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The Evolution of Flower Size and Flowering Behaviour in Plants: The Role of Pollination and Pre-Dispersal Seed Predation.

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Thesis for the degree of Master of Philosophy

June 2008

CONTENTS

Chapter 1. Introduction

Floral traits, as primary sexual characteristics of plants that rely on third party organisms for gamete transfer, represent aspects of plant physiology open to study for evidence of evolutionary selective pressure affecting physically observable characteristics of organisms.

The mean flower size adopted by a species may be a compromise between the need to attract pollinators (selection resulting in big flowers) and the need to avoid pre-dispersal seed predators (selection resulting in small flowers). If both selection pressures are indeed found within the same plant species, a combination of field data on both seed set success and seed predation may reveal a capitulum size that is maintained by these competing forces. Timing of flowering (i.e. the calendar slot in the season) may also be subject to selection by these two sets of organisms..

The research described here has set out to investigate these questions by posing several hypotheses that support the concept of competing selective pressures, and seeking to provide their proof by a combination of field data collection and theoretical evolutionary modelling.

1.1 Aims

This research aims to investigate the evolutionary role of predator-host and pollinator-plant relationships in the evolution of floral traits. Research was carried out on various species of Asteraceae, which were considered to be suitable for study, both because of existing research work, which identified pre-dispersal seed predation as existing at significant levels within the family, and also due to their composite nature, that demonstrates a simple relationship between flower size and fecundity.

Several hypotheses can be identified, each of which has to be confirmed in order to make the case for an overall hypothesis that pre-dispersal seed predation, and pollination success and seed potential, act as competitive selective pressures on floral traits within certain species of Asteraceae:

1 Pre-dispersal seed predators tend to select larger flower heads as oviposition sites in a repeatable pattern over time and population.

2 The presence of seed predators within flower heads reduces their fecundity.

3 The size of flower heads is correlated with successful seed set.

4 The pattern of seed predation over the course of a season is repeatable over several seasons and locations.

5 The combined effect of selective infestation of flower heads, and reduction in fecundity, and correlation of flower size and seed set, leads to an evolutionary pressure that selects for an optimum flower size in subsequent generations.

6 The combined effect of the pattern of predation and reduction in fecundity leads to an evolutionary pressure that selects for phenological patterns in subsequent generations.

In order to test these hypotheses, two approaches have been followed:

Firstly field research has been undertaken to collect and measure the size of flower heads in three species of Asteraceae, discriminating by means of predator selection for oviposition (1); collection of flower heads has been undertaken to determine seed set, discriminating by means of capitulum size and predator oviposition (2 and 3); by sampling over several full seasons, patterns of predation have been determined that can be compared with the species phenology (4).

Secondly, a mathematical model has been constructed to combine the effects of infestation pattern, fecundity levels, and phenological patterns, to examine the effects on subsequent generations, and suggest an evolutionary role for seed predation (5 and 6).

These researches are described below, following an extensive literature review, Chapter 2 that seeks to underpin the validity of the hypotheses. Chapter 3 then describes the methods of data collection, as well as detailing the data set itself.

The measurable hypotheses are tested in Chapters 4 (predation pattern), 5 (fecundity and seed set) and 6 (phenological patterns), followed by a description of the mathematical model used to investigate the combination of these effects on floral size and phenology, together with details of the results suggested by the model in Chapter 7. Finally a general discussion and conclusion is presented in Chapter 8.

Chapter 2. Literature Review

Predator-host interactions have been studied extensively over many years, and have formed an important basis for ecological studies such as the classic paper 'The Natural Control of Population Balance in the Knapweed Gall-Fly (*Urophora jaceana*)', Varley (1947), which developed an analysis of the mutual effect of parasitic and other factors on the population density of one of the Tephritid predators of *Centaurea nigra*. However, such interactions can be complex. Ostergard and Ehrlen (2005), investigating how seed predators can affect population dynamics and trait selection in their host plants, assessed the relative importance of host plant distribution, alternative hosts and environmental factors for variation in predation in a system with three host plants, a specialist and a generalist pre-dispersal seed predator. They concluded that many factors influence the strength of plant-seed-predator interactions, and that their relative importance depended on the degree of specialization involved in the interaction, which can 'result in highly complex selection mosaics and co-evolutionary trajectories'. Cariveau et al. (2004) examined the importance of pollination against pre-dispersal seed predation to selection on plant and floral characters, using *Castilleja linariaefolia*, concluding that the remarkable intraspecific variation in plant and floral characters exhibited by some flowering plants is likely the result of selection driven, at least in part, by pollinators in concert with antagonists, such as pre-dispersal seed predators.

The main purpose of the present study is to investigate insect herbivory as a source of evolutionary change in various floral traits, such as flower size and flowering phenology. Several workers have directly measured factors that might demonstrate that this premise is valid, and much of this work has been undertaken in the search for bio control agents of xenospecies such as members of the *Centaureae*, where they form a significant threat to cropping of commercial cereals. Clement and Sobhian (1991) tested the specificity of Tephritid species feeding on *Centaurea solstitialis* L., the yellow starthistle, with a view to their use in biological control, and Clement (1994) extended this work to various ecotypes of C. *solstitiali*. Story et al. (1982, 1984, 1985), Good (1992), Kinkorova (1991), Rieder et al. (2001), Marshall and Storer (2006) and Crowe and Bourchier (2006), all describe the attacks of *Urophora* species on different *Asteraceae,* noting different aspects of the insects ecology and their effects on predation, whilst Kinkorova and Mickova (2006) show that *Chaetostomella cylindrica* has a more polyphagous nature when compared with the more specialised *Urophorae*. Predation is not limited in its commercial effects to its effects on xenospecies however, Cummings et al. (1999) noted significantly increased predation levels in F1 hybrids of *Helianthus anuus* when compared with natural plants, and speculated that this might be evidence for natural resistance as an acquired trait within populations.

