

## A Review of Biological Weed Control

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Biological control would appear to be the perfect solution for pest, disease and weed control in organic and conventional agriculture (Cooke, 1988). In its widest sense it could be said to include such basic practices as crop rotation but the term biological control is now usually restricted to the deliberate application or manipulation of some natural control agent. There is considerable potential for encouraging the use of native biocontrol agents against weeds (Liebman & Davis, 2000). Substantial research effort has been put into biological control (Powell, 1990). There have been a number of successes and some failures worldwide (Crawley, 1989). In Europe, COST-816, a co-ordinated research programme for the biological control of weeds was aimed at making a concerted effort to develop a general protocol, to integrate biological into other weed control strategies, to resolve any potential conflicts and to establish a list of target weeds (Müller-Schärer & Scheepens, 1997). However, the application of biological weed control in agricultural systems in Europe has proved difficult (Müller-Schärer *et al.*, 2000). Wapshere *et al.*, (1989), reviewed the different approaches to the control of weeds by biological methods and the steps normally followed when introducing a biocontrol agent.

Classical (or inoculative) control describes the introduction of host-specific, exotic natural enemies to control alien weeds. Inundative (or augmentative) control involves the mass production and release of native (usually) natural enemies against native (usually) weeds. Conservative control is an indirect method whereby the natural level of the pests that attack the native insects that feed on the particular native weeds are reduced and maintained at a low level. This is a long term strategy that requires a detailed knowledge of the ecology of the target weeds. Broad spectrum control (or total vegetation control) as the name implies, does not target a single weed and often refers to modification of a whole habitat.

Biological weed control may put permanent pressure on a target weed by the 'one-off' application of a control agent that is able to maintain itself for a significant period after release (van Zon & Scheepens, 1979). Effective control may take time to be achieved and the control agent is likely to spread beyond the area of release. This approach is best suited to relatively stable vegetation. The alternative is to apply a control agent that is only active for a restricted period and will not persist in the environment. It has the advantage that an optimum dose can be calculated and any effect is contained within the treatment area. This approach could be used to target a problem weed in an annual cropping system. Before any of this can happen, production methods must be developed to accumulate sufficient of the control agent for it to be commercially viable (Powell, 1990). Techniques for stabilising or maintaining the agent and for its application to the target area or individual plant also need to be evaluated.

The effectiveness of selected control agents should be observed on the target weed within its native range in situations that most closely mirror the infested region (Wapshere, 1982). It is essential that biocontrol agents are thoroughly tested for host specificity so that they do not pose a threat to other plant species on release. An

example of the systems needed for the importation, testing and release of biological control agents is outlined by Shepherd (1993). The potential harmful effects of introducing non-indigenous species for biological control purposes are reviewed by Simberloff & Stiling (1996). The protected crop situation is ideal for introduced biocontrol agents, which remain contained because they will not survive outdoors in the UK. It is more difficult to control the likely spread of agents that are intended to be released into the open.

The prediction of how biological control may affect the interaction between species, and influence the life cycle of non-target species is extremely complicated. The example that is often quoted is the decline of the large blue butterfly (*Maculina arion*) following the attempt to control rabbits by introducing the *Myxoma* virus into the UK. Reduction of the rabbit population, lessened the grazing of natural grassland, and colonies of the ant (*Myrmica sabuleti*) that 'nursed' the caterpillars of the butterfly did not thrive in the altered habitat. There is the additional concern that the control agents may continue to evolve, and that changes in host specificity could occur by natural selection or mutation. The assessment of the extent of the potential risks involved in biological control remains a contentious issue (Simberloff & Stiling, 1996). Even if there were no risk to non-target species, there could still be a conflict of interests because some may perceive a particular plant as a weed while others see it as a desirable wild flower, or even a potential crop.

