces, cubes of about 3 mm square being about right, and should be replaced regularly.

Care of eggs, larvae, and pupae

Many species of ladybird will mate and lay eggs very readily when kept in Petri dishes or other suitable containers, as long as they are provided with a plentiful supply of live food. In most cases, females attach their eggs to the surface of the Petri dishes in small batches. It is difficult to remove eggs without damaging them, and adult ladybirds often eat their own or other females' eggs. Therefore, Petri dishes should be checked for eggs at least once a day, and if eggs have been laid, the adults should be transferred to a new dish.

In fact, if a high reproductive rate is required, it is good practice to transfer adults to a clean container each day, whether eggs have been laid or not. The effect of clean containers is to increase mating frequency, presumably because old containers become saturated with pheromones after a time. In turn, repeated matings promote a high rate of oviposition. The disposable plastic Petri dishes can be used many times, provided they are washed in hot clean water after the ladybirds have been removed. It is not advisable to use detergent.

Petri dishes which contain eggs can be stored at room temperature. At 21 °C the eggs will hatch in about 5 days. At lower temperatures the eggs will take longer to hatch; at higher temperatures less time. The hatch rate declines if eggs are kept at less than 8 °C. Eggs should be checked each day for newly hatched larvae. The eggs are generally yellow, and begin to darken and turn grey about 12–24 hours before hatching. Newly hatched larvae stay on their egg shells for a few hours after crawling out of them and their first meal consists of their shells. Normally, all fertile eggs in a batch hatch together, and it is important to provide them with aphids immediately for two reasons. Firstly, if these newly hatched larvae cannot obtain food within about 30 hours of hatching, they will die. Secondly, the larvae may start to eat each other, or if the Petri dish contains any further batches of eggs which have not yet hatched, the young larvae will eat these if aphids are not provided (figure 10). Infertile eggs shrink, become wrinkled, and eventually turn dark orange. They may be discarded.

Figure 10 Young larvae of the Eyed ladybird (*Anatis ocellata*) cannibalizing eggs.

Live aphids should be given to larvae each day from the time they hatch. There is no need to move larvae to fresh dishes, but dead aphids and other debris should be brushed out of the Petri dishes regularly. If large numbers of larvae hatch in a dish, it is desirable to move some to another dish. Ladybird larvae are highly cannibalistic if there is a shortage of food at any time. Larvae which are ecdysing or pupating are particularly vulnerable to attack. To avoid cannibalism give larvae a surplus of aphids and do not attempt to keep more than about ten larvae in a single Petri dish after the second instar. Larvae are quite easy to transfer to other dishes with a paint brush.

It is best to feed larvae with live aphids. However, if living aphids are in short supply, frozen ones may be used. Artificial food should only be used as a last resort. Most larvae will feed on it, but rate of development is slow, larval mortality tends to be high, and the resulting adults are abnormally small.

Ladybird cultures may, from time to time, become infected with viral or bacterial diseases. These are usually highly infectious. To prevent initial infection, Petri dishes should be kept as clean as possible. If any dishes are found to contain dead larvae which have apparently succumbed to disease, these dishes should be immediately isolated from all other stocks. Live larvae in these dishes should be carefully transferred to new dishes. The dead larvae and the dishes containing them should be immediately thrown away, or the larvae should be discarded and the dishes washed thoroughly in disinfectant and rinsed several times before being re-used. When dealing with infected stocks it is imperative to wash your hands properly, and disinfect any equipment being used (paint brushes, forceps etc.) in methylated spirit before returning to healthy stocks. Healthy larvae from infected dishes should be kept completely separated from other stocks until they attain the adult state.

When kept at around 21 °C with a good supply of aphids, the larvae of the 2-spot feed up in about 16 days. When ready to pupate, they cease feeding, and attach themselves to the sides or bottom of the Petri dish by an anal cremaster. They shrink in size and assume a hunched pre-pupal posture (figure 11) before shedding their final larval skin. Care should be taken not to disturb ecdysing or pupating larvae.

When all the larvae in a dish have pupated, any debris in the dish should be brushed out. Dishes, kept at 21 °C, may then be left until the adults begin to emerge, about six to eight days later at 21 °C. Emerging adults usually clamber out of the pupal case and then rest upon it while the membranous hind wings expand and dry, and while the elytra harden and begin to colour (figure 12). At this stage they should not be disturbed. If kept at room temperature with a good supply of aphids many species will be able to mate and lay eggs about a week after emergence.