The success of Tephritid species in controlling populations of *Asteraceae* points to their ability significantly to affect fecundity, and this implies a possible evolutionary path if fecundity is differentially affected by varying floral and ecological traits. A number of papers demonstrate that this may indeed the case. Exclusion experiments have shown that insect herbivory limits seed production, Maron (2002), and seedling recruitment Louda et al. (1990), where a 3-fold reduction in viable seeds was noted, leading to a 6-fold decrease in seedling establishment and a 6- to 37-fold reduction in the eventual number of new adults of the Platte thistle *Cirsium canescens* in the USA. Kelly and Dyer (2002) hypothesise that inflorescence-feeding insects can influence population growth rate, and have the potential to act as a selective force for the evolution of traits in *Liatris cylindrica*, an iteroparous perennial, while Louda and Potvin (1995) suggest that insect feeding restricted the phenology of flowering and pollination, concluding that the inflorescence-feeding insect herbivores limited seed production, seedling recruitment, plant density, and maternal fitness of *Cirsium canescens* under natural conditions. Brody (1992) studied plant density dependence on attack frequency in pre-dispersal seed predation, again suggesting a role for pre-dispersal seed predation in the evolution of floral traits, a conclusion shared by Crawley (2000) in a wide ranging overview of seedling recruitment.

Zimmerman and Brody (1998) observed that the Dipteran Hylema, which oviposits on *Ipomopsis aggregata* and *Polemonium foliosissimum*, use some measure of floral morphology in choosing where to lay its eggs. Ehrlen (1996) observed that predation in the legume *Lathyrus vernus* was favoured in individuals with larger inflorescences, and was positively correlated with the average density of seeds within plots. Brody and Morita (2000) conclude that ovipositing insects appear to choose or manipulate flowers to ensure seed set, and thus larval resources. Such effects are not limited to forbs, as Grieg (2003) showed in a study of forest trees in Costa Rica whish suggests that seed predation on the latter species may limit seedling recruitment, and Calvo-Irabien and Islas-Luna (1999) showed that pre-dispersal predation limits seedling recruitment of herbs living within mature forest.

These papers suggest an evolutionary role for seed predation, so it would be natural to investigate which floral traits might be affected, and thus form a basis for further study.

Flower size is a clear candidate for differentially selected predation, as for pollination, and a number of studies have investigated this, and Hainsworth et al. (1984) suggest that *Ipomopsis aggregata* should be under selective pressure to maximize inflorescence height, as well as generation time, although the ecological implications of resource limitation could result in advantages for inflorescences of intermediate height. In further work on *Ipomopsis aggregate* its predators and pollinators, Campbell et al. (2002) looked at experimental arrays of flowers and found that predator oviposition correlated well with flower size, which also affected pollination rates. They again suggested pre-dispersal seed predation as a selective force on floral traits, including size, and Petersen (2000) investigated the effect of flower size on both pollination success and pre-dispersal seed predation, again finding a correlation with both. He also suggested that predatory pressure may act as a constraint to inflorescence size.

Fenner et al. (2002), in work that directly inspired this research showed a consistent preference for larger flower size by predators in a wide range of Asteraceae. Although, as with many studies, these data are relatively restricted in both space and time, begging the question of how such preferences varied throughout the full flowering season, and between populations.

Straw (1989b) described experiments that showed how such flower selection might be attained, as Tephritid females used their abdomen and ovipositor to measure the depth of bracts and thus determine whether or not to lay their eggs. During the course of such research, specifically into the relationship between floral size and predation, it is important not to dismiss the possibility of direct effects of predation itself on the size of the predated flowers. There is little literature that addresses this directly, but Harris and Shorthouse (1996) do provide some evidence that *Urophora. quadrifasciata*, a tephritid that features strongly in the research data below, produced galls in *Centaurea nigra* plants that were relatively low nutrition sinks, not increasing calorific value of the flower head.

Another trait that could be hypothesised as readily affected by differential predation rates is flowering phenology, and several papers have addressed this issue. Albrectsen (2000) studied the relationship between a Tephritid fly and its host plant *Tripolium vulgare* (Asteraceae), concluding that pre-dispersal seed predation, like pollination, may act as a selective force on flowering phenology, and Wright and Meagher (2003), suggest that seed predators play an equally important role in determining flowering phenology as pollinators. However they also note that predation might allow resource reallocation to the production of subsequent flowers, showing the complexity of such interactions. Freeman et al. (2003), in forcing delayed flowering by clipping experiments, noted reduced rates of predation, indicating an escape route from predation by means of changing phenology, but Dierenger (1991) found a greater role for variable rates of growth, determined by local biotic or edaphic factors, to be more important than either pollination or predation in generating phonological variation.

Desteven (1981) looked at the effects of early fruiting of a tropical shrub under predation from moth larvae, and noted that early fruiting avoided predation. She also found that relative sub-population made little difference to predation levels, indicating that foraging ranges of predators can be wide. Both these observations highlight the need to consider predator behaviour as a vital part of any hypothesis regarding their effects on plant evolution.

Following on from Desteven's observations that early flowers can escape all predation, one can look even further than micro evolutionary change. Hendry and Day (2005) speculate that differential flowering times within populations may be a path to speciation. Green et al. (1975) and Fenner (1985) also postulate that pre-dispersal seed predation may affect flowering phenology. Fenner also noted high levels of predation in Asteraceae.

In *Lavandula stoechas*, a Mediterranean shrub commonly occurring in southern Spain, Herrera (1991) studied the effects of pre-dispersal predation. The small flowers aggregate into dense, head like inflorescences, as in the Asteraceae, with large variations in fecundity. Predation accounted on average for a 31% reduction in fruit set, and was found to be non-randomly distributed within inflorescences, with most damage concentrated on late fruits.

As with other traits, pollination rates are also affected by flowering time. Mahoro (2003) found that early flowering was worse for pollination than predation, where in this case the earlier flowering of two related species suffered more severe insect attacks than the late flowering species in two years out of four, this was against his expectation, as in Mahoro (2002) it was noted that pre-dispersal seed predation mitigated the disadvantage of early flowering in Ericaceae, where a previous correlation analysis between flowering schedule and fruit set suggested that early-flowering individuals were at a disadvantage regarding pollinator availability. Such findings suggest that individual patterns of predation and pollination need individual study.

An alternative host avoidance strategy might be to increase the allocation of resources to flower and seed production, and although Fenner (1985) shows, for some species, an inverse relationship between attack patterns and the local density of flower heads, Ohashi and Yahara (2000) both predicted that an increase in predation would actually lead to a reduction in flower production, and observed the same for *Cirsium purpuratum*, concluding that increased flower production may not always improve reproductive success. However, Ohashi and Yahara also noted that, in this species, a greater density of florets or flower heads on a plant did not increase the number of mature seeds produced, so in this plant the total resource allocation might not change in this way.