### **Classical biological control**

Classical biological control with insects and with micro-organisms (Evans & Ellison, 1990) has been successfully applied in South Africa (Morris, 1991), Australia (McLaren, 1993), the USA and elsewhere. It continues to be an important area of study, particularly in non-European countries (Quimby *et al.*, 1999). In 1992, classical biological weed control was evaluated by 56 countries in over 700 trials involving 144 weed species and using 370 control agents (Igrc & Maceljski, 1993); but only one trial was listed for the UK in that year. Insect damage to weeds can include the stripping of leaves and flowers, root feeding, tunnelling in the leaves and stems, the formation of galls and the hollowing out of seeds (Andres, 1982). In galls, the insect damage causes redirection of nutrients to the gall tissue.

Many of the annual weed species in the UK have been introduced at some time in the past and could be considered candidates for classical biological control. However, since their arrival most have become an established part of the flora and as such their wholesale destruction by exotic pests or diseases would not be welcomed. It has been suggested that some of the introduced, invasive perennial weeds such as giant hogweed (*Heracleum mantegazzium*), Himalayan balsam (*Impatiens glandulifera*) and the Japanese knotweeds (*Reynoutria* spp.) would be ideal candidates for classical biological control (Child *et al.*, 1993; Evans & Ellison, 1990; Fowler *et al.*, 1991). However, the giant Japanese knotweed (*Reynoutria sachalinensis*) has been shown to be a source of a natural fungicide (Maché, 1991/92), and it is possible that the other weeds may have some, as yet undiscovered, desirable feature. If so, an introduced biocontrol agent could itself need to be controlled in the future. It would be like introducing Colorado beetle (*Leptinotarsa decemlineata*) to control volunteer potatoes and then discovering that potatoes were good to eat!

In the UK, the only candidate for classical biological control has been the perennial weed bracken (*Pteridium aquilinum*) (Fowler *et al.*, 1989; 1991). However, attempts to use the caterpillars of two species of South African moth as potential biocontrol agents have not however been successful. The introduction of a classical biocontrol agent does not have to be deliberate. The rust *Puccinia lagenophorae* is of Australian origin where it attacks a range of *Senecio* species. It was unknown in Europe before 1960 but since then it has been recorded in France and the UK on groundsel (*Senecio vulgaris*) (Evans & Ellison, 1990). The rust does not kill the weed but makes it less competitive. Higher yields have been recorded in lettuce with rusted groundsel present compared with the presence of rust-free plants (Paul & Ayres, 1986).

There are examples of biological control by endemic phytophagous insects but in other situations the insects may be pests of desirable plants. Gliessman (1984) reported that in the US, flea beetles had attacked wild radish (*Raphanus raphanistrum*) and wild mustard (*Brassica campestris*) in preference to collards. In the UK, flea beetles have also been seen to reduce the number of wild radish seedlings (*Raphanus raphanistrum*) that emerged in a crop of drilled onions (Personal experience). However, it is unlikely that growers would want to encourage an increase in flea beetle numbers. Carabid beetles are predators of harmful insects but some feed solely on plant material, particularly weed seeds (Lund & Turpin, 1977). Different species are likely to vary in their preferences for the seeds of individual weed species (Tooley *et al.*, 1999). In tests in the USA, common chickweed seed was the preferred food of the four Carabid species tested.

### **Inundative biological control**

The inundative method of biological control involves the culture and release of large numbers of a biocontrol agent into the region or field where the target weed needs to be controlled. It has the advantage that native organisms can be used but there is still a requirement for host specificity (Weidemann & Tebeest, 1990). Some agents, particularly plant pathogens (mycoherbicides), can be applied as sprays in the same way as conventional herbicides. Fungal spores formulated as mycoherbicides are intended to kill specific weeds within 3-4 weeks of application (Templeton, 1985).

The history of development, current position and the future prospects of bioherbicides are reviewed by Wall (1995) and by Watson (1989). The mycoherbicides in particular offered much promise but there have been many technical difficulties to overcome in the culture, storage and application of a biological material (Weston, 1999). Formulation and spray application play an important part in microbial herbicide efficacy (Lawrie *et al.*, 1999). In the UK the potential for bracken control with mycoherbicial formulations has been investigated by Munyaradzi *et al.*, (1990). Their results indicate that to improve inoculum retention and ensure effective disease development an adjuvant is needed. Bioherbicides have the dual hurdles of the regulations that apply to biological control agents as well as those that apply to a conventional pesticide. Success also depends on collaboration between individuals from several disciplines, and between the public and private sectors (Templeton, 1988). Commercial products have been developed based on mycoherbicides but success has been limited (Bannon, 1988; Greaves & Maqueen, 1990). Four mycoherbicides have been developed and registered for use in North America (Boyetchko, 1997). The attitude of plant protection companies to bioherbicides is

discussed by Wilson (1990). Even if production problems are solved, to be successful, bioherbicides need to match chemicals in efficacy and ease of application if they are to be commercially viable; market size is also an important factor.