The 2-spot ladybird is one of the species which will brood continuously throughout the year. Maintained at 21 °C, eight generations can be produced per annum. At higher temperatures, egg, larval, and pupal development are more rapid, and at 28 °C 12 generations can be produced in a year by taking the first emerging progeny from each generation to begin the subsequent one.

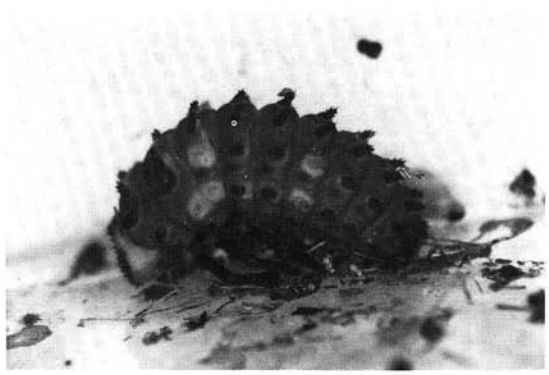

Figure 11 Pre-pupa of Eyed ladybird (*Anatis ocellata*).

Rearing other species of British ladybird

The basic techniques we have described for the 2-spot ladybird can be used unaltered for some of the other species of carnivorous British ladybird. However, many require minor or occasionally major modifications of this basic technique. Larger species of ladybird should be given more room, so less should be kept in a single Petri dish, the converse obviously being true for smaller species. Some species are rather more prey-specific than 2-spots and do not breed well on a diet of pea aphids. There is variation in the acceptability of our artificial diet to different species. Some species oviposit better if plant material is added to the Petri dish. Not all species will brood continuously. All British ladybirds overwinter as adults,

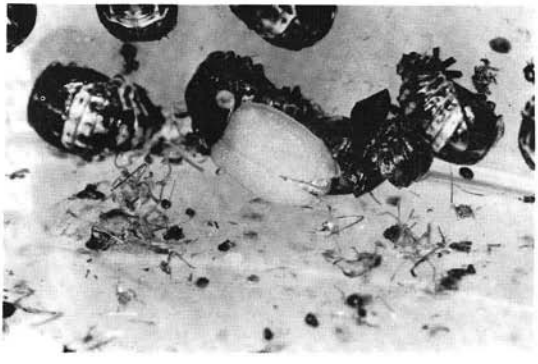

Figure 12 Newly emerged 2-spot ladybird (*Adalia 2-punctata*) on pupal case with other unhatched pupae.

and in some cases a specific environmental cue is necessary to trigger mating and oviposition. A range of modifications which we have found helpful in breeding the 20 species of predatory British ladybird is given in table 6.

Non-predatory ladybirds

Four British ladybirds are not predatory. One of these, the 24-spot is a true vegetarian. Both larvae and adults feed on the leaves of a variety of plants (e.g. campions, clovers, vetches, some grasses). They are relatively easy to breed in captivity as long as fresh cut food plant is provided daily or if they are kept in cages containing suitable potted food plants.

The other three species all feed on mildews (*Erysiphaceae*), both as adults and larvae. The 22-spot feeds on mildews growing on a variety of plants, but is most commonly associated with umbellifers. It is quite easy to breed if leaves of hogweed (*Heracleum sphandylium*) or wild angelica (*Angelica sylvestris*) covered in powdery white mildew are provided. The Orange ladybird (*Halyzia 16-guttata*) is also quite easy to rear in captivity. It favours mildew growing on the leaves of sycamore, and again the most important factor which contributes to rearing this species successfully is that fresh, mildew-covered sycamore leaves are provided daily.

The third mildew-feeding species is the 16-spot ladybird, the smallest of the British ladybirds. It feeds as both an adult and a larva on the mildews that grow on a variety of low-growing plants. However, we have not as yet been able to induce oviposition in captivity.

Acknowledgements

We wish to acknowledge with thanks those who have helped us, during the past nine years, in developing the various techniques we have described here. Ms J. Weir was involved in setting up the first 2-spot cultures in 1981 with M.E.N.M. Dr S. A. Henderson and Mr S. Albrecht concocted and developed the first usable artificial food. Mr D. Parkinson and Mr A. Feast have advised us on plant hygiene and plant pests and have grown hundreds of thousands of broad bean plants for us over the past nine years.

We have received advice on ladybirds from a number of people over the years. In particular we wish to acknowledge the help of Mr R. D. Pope and Dr J. Muggleton. A great many people have sent us information on ladybirds since we began the Cambridge Ladybird Survey in 1984. Much of the information on habitat preferences, preferred host plants, and overwintering sites has resulted from their efforts. We are grateful to them all, whether they have been involved as our own survey recorders, or through the WATCH Ladybird Spots carried out in 1986 and 1987.