Hemborg and Despres (1999) studied the effects of predation on *Trollius europaeus*, a forb that exhibits both single and multi-flowered traits. Although the predator in question also acted as pollinator, adding a further selective pressure on the insects, they noted greater attraction to the larger early flowers of the multi headed plants than on the secondary heads, without this resulting in higher overall predation per plant, multi-headed flowers having a greater seed output., although they tended to be rarer. However, their conclusion was that in this specific host-predator interaction the distribution of morphs was more likely to be controlled by environmental factors.

Leimu et al (2002) found considerable differences in populations when studying the effects of selection from seed predation on flower number in *Primula veris*, as did Ehrlén (1996) studying *Lathyrus vernus*. However, in *P. veris* individuals with a higher number of flowers almost always produced more seed capsules than individuals with a lower flower number and even though larger inflorescences had a higher number of damaged capsules in some populations, they still had more undamaged capsules.

Masting is another form both of phenological pattern and resource allocation that has evolved in many plant lineages. In grasses Kelly et al. (1992) found that seed set in a masting species was negatively correlated with seed predation, suggesting that masting may be effective at reducing seed predation in the species under investigation, a conclusion that Kelly and Sullivan (1992) reiterate. Sullivan and Kelly (2000) tried to extend this into some non grass species in New Zealand, but found a strong cross correlation of the effect with altitudinal setting, resulting in masting being most common in higher areas where predation was lower, concluding that in this case the effect of predation was to limit the elevational range of some species that had already evolved the masting habit. The effects of any single parameter in plant insect interactions are clearly both complex and difficult to entangle from the many aspects of life history, as Kelly et al. (2002) note, having shown evidence that, whilst masting in a wide range of plants did produce some advantage in insect predator satiation, it led to a significant and balancing increase in that due to birds, concluding that masting may be favoured by a number of different selective pressures, depending upon the species involved. To complicate the situation further, Forget et al. (1999) found that in a tropical forest it was the post dispersal predators that were sated, rather than the pre-dispersal insects.

Seed production per flower head might also be selected for under pressure from predation, and Herrera (1984) noted a steady increase in fruit attack incidence with increasing number of seeds in the fruit, leading to a significantly increased probability of individual seed success in fruit that contained a single seed over those containing two or more. These effects may differ depending upon the intensity of the predation, as Cipollini and Stiles (1991) discuss. Under intense infestation levels, garden grown beans, which exhibit a negative correlation between seed pod size and seed number, tended to move towards smaller seed size in successive years, even though this resulted in a reduced early growth patterns. Here, a Bruchid, whilst infesting almost all seed pods, tended to concentrate oviposition in the larger seed with the pods. However, as has been seen in many aspects of these relationships, individual interactions have very different outcomes, whilst Briese (2000) noted positive correlations between attack level and seed production in thistles in Greece, Figueroa et al. (2002), studying a montaine forest in Chile found that groups of species suffering similar levels of seed losses to pre- or post-dispersal predators did not share any particular seed characteristics, suggesting that differences in the seed traits studied seem to be relatively unimportant in determining variation in seed predation. Once again, the importance of detailed study on an individual host – predator interaction becomes clear.

Predation has also been postulated as selecting for different colour morphs in a wild radish, and Irwin (2003) suggests that this might counterbalance selection by pollinators, a pattern that reflects itself in a number of different traits including size and phenology. Countering this direct effect, Whitney and Stanton conclude that pleiotropic effects of colour morphism may be a better explanation for differential predation in an acacia, where the predation was nocturnal, and the colour differences could not be detected visually. Mack (2000) has suggested that seed predation might have driven the evolution of fleshy fruits, only secondarily becoming structures that promote dispersal, and Raghu et al. (2005) demonstrate that early seed pod dehiscence can control rates of seed loss from pre-dispersal predation.

These studies demonstrate the wide range of traits that might be affected by differential predation, as well as the complex nature of host predator interactions, which, from an evolutionary perspective may easily be masked by purely ecological effects, noted by Janzen (1975) in studies on Barro Colorado, leading him to comment on the importance of studying individual interactions. As Fenner and Lee (2002) show, and the large body of work devoted to the use of seed predators as biological control agent, lack of predators can set species free in new environments. Such new environments might not need to be as distinct as New Zealand for the European Asteraceae studied by Fenner and Lee. Rand (2002) shows how herbivore pressure might maintain distinct abundance patterns in intertidal species of a salt marsh. Similar points are made by Reader (1992), reflecting on a study of variation in density of three forb species in ridge-hollow patterns of ancient cultivation, supporting theories that consider differential herbivory to be a primary control of plant frequency on an environmental gradient. That local plant densities can be affected is demonstrated by various works, Kaye (1999) suggested that predation was a serious threat to local populations of a rare Astralgus species in the western USA, work similar to that of Bigger (1999), and Leimu and Syrjanen (2002) stressed the difference in predation levels on a population level rather than local density fluctuation.

Clearly many extraneous factors can mask or mitigate against evolutionary pressures solely from predation. Requirements for pollination have long been expected to produce evolutionary pressure to increase seed production that runs directly counter to predation. In a series of papers on the pollinators of Centaurea species Lack (1976) (1982) proposed pollination pressure as a source of floral morph difference and phenological differences. Galen (1996) observed rapid morphological change in corolla form of an alpine flower in experimentally controlled pollination, whilst Young (2002), investigating differences between day and night visitors hypothesised that floral phenology is an adaptation to expose flowers to the most effective pollinators. Armbruster (2001) studied detailed floral form adaptation in terms of the physical mechanism of pollen transfer, revealing the different levels of detail at which interactions may be investigated.