The specificity of a bioherbicide is increased where the susceptibility of the target organism can be enhanced. This may allow a selected area of a weed to be controlled without affecting nearby plants of the same species. Isolates of *Xanthomonas campestris* pv. *poae*, have some activity against annual meadow grass (*P. annua*), a lawn weed (Imaizumi *et al.*, 1997; Johnson *et al.*, 1996). The inoculum is taken up more readily through cut surfaces so mowing makes the weed more susceptible than plants outside the mown area that are undamaged. There can also be synergistic effects between two pathogens. In groundsel (*Senecio vulgaris*), plants naturally infected with the rust *Puccinia lagenophorae*, were killed by inoculation with the pathogen *Botrytis cinerea*, while healthy plants were not (Hallett *et al.*, 1990). It has been suggested that improvements could be made in the activity and host specificity of mycoherbicides through techniques of genetic manipulation (Bailey, 1990; Sands *et al.*, 1990). Such an approach is unlikely to be approved of by organic farmers.

A further development of the concept of bioherbicides is the isolation and application of just the toxin responsible for killing the weeds as a 'natural herbicide' rather than applying the living organism (Hatzios, 1987; Poole & Chrystal, 1985). The chemical would be easier to store, formulate and apply than a mycoherbicide, and without the risk of proliferation in the environment; but production and development costs may still be prohibitive (Froud-Williams, 1991). The isolated phytotoxins may exhibit similar host and non-host specificity to the pathogen. AAL-toxin, a natural metabolite of the pathogen *Alternaria alternata* f. sp. *lycopersici* has been tested on a range of crop and weed species, and has been patented as a herbicide (Abbas *et al.*, 1995). There is increasing interest in studying the mechanisms and sites of action of natural phytotoxins to aid the search for new herbicides (Duke *et al.*, 1997; 2002). Bacterial and fungal microbes produce a wide array of phytotoxins with the potential to be used as herbicides (Duke *et al.*, 1991). The acceptability of 'natural' herbicides to the organic standards authorities is unclear (Stopes & Millington, 1991).

Studies on bioherbicides have concentrated mainly on foliar treatments using fungi. Soil micro-organisms are often overlooked but also include important plant pathogens. Several are being investigated as potential biocontrol agents particularly for control of grass weeds such as downy brome, wild oat and green foxtail (Boyetchko, 1997). Rhizobacteria are able to colonize the root surfaces of weed seedlings and suppress plant growth, reducing weed density, biomass and seed production (Kremer & Kennedy, 1996). Plants may not be killed but their competitive ability is much reduced. The method of application of a biocontrol agent to soil may influence its effectiveness (Rhodes, 1990). Timing can be important and control release formulations may be of value where the target plant emerges over an extended period. Where placement close to the host root is required application aided by irrigation is used, as with *Phytophthora palmivora* sold as Devine for the control of milkweed vine in citrus.

Although much of the work on biocontrol agents has concentrated on the growing weed plant, there is considerable potential for using micro-organisms to manipulate or deplete the soil weed seedbank (Kremer, 1993). The persistence of weed seeds in the

soil is the key to their success in continuing to emerge despite repeated control measures over many years. Greater predation or an increase in natural decay would reduce the soil seedbank and hence future weed populations. Studies of weed seed survival in soil suggest that there can be significant effect of cropping system, tillage and burial depth (Ullrich *et al.*, 2007). The differences in survival can only in part be explained by soil microbial activity.