Miss T. M. O. Harris read and corrected the manuscript, making many useful suggestions. Mrs J. Hunt typed the manuscript.

Finally, we wish to thank Dr P. O'Donald and Mr R. Ison for giving us such tremendous support and assistance

Table 6 Recommended modifications to basic culturing regime for breeding and rearing carnivorous British ladybirds

2-spot ladybird	Basic regime—see text.
13-spot ladybird Adonis' ladybird Water ladybird 10-spot ladybird 5-spot ladybird Cream-streaked ladybird Cream-spot ladybird 14-spot ladybird	As basic 2-spot regime.
7-spot ladybird 11-spot ladybird Scarce 7-spot ladybird	Continuously broods. Needs very large numbers of aphids to maintain high oviposition rate. Adults do not do well on artificial medium.
18-spot ladybird	Will not brood continuously. Require period of low temperatures (min. 4 weeks at less than 4 °C) followed by period of increasing day length to trigger mating and ovipositing. Mated females may be collected from April to June.
Eyed ladybird	As 18-spot, but mated females may be collected from March to June. Larvae should be given a large excess of aphids as they are extremely voracious and cannibalistic.
Pine ladybird	As 18-spot, but mated females may be collected from February to June. Oviposition rate increased if a few pine needles are added to Petri dish. Also breeds well on a diet of adelgids.
Larch ladybird	Continuously broods. Will tolerate large amounts of artificial food in place of aphids with only a small reduction in oviposition rate. Oviposition rate increases if adelgids are given in diet, and if pine needles are added to the Petri dish.
Heather ladybird Kidney-spot ladybird	Continuously brood at low rate on pea aphids. Oviposition rate increases substantially if fed on scale insects (coccids) which are the natural food of these species.
Striped ladybird	Difficult to breed in captivity unless fed on aphids from Scots pine. Mated females may be collected from March to June: these will oviposit for a short time when fed on pea aphids, but oviposition falls off unless given Scots pine aphids. Larvae will feed on pea aphids, but development is slow, mortality high, and the resulting adults small.
Hieroglyphic ladybird	We have not been able to breed this species of ladybird in captivity. We suspect a specific heather-associated prey species is required to promote mating and egg-laying. Larvae swept from heather in June/July will develop normally on pea aphids.

in the development of our general ladybird work at Cambridge.

Much of our work has been carried out in laboratory premises funded by a Wolfson Foundation Grant. The work on sexual selection in 2-spot ladybirds for which many of the techniques herein described were developed, has been supported by a series of SERC Grants (GR/B/30807; GR/C/39965; GR/D75533).

References

Blackman, R. L. (1967) The effects of different aphid foods on *Adalia bipunctata* L. and *Coccinella 7-punctata* L. *Annals of applied Biology*, **59**, 207–219.

Dixon, A. G. F. (1958) The escape responses shown by certain aphids to the presence of the coccinellid *Adalia decempunctata* (L). *Transactions of the Royal Entomological Society of London*, **110**, 319–334.

Hodek, I. (1973) *Biology of Coccinellidae*. The Hague: Junk. Prague: Academy of Science.

Majerus, M. E. N. (1985) Some notes on ladybirds from an acid heath. *Bulletin of the Amateur Entomologists' Society*, **45**, 31–37.

Majerus, M. E. N. (1988) Some notes on the 18-spot ladybird (*Myrrha 18-guttata*). *British Journal of Entomology and Natural History*, **1**, 11–13.

Majerus, M. E. N. and Kearns, P. W. E. (1989) *Ladybirds*. Naturalists' Handbook 10. Richmond, Surrey: Richmond Publishing Company.

Majerus, M. E. N., Kearns, P. W. E., Forge, H., and Burch, L. (1989) Ladybirds as teaching aids: 2 Potential for practical and project work. *Journal of Biological Education*, **23**(3), in press.

Pope, R. D. (1953) *Coccinellidae and Sphindidae. Handbook for the identification of British Insects Vol. V(7)*. London: Royal Entomological Society.

The authors

The authors are in the Department of Genetics, University of Cambridge, Downing Street, Cambridge CB2 3EH. Dr M. E. N. Majerus of Clare College is a lecturer, Dr P. W. E. Kearns of Darwin College is a post-doctoral research associate, and Mrs H. Forge and Miss H. Ireland (until her recent departure) are technicians.