As well as the differing pressures from species interactions such as pollination and predation that affect plant ecology and evolution, plant-insect interactions are all two way processes, with gains and losses made by pollinator and predator as well as hosts, and many researchers have looked at them from this perspective. Although this research is mainly based upon the effects on plants, it must also be informed by this view, indeed Duggan (1985) concluded that in Lady's Smock, the plant-herbivore relationship is asymmetric, with the plant having a much greater effect on the herbivore than the herbivore has on the plant, and Leimu and Lehtila (2006) conclude that leaf herbivory contributed more strongly to host plant fitness than seed predation in the perennial herb *Vincetoxicum hirundinaria.*

There are several general descriptions and overviews of pre-dispersal seed predating insects and their attack patterns. Freese (1994) conducted a wide ranging study of the phytophagous insects of seven thistles, Straw (1989a) presents an overview of the guilds of Tephritid herbivores, and their attack patterns, and Zwolfer and Stadler (2004) describe the organization of phytophagous guilds in *Cardueae* flower heads. More specifically Riverolynch and Jones (1993) made experimental studies of the specific ovipositional behaviour of a Tephritid in *Cirsium palustre*. In terms of their evolutionary effects on hosts, the ecology of predators might have important connotations, especially in terms of the level of selective pressure that they apply. Dempster Atkinson and Cheesman (1995), demonstrated the variability of local predation on populations of *Centaurea nigra* over a number of years. Local extinction was found to be a regular feature of these populations, especially on the smaller patches of the plants, offering the possibility of relief from selective pressure for the plants, while in the second part of the same study, Dempster Atkinson and French (1995) show that how predator migration can re-introduce selective pressure into plant populations, postulating plant density dependence as a factor increasing probability of migration. In a further study, Halley and Dempster (1996) found that although the rate of immigration appeared proportional patch size, distance between patches was significantly less than the measured dispersal distance of all the species studied. Schlumprecht (1989) also studied Tephritid dispersal rates, calculating an annual migration rate of up to 100m per generation.

Host plant ecology can also affect their predators' behaviour, as Lalonde and Roitberg (1994) showed, where local pollen shortage in diecious *Cirsium arvense*, leading to low seed availability, significantly reduced oviposition choice, and van Poeke and Dicke (2002) demonstrated a mechanism whereby volatiles released during herbivory was used as a cue by the herbivore's parasitoid.

Although insects may be tied to individual hosts, it is rare for hosts only to have a single herbivore, which might lead to interspecific competition for oviposition sites, or character displacement to separate overlapping predator niches. Berube (1980) found just such evidence in the attacks of the Tephritidae *Urophora affinis* and *Urophora quadrifasciata* on *Centaurea diffusa*, although Burkhardt and Zwolfer (2002) did not find such evidence between *Urophora jaceana* and *Urophora quadrifasciata*, both predating *Centaurea jacea* as host plant, perhaps due to the different levels of specificity of the predators. In a further study of *Urophora affinis* and *Urophora quadrifasciata* Myers and Harris (1980) suggested that pheromones may be used to avoid competition for oviposition sites, Frenzel et al. (1990) identified just such a pheromone in the rectal ampulla of male *Urophora cardui* and *Urophora stylata*.

Competition can lead to complex behaviour. Reader (2003) describes that between two gall formers in reeds, where the favoured competitor demonstrates facultative kleptoparasitism by taking over the galls of the other, increasing its food supply and in turn improving its defence against its parasitoid wasp, and Williams et al (2001) show how both *Terellia ruficauda*, infesting *Cirsium palustre*, and its parasitoids appear to select their oviposition sites to spread the risk of losses through factors such as mammal herbivory that may damage dense clusters of their hosts.

As with their ecology, insect evolution is clearly affected by their interactions with hosts. Abrahamson et al. (2001) and Abrahamson et al. (2003) provide evidence of sympatric speciation in insects due to floral evolution producing genetically differentiated and reproductively isolated host races, and Berube and Myers (1983) describe the reproductive isolation of *Uphora* species in Centaurea. In another example of character displacement, Morrow et al (2000) describe the close genetic similarity between two sympatric species of tephritid fruit fly that predate the same fruits, but are reproductively isolated by mating time. Hosts and predators are well known to follow co-evolutionary paths, although it may be very hard to determine how each event affects the next, although Brandle et al. (2005) carried out a phylogenetic study of thistles and their predators, finding in general that Urophora speciation events lagged behind those of their hosts.

From all of the above there is abundant evidence of pre-dispersal seed predation acting as a potential selective pressure on floral traits, albeit within an extraordinarily complex set of overlapping plant-insect interactions. In order to turn such clues as are found in the literature of predation into a realistic hypothesis that predation is indeed a selective pressure in certain individual interactions, information on a number of areas of plant and insect life needs to be determined. Firstly, in any evolutionary explanation, there must be evidence that the traits under study are actually heritable, rather than being the result of extra-somatic factors. Once again, the literature provides much evidence that the floral traits under study do have heritable components.

Geber and Griffen (2003) review a wide range of papers, with a range of results, but showing various traits to be heritable, as well as identifying some specific genetic sites of selection. Mazer and Delesalle (1996) note the difficulty of estimating the genetic component of phenotypic variation in modular organisms, as their phenotype changes with age, but they measured sufficient genetic variation among populations of *Spergularia marina* to suggest that floral traits are open to evolutionary change. Campbell (1996) measured genetic variances, heritabilities, and genetic correlations of floral traits in *Ipomopsis aggregata,* demonstrating several traits open to evolutionary selective pressure.

More specifically, flower size and form, have been shown to be heritable, such as the discoid shape of the *Leucanthemum vulgaris* Bogle (1983), corolla width in the of wild radish *Raphanus sativus* Young et al (1994), flower size, and thus seed production, in *Mimulus guttatus* van Kleunen and Ritland (2004) and both flower size and number Worley and Barrett (2001).

The heritability of phenology is well researched in a range of plant families. Akeroyd and Briggs, (1983) showed that flowering time is preserved in *Rumex crispus* when it was transplanted from different populations that themselves flowered at differing times, and Pors and Werner (1989) showed how flowering ranks within a long established population of *Solidago Canadensis* a clonal plant. Transplant experiments showed that this ranking was maintained over several years in a new environment. Similar experimental evidence was found in *Senecio intergrifolius* by Widen (1991) who noted ‘a significant correlation between phenological rank order of mother plants in natural populations and their progenies in cultivation’.

Ollerton and Lack (1998) studied similar traits in *Lotus corniculatus*, again noting a strong correlation between phenology of individual plants over a number of years, in a population with significant variation of flowering among its individuals. They also found that flowering time was correlated with plant size. Pilson (2000) studied the phenology of *Helianthus annuus* in the light of pre-dispersal seed predation. Having determined a genetic variability in flowering time that was heritable, he hypothesised phenology to be open to selective pressure by herbivory. Further analysis identified two out of a number of herbivorous predators that accounted for all selection on phenology, demonstrating that, in this species flowering phenology is a direct consequence of insect attack.