### **Conservative biological control**

Conservative biological control requires a detailed ecological knowledge of the weeds and control agents involved. It has received little attention and remains largely a theoretical concept based on a reduction in the native parasites, predators and diseases that attack the native biocontrol agents of the target weed (Wapshere *et al.*, 1989).

### **Broad spectrum biological control**

The oldest example of broad-spectrum biological control is the use of grazing animals and birds (cattle, sheep, horses, goats, ducks, geese etc.) to maintain pasture. In aquatic situations, the use of grass carp (*Ctenopharyngodon idella*) and other phytophagous fish has been investigated. In Australia, goats have been used to control blackberry (*Rubus fruticosus* agg.) (Dellow *et al.*, 1988). Also in Australia, targeted grazing of a range of thistles was shown to be possible with goats but sheep were not as effective (Holst & Allan, 1996). In cereals, sheep grazing in spring is a traditional practice of many organic growers to aid weed control. In the UK, the effect of weeding and sheep grazing on grain yield and quality of organic wheat has been investigated by Cosser *et al.*, (1997). Weeding increased grain yield but grazing reduced ear number. In Malaysia, weed control by sheep in permanent tree crops reduced weeding costs by up to 26% and provided additional profit from the sale of mutton (Stöber, 1993). It is known that different breeds of livestock vary in their grazing or browsing preferences and abilities and should may be taken into account for improved weed control (Soil Association, 2002).

On common land in Surrey (UK) where invasive Michaelmas daisies were a problem, Sussex cattle were found to graze and even uproot the plant (Anon, 2003). The Sussex breed are known to be foragers as well as grazers and are ideal for improving a grassland habitat.

An organic farmer in Canada has used geese to help with weed control in strawberries (Wise, 2000). The geese do not eat the strawberry leaves but consume the foliage of many different weeds although not thistles. Smaller breeds of geese do less damage by trampling. The birds are excluded before the fruit begins to ripen. They are selective in certain crops. However, they are said to favour grasses and may eat crops such as sweet corn in preference to broad-leaved weeds (Smith *et al.*, 2000).

A more extensive discussion of the use of livestock for weed management is given elsewhere in the 'organicweeds' website.

### **Arbuscular-mycorrhizal fungi**

Many aspects of plant development are influenced by arbuscular-mycorrhizal fungi (AMF), which form symbiotic relationships with certain plant species (Jordan *et al.*, 2000). Germination and early growth can be affected. Drought tolerance may be enhanced and other the effects of other forms of stress may be alleviated. In agroecosystems AMF may affect the ecology of weeds in a number of ways. Sustainable agriculture puts more organic matter into the soil, promoting biological activity and increasing AMF abundance. The fungi colonize plant roots and form mycorrhizal associations that benefit particular host plant species. Other non-host plant species may suffer reduced vigour in the presence of AMF mycelia. Exposure to an AMF inoculum has been shown to cause a 90% reduction in biomass in the non-host weed species common amaranth (*Amaranthus retroflexus*). Other weed species that suffer in the presence of AMF include fat-hen (*Chenopodium album*), pale persicaria (*Persicaria lapathifolium*), broad-leaved dock (*Rumex obtusifolius*), corn spurrey (*Spergula arvensis*) and charlock (*Sinapis arvensis*). It may prove to be a broad-spectrum biocontrol agent of non-host weed species. However, there are some weed species that benefit from AMF including ribwort plantain (*Plantago lanceolata*).

### Allelopathy

Within the broadening perceptions of biological control, allelopathy can be legitimately regarded as a component of biological control (Lovett, 1991). Allelopathy refers to the direct or indirect chemical effects of one plant on the germination, growth, or development of neighbouring plants. The effect is exerted through the release of allelochemicals by the growing plant or its residues. Microorganisms may also play a role in the production of these chemical inhibitors. Allelopathy has been considered a defence mechanism in plants (Lovett, 1982). It makes a significant contribution to the process of plant succession (Numata, 1982). The broad ecological role of allelopathy is discussed at length by Rice (1984). Widdowson (1987) considers the allelopathic interactions that are important in progressing towards holistic agriculture.