Heritability may not always be evident in the simple Mendelian sense as Case et al. (1996) discovered. Environmental factors that affected flowering traits of one generation of *Plantago laneoleta* appeared to affect both second and third generation seedlings, in a complex environmental genetic interaction, and Meagher et al (2005) suggest that DNA content in *Silene latifolia* correlates well with flower size variation, indicating a quantitative model rather than a direct Mendelian for floral evolution, a conclusion also found by Hansen et al. (2003), who predicted however that most floral traits would not be expected to change by more than a fraction of a percent per generation in *Dalechampia scandens* (Euphorbiaceae).

Much work has been done at the molecular level in recent years, much of it adding to knowledge of the complicated nature of evolutionary paths. Worley and Barrett (2000) found significant links between genes controlling resource allocation and genes with opposing effects on flower size and number, and suggest that these are due to the hierarchical nature of floral display, but that these effects are themselves means whereby evolutionary selection might work. Armbruster (1997) showed that both defence and pollination attraction adaptations may be derived from the same pre-cursors.

Plant genetics and evolution can often differ from standard Mendelian models, polyploidy being a prime example, and the several different life cycles can further complicate matters. In dioecios plants, there can be significant differences between the effects of paternal and maternal lines, such as demonstrated by Kelly (1993) in *Chamaecrista fasciculata*, a temperate summer annual, where paternal families showed 'no evidence of heritable variation for two estimates of plant size, six measures of reproductive phenology or two fitness components' whilst 'maternal estimates of heritability suggested the influence of maternal parent on one estimate of plant size and four phenological traits'. Gynodioecity offers another life cycle that can be influenced be insect interaction, and both Marshall and Ganders (2001) and Asikainen and Mutikainen (2005) suggest differential predation on hermaphrodite and female flowers as a means of sustaining it.

Once again, published works show the complex nature of plant-insect interaction, making simple hypotheses hard to sustain, and suggesting that although there may be strong selective pressure for floral evolution in some cases, moving from the specific to the general would be hard to argue, and thus that study should concentrate on individual detail. Such difficulties are augmented by the many other interactions that plant species are involved in. Pollinator traits, for instance are shown to correlate with flower size in 15 Asteraceae, Dlussky et al. (2004). Acosta et al. (1997) show how differential resource partitioning as plants age may affect fitness at different ages, Gange et al. (2005) looked at the effects of mycorrhizal symbiosis in *Leucanthemum* vulgare and found that it increased floral traits leading to increased infestation, as did root herbivory in *Cirsium palustre* Masters et al. (2001). Mammal herbivory can often affect local populations, although Amsberry and Maron (2006), studying Asteraceae in the USA concluded that in general insect herbivory is a greater cause of seed loss. Habitat fragmentation can significantly reduce the effects of predation Chacoff et al. (2004), allowing refuges of original genetic traits to be maintained within meta-populations, whilst predation can itself maintain equality in fertility levels between related species by depressing fitness in one that would normally exhibit increased fertility, Lavergne et al. (2005).

Complex systems often resolve themselves into stable states, with deviations being restored by a number of compensating feedback systems, which may well be the basis for the pattern of evolutionary stasis observed over long terms. The various interactions between pre-dispersal seed predators, pollinators, host plant, parasitoids, mycorrhizal fungi, and many other organisms that affect each other may well be a case in point. Many publications have noted the opposing selective pressures that form the basis of these patterns. Most studied are the differing pressures of pollination and predation in direct opposition to each other. Brody (1997) and Brody and Mitchell (1997) studying *Ipomopsis aggregata* note the importance of both pressures, emphasising the need for studies to look at both if their evolutionary impacts are to be studied. Kudoh and Whigham (1998) found countreracting pressures between pollinators and predators on floral size, measured by petal length, in *Hibiscus moscheutos*, and Herrera (2000) analysed the sign and magnitude of selection on *Paeonia broteroi* from each, and found them to be opposing, and of roughly similar magnitude, a conclusion similar to those of Galen and Cuba (2001) and Ehrlen et al. (2002).

Further studies have included other factors together with pollination and predation. Steffan-Dewenter et al. (2001) looked at the effects of habitat complexity, and found that both the number of flower-visiting bees and the proportion of flower heads damaged by seed predators increased with landscape complexity, however the mean number of seeds set per flower head did not increase which they attributed to the counterbalancing effects of pollination and seed predation. Brys et al. (2004), working with *Primula vulgaris*, a declining plant in Belgium, also looked at the effects of habitat fragmentation. They found that predation increased with floral display, but that the effect was not sufficient to be a selective pressure over large display for improved pollination. In a very different environment, the cloud forests of the Columbian Andes, similar conclusions were made by Garcia-Robledo et al. (2005) who conclude that ‘our study suggests that the interaction of two ecological processes, pollination and pre-dispersal seed predation, may cancel each other's effects under natural conditions’.

It may not always be pollination in opposition to predation. Geritz (1998) studied the evolution of seed size under the Evolutionary Stable Strategy approach, in a co-evolutionary model of seed size and predation, predicting that the ESS would result in significant seed size variation, while Graham (1995) showed that competing species of galling insects strike their own balance in their parasitism that does not excessively threaten the plant species on which it depends.

Clearly the effects of pre-dispersal predation and pollination are well studied. But these papers point to an over-riding complexity that demands not only the study of plant insect interactions on an individual case by case basis, but also a longer term approach that can look at the longer term effects and competing pressures that result in the enormous variety of symbioses, mutualisms, parasitisms and commensalisms that characterise them.

Chapter 3. Field research

In order to address the first four hypotheses outlined in the introduction; that predators tend to select larger flower heads as oviposition sites in a repeatable pattern, that the presence of seed predators within flower heads reduces their fecundity, that successful seed set is correlated to flower size, and that the pattern of seed predation over the course of a season is repeatable over several seasons, a programme of field research was undertaken. This programme has consisted of identifying naturally growing populations of Asteraceae, and sampling them by means of flower head removal and inspection to determine levels of predation and pollination success, at regular intervals throughout a number of growing seasons.

3.1 Population search

Species suitable for study were identified initially, in conjunction with Dr. Fenner, by considering a combination of availability in the area, previously collected data, and suitability of seed size. Populations of suitable plant species were identified by a geographic search of sites close to roads in South Hampshire. The search started in the ecology meadows at Chilworth, and moved north and west from this point. The following locations were used:

Cirsium palustre

This species was only sampled in 2001. Levels of predation were found to be generally low, indicating that for this species seed-predation may not be a significant selective force on floral size or phenology, so following discussion with the project supervisor it was decided to curtail its use in the study.

Site 1	Old orchard at Chilworth. SU 405 183. 20m x 10m.
Site 2	Meadow edge at Chilworth. SU 407 183. 40m x 5m.
Site 3	Forestry commission’s drive in Castle Lane Chandlers Ford. SU 423 197. 50m x 20m.

Tripleurospermum inodorum

This species was sampled in 2002 and 2003. Following investigation of the effects of infestation on seed set it was found that the infesting larvae were not significantly reducing fecundity as they remained within and consumed only the receptacle material, damage which appeared not to influence seed set. Clearly in this species the second hypothesis is not supported, demonstrating, as with *Cirsium palustre* that the effects under investigation are not universal within the Asteraceae.

Site 1 Roadside verge opposite Little Somborne church. SU 382 327. 10m x 3m.
Site 2 Farm entrance in lane from Little Somborne to Sandy Down. SU 388 334. 10m x 10m.
Site 3 Field track close to Farley Mount Winchester. SU 387 298. 10m x 5m.

Leucanthemum vulgare

Site 1 2001, a site close to the orchard at Chilworth. SU 406 183. 10m x 10m.
2002 onwards a site in the meadow edge at Chilworth. SU 407 183. 30m x 30m.
Site 2 Roadside bank close to the western entrance to Shawford Down. SU 467 248. 10m x 4m.
Site 3 Roadside bank at the new Hockley roundabout Winchester. SU 473 264. 20m x 8m.

Centaurea nigra

Site 1 Old orchard at Chilworth. SU 406 183. 5m x 5m.
Site 2 Open meadow at Chilworth. SU 407 183. 10m x 5m.
Site 3 Open downland at Shawford. SU 472 248. 10m x 10m.

At each site, a boundary was determined within which sampling took place. For some sites, the boundary was set by the population itself, where this could be seen to be discrete, otherwise it consisted either of natural features such as the roadside or a track or field edge, or was laid out with bamboo canes at each corner.

3.2 Flower head collection

For each species, flower heads were gathered only at a specific stage of flowering:

Cirsium palustre	all florets visible, no browning of florets.
Tripleurospermum inodorum	floret discs all yellow and rounded, no browning of florets.
Leucanthemum vulgare	between 10% & 100% of florets open, no browning of florets.
Centaurea nigra	all florets visible and head opening out, no browning of florets.

The criteria for collection were selected to correspond with the period of floret maturation, when resource flow to the florets would be continuous. In order to obtain data relating to the overall period of flowering of each population the density of open flowers at each site, and on each visit, was measured by counting numbers within a defined area of the site.

Sites were visited twice weekly during the periods when flowers were open, and on each visit heads were removed from random positions within the plots. In each plot, twenty random positions were calculated before collection. This process was carried out in a set of square axes, and the resulting coordinate pairs located approximately within the plots visually. Once at the coordinate point a central flower head was identified, and the nearest five or ten suitable heads were removed.

Where possible, 100 heads were sampled at each visit. Where less than 100 heads were available, all suitable heads in the plot were taken. The heads were placed in plastic bags labelled with the species name, site name and date. Samples were analysed within two to three days of sampling.

2001 Samples were taken from the populations of *Cirsium palustre*, *Leucanthemum vulgare* and *Centaurea nigra*.

2002/2003 Samples were taken from the populations of *Leucanthemum vulgare*, *Centaurea nigra* and *Tripleurospermum inodorum.*

2004-2006 Samples were taken from the populations of *Leucanthemum vulgare* and *Centaurea nigra*.

This method of collection was determined as a practical solution to a common method of locating flowers within the plots, which varied in size from 40 to 1000 square metres in size, where some 100 heads were to be sampled. It might be suggested that it opens the prospect of some non-independence of data as ovipositing and pollinating insects might follow a similar pattern of infesting or visiting certain patches within the sampling area rather than randomly moving from flower to flower. This is a criticism that has to be taken as a possibility and one that was not specifically considered at the start of the project. However, within the smaller patches, the location of 100 flower heads within areas of up to 100 square metres by this method, rather than by calculating 100 separate locations is more practical, and the average distance between locations actually sampled varies between 2 and 5 metres for plots of 40 and 100 sq. metres respectively. In addition, the length of time that plant heads remain within available for sampling is generally longer than the intervals between sampling, allowing at least two opportunities for an individual head to be collected.

The possibility of non-independence would be more serious if the data showed high levels of density dependence in levels of infestation, which would indicate a relatively small foraging range for individual insects, and this aspect has been studied in section 3.7. However, the criticism remains, and perhaps suggests that the results of analysis be judged by a more stringent standard when applying statistical techniques, $p<0.02$ rather than $p<0.05$.

One further criticism of the technique is that of destructive sampling. This might have some validity at the extremes of date sampling where some plots were sampled for all of their heads. The data relating to the phenology of flowering could be examined firstly to see if this affected flower density over the course of the five years, and secondly in relation to frequency of infestation to see whether clear sampling at extreme dates might show an effect.

3.2.1 Measurement technique

For each species a characteristic flower dimension was identified to serve as a measure of flower size:

Cirsium palustre	external capitulum diameter.
Tripleurospermum inodorum	internal receptacle width.
Leucanthemum vulgare	internal receptacle width.
Centaurea nigra	external capitulum diameter.

All measurements were made by vernier calliper. For the first half of the 2001 data, a standard calliper with visual reading of the vernier was used. For all later readings a calliper with digital readout was used. Readings were recorded to the nearest 0.1mm.

For species with external dimension, the capitulum diameter was measured directly, ensuring that no additional tissue was encompassed. For species with internal measurement, the head was cut in two with a scalpel and the callipers overlaid onto the receptacle. Flower dimensions were recorded on a paper run sheet for later entry onto a laptop computer.

Following the measurement, the flower heads were cut into four equal parts and visually inspected for the presence of fly larvae, no attempt was made to identify unhatched eggs. In general, infestation was recorded on the run sheet as either present or not.

For *C. palustre* and *T. inodorum*, the type of infestation noted was consistent for all infested flower heads. In the course of 2001 it was noted that *C. nigra* was infested both by free-living non gall-forming larvae, and by gall-forming larvae. In 2002, all *C. nigra* heads were recorded as either being free of infestation, galled, non-galled, or both galled and non-galled.

From 2003 onwards, infestation of *L. vulgare* was discriminated between Tephritid larvae living within the florets, and receptacle based larvae of a midge, not being considered within the project.