Allelochemicals may be present in the mucilage around a germinating seed (Kosemura *et al.*, 1993), in leachates from the aerial parts of plants (Tukey, 1966), in exudates from plant roots (Weston *et al.*, 1997), in volatile emissions from the growing plant (Charron *et al.*, 1995), and among decomposing plant residues (Bewick *et al.*, 1994). There has been much study of the biochemical aspects of allelopathy and investigation of the effect of plant extracts and leachates on seed germination and seedling growth in the laboratory (Waller, 1989). In the field, the evidence for allelopathy has largely come from studies of the use of organic mulches and cover crops to suppress weed emergence (Putnam *et al.*, 1983; Putnam & Defrank, 1983).

The effectiveness of living mulches, intercrops or smother crops may in part depend on their allelopathic ability. The decomposition products of organic mulches and cover crops residues may continue to prove toxic to weeds in subsequent crops. Unfortunately, such phytotoxins are also know to reduce the germination and development of drilled small seeded crops. Even the growth of transplanted crops may be checked (Russo *et al.*, 1997). Newman (1982), and Saxena *et al.*, (1996) have reviewed the relevance of allelopathy to agriculture where the toxicity of plant residues is particularly important because of the adverse effects it can have in rotational crops. While allelopathic crops or their residues inhibit the growth of

certain weeds (Steinsiek *et al.*, 1982), weeds such as fat-hen (*Chenopodium album*) that have allelopathic ability, may also influence the growth of some crops (Goel *et al.*, 1994; Qasem & Hill, 1989). Weeds can also inhibit the growth of other weeds (Anaya *et al.*, 1988).

The potential use of allelopathy for weed management has been reviewed by Altieri & Doll (1978), Numata (1982), Putman (1985), and Putnam *et al.*, (1983). Allelopathy could be used to manipulate the crop-weed balance by increasing the toxicity of the crop plants to the weeds. Where a crop has only a limited allelopathic effect it may still be sufficient to reduce the emergence of difficult to control weeds in the crop row, leaving only the inter-row weeds to be controlled mechanically. Studies have been made to evaluate the allelopathic ability of different cultivars in a limited range of crops e.g. peppers (Gonzalez *et al.*, 1993), oats (Fay & Duke, 1977), and cucumber (Putnam & Duke, 1974). The use of techniques that might allow genetic transfer of allelopathic ability into crop plants (Putnam, 1985), is unlikely to be an acceptable to organic practitioners. Other studies have evaluated crop cultivars for their tolerance to the allelochemicals produced by weeds (Ray & Hastings, 1992).

Another approach has been to determine the crops that contain those chemicals or their precursors with the potential to suppress weeds. The glucosinolates for example, precursors of several toxic metabolites including isothiocyanates, are found principally among members of the Cruciferae (Duncan & Milne, 1989). The effect of breakdown products from different parent glucosinolates was tested on the germination of white mustard and perennial ryegrass seed (Bellostas *et al.*, 2006). The chemicals were not equally effective and the response of the test species also differed. Grossman (1993) discusses the potential use of brassicas as alternatives to herbicides and soil fumigants for weed, pathogen and nematode control. Caliente mustard incorporated into the soil can act as a bio-fumigant (Gill, 2004). In moist soil, the chopped plant material releases isothiocyanate gas that sterilises the soil. The plants can grow up to 2 m tall but require high nitrogen levels to do this.

There have been suggestions that the allelochemicals themselves (Chung & Miller, 1995), or synthetic derivatives (Macias *et al.*, 1997) could form the basis of 'natural' herbicides (Newman, 1982; Duke *et al.*, 2000; 2002). The term 'natural plant protection agents' has been used to describe the products produced by enzyme hydrolysis of glucosinolates that have been shown to inhibit seed germination (Bellostas *et al.*, 2006).

Another approach has been the use of waste products from plants that are processed for food or oil (Duke *et al.*, 2002). Corn gluten meal, a bi-product of the wet-milling process is used for pre-emergence weed control. Seed meal waste from crambe (*Crambe abyssinica*) and from glucosinolate-containing plants also have the potential to be used for weed management. Olive processing wastes, in both liquid and solid forms have been evaluated for weed control in wheat, maize and sunflower (Boz *et al.*, 2003).

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