3.2.2 Characteristics of the floral populations

The sampling of populations in flower during the course of the research resulted in large sets of data on flower size and infestation for four species, which are summarised in Table 1.

Table 1 Descriptive statistics for all sampled flowers

Cirsium palustre

Descriptive Statistics

N	3836
Minimum	3.90
Maximum	11.30
Mean	6.95
Std. Deviation	1.02

Tripleurospermum inodorum

Descriptive Statistics

N	9189
Minimum	2.50
Maximum	16.20
Mean	9.42
Std. Deviation	2.07

Leucanthemum vulgare

Descriptive Statistics

N	13900
Minimum	1.40
Maximum	12.50
Mean	6.63
Std. Deviation	1.62

Centaurea nigra

Descriptive Statistics

N	23507
Minimum	5.30
Maximum	15.70
Mean	10.02
Std. Deviation	1.31

In order to investigate the presence of predator choice within these samples, an initial investigation of the data is required to determine the basic nature of the data. The samples can be separated into categories on the basis of predator choice for oviposition, by placing them into groups by the presence of, and type of, infestation observed when dissected. Looking at these categories by sampling site and year reveals the following.

Table 2 Sample count and frequency of infestation

Cirsium palustre (2001)

	Frequency			Percent		
	Valid			Valid		
Site	Uninfested	Infested	Total	Uninfested	Infested	Total
1	1440	18	1458	98.8	1.2	100.0
2	1287	23	1310	98.2	1.8	100.0
3	1012	56	1068	94.8	5.2	100.0

Tripleurospermum inodorum

		Frequency			Percent		
		Valid			Valid		
Site	Year	Uninfested	Infested	Total	Uninfested	Infested	Total
1	2002	1329	1061	2390	55.6	44.4	100.0
	2003	1131	274	1405	80.5	19.5	100.0
2	2002	1538	1097	2635	58.4	41.6	100.0
	2003	831	455	1286	64.6	35.4	100.0
3	2002	217	257	474	45.8	54.2	100.0
	2003	601	398	999	60.2	39.8	100.0

Leucanthemum vulgare (Floret infestation by *T. neesii*, receptacle by a midge)

Infestation

		Frequency					Percent				
		Valid					Valid				
Site	Year	Uninfested	Floret	Receptacle	Both	Total	Uninfested	Floret	Receptacle	Both	Total
1	2003	281	608	5	6	900	31.2	67.6	.6	.7	100.0
	2004	143	139	29	35	346	41.3	40.2	8.4	10.1	100.0
	2005	216	287	10	27	540	40.0	53.1	1.9	5.0	100.0
	2006	211	232	8	11	462	45.7	50.2	1.7	2.4	100.0
2	2003	577	185	250	115	1127	51.2	16.4	22.2	10.2	100.0
	2004	290	63	179	70	602	48.2	10.5	29.7	11.6	100.0
	2005	542	31	108	17	698	77.7	4.4	15.5	2.4	100.0
	2006	426	46	102	9	583	73.1	7.9	17.5	1.5	100.0
3	2004	853	75	144	21	1093	78.0	6.9	13.2	1.9	100.0
	2005	962	39	77	8	1086	88.6	3.6	7.1	.7	100.0
	2006	658	50	45	6	759	86.7	6.6	5.9	.8	100.0

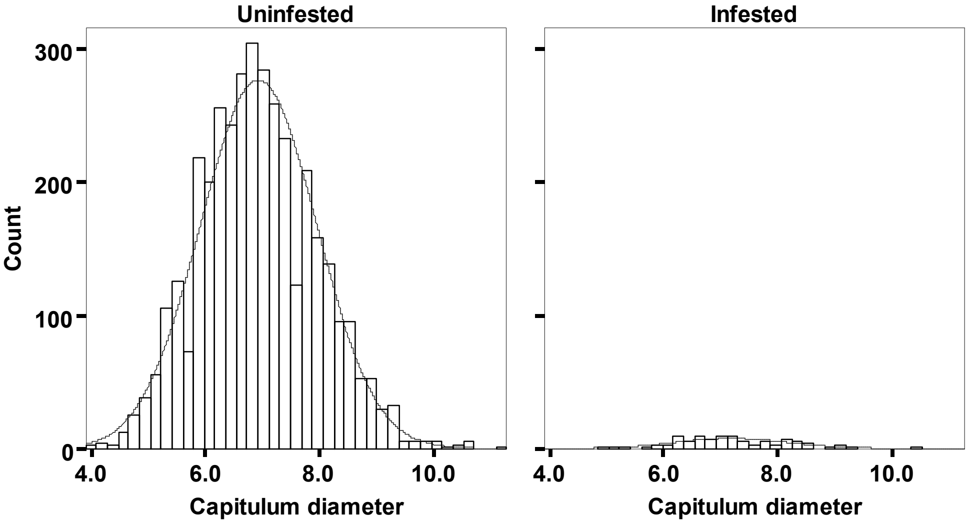
Centaurea nigra **(Non-gall infestation by *C.cylindrica*, gall by a Urophora spp.)**

Infestation

		Frequency					Percent				
		Valid					Valid				
Site	Year	Uninfested	Non-gall	Gall	Both	Total	Uninfested	Non-gall	Gall	Both	Total
1	2002	859	724	84	1	1668	51.5	43.4	5.0	.1	100.0
	2003	311	897	185	34	1427	21.8	62.9	13.0	2.4	100.0
	2004	940	609	245	12	1806	52.0	33.7	13.6	.7	100.0
	2005	684	389	320	15	1408	48.6	27.6	22.7	1.1	100.0
	2006	591	418	141	9	1159	50.9	36.0	12.2	.8	99.9
2	2002	503	456	366	25	1350	37.3	33.8	27.1	1.9	100.0
	2003	305	740	234	47	1326	23.0	55.8	17.6	3.5	100.0
	2004	720	427	344	12	1503	47.9	28.4	22.9	.8	100.0
	2005	577	321	307	13	1218	47.4	26.4	25.2	1.1	100.0
	2006	481	250	203	10	944	51.0	26.5	21.5	1.1	100.0
3	2002	622	440	545	7	1614	38.5	27.3	33.8	.4	100.0
	2004	1032	512	451	44	2039	50.6	25.1	22.1	2.2	100.0
	2005	787	373	400	18	1578	49.9	23.6	25.3	1.1	100.0
	2006	742	252	373	32	1399	53.0	18.0	26.7	2.3	100.0

In order to investigate whether flowers chosen for oviposition show a significantly different average size from those uninfested, some statistical test is required. Choice of statistical test requires further investigation of the nature of the distribution of the data in each category. Normally distributed samples can be investigated by simple ANOVA if they are normally distributed, and also pass the test for homogeneity of variance. Normal samples with non-homogeneous variance can be testes using the t statistic, if comparing only two samples. Plots of the distribution of sizes on the basis of infestation are shown in Figure 1, together with the results of a Kolmogorov-Smirnov test of normality, and Levene’s test for homogeneity of variance.

Figure 1 Size distribution of all sampled flower heads
***Cirsium palustre* (2001)**



One-Sample Kolmogorov-Smirnov Test

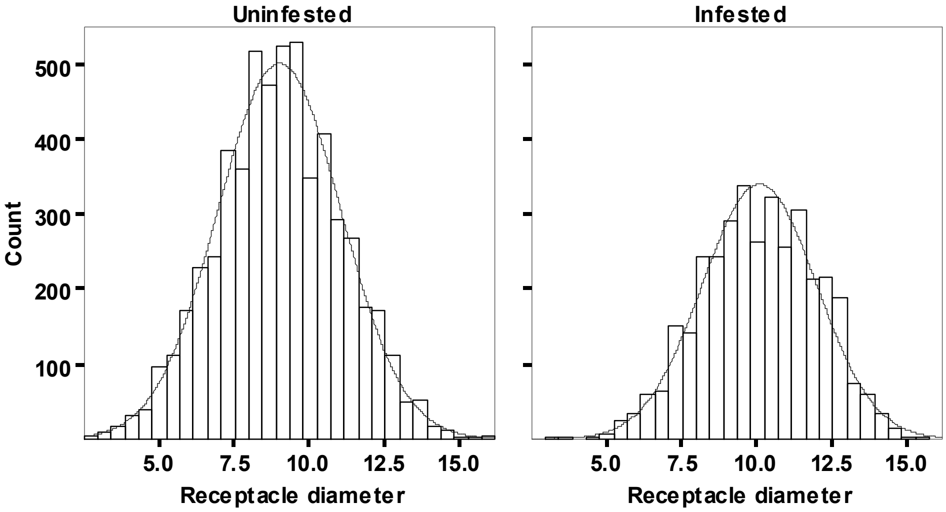
Infestation			Capitulum diameter
Uninfested	Normal Parameters	Mean	6.939
		Std. Deviation	1.024
	Kolmogorov-Smirnov Z		2.330
	Exact Sig. (2-tailed)		1.000
Infested	Normal Parameters	Mean	7.207
		Std. Deviation	.956
	Kolmogorov-Smirnov Z		.787
	Exact Sig. (2-tailed)		.539

Test of Homogeneity of Variances

Capitulum diameter

Levene Statistic	df1	df2	Sig.
1.346	1	3834	.246

Tripleurospermum inodorum (2002-2003)



One-Sample Kolmogorov-Smirnov Test

Infestation			Diameter
Uninfested	N		5647
	Normal Parameters [a,b]	Mean	9.0070
		Std. Deviation	2.05452
	Kolmogorov-Smirnov Z		1.504
	Exact Sig. (2-tailed)		1.000
Infested	N		3542
	Normal Parameters [a,b]	Mean	10.0911
		Std. Deviation	1.90162
	Kolmogorov-Smirnov Z		1.975
	Exact Sig. (2-tailed)		1.000

a. Test distribution is Normal.

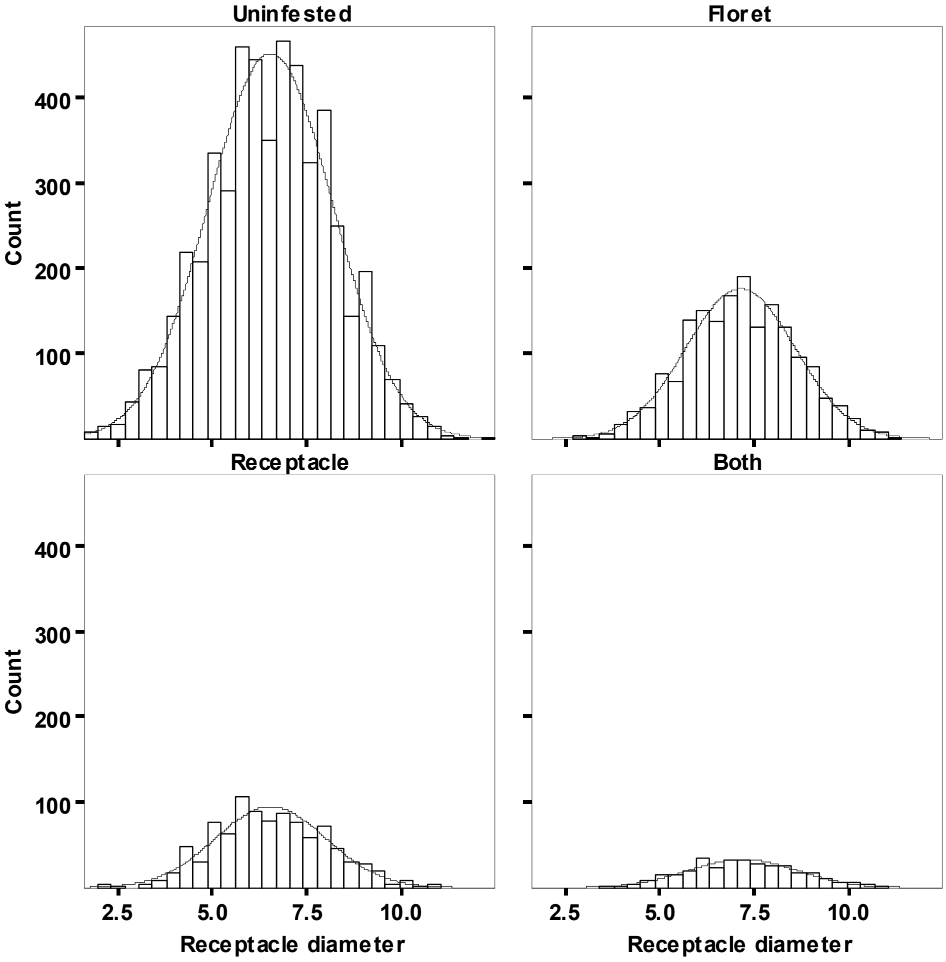
b. Calculated from data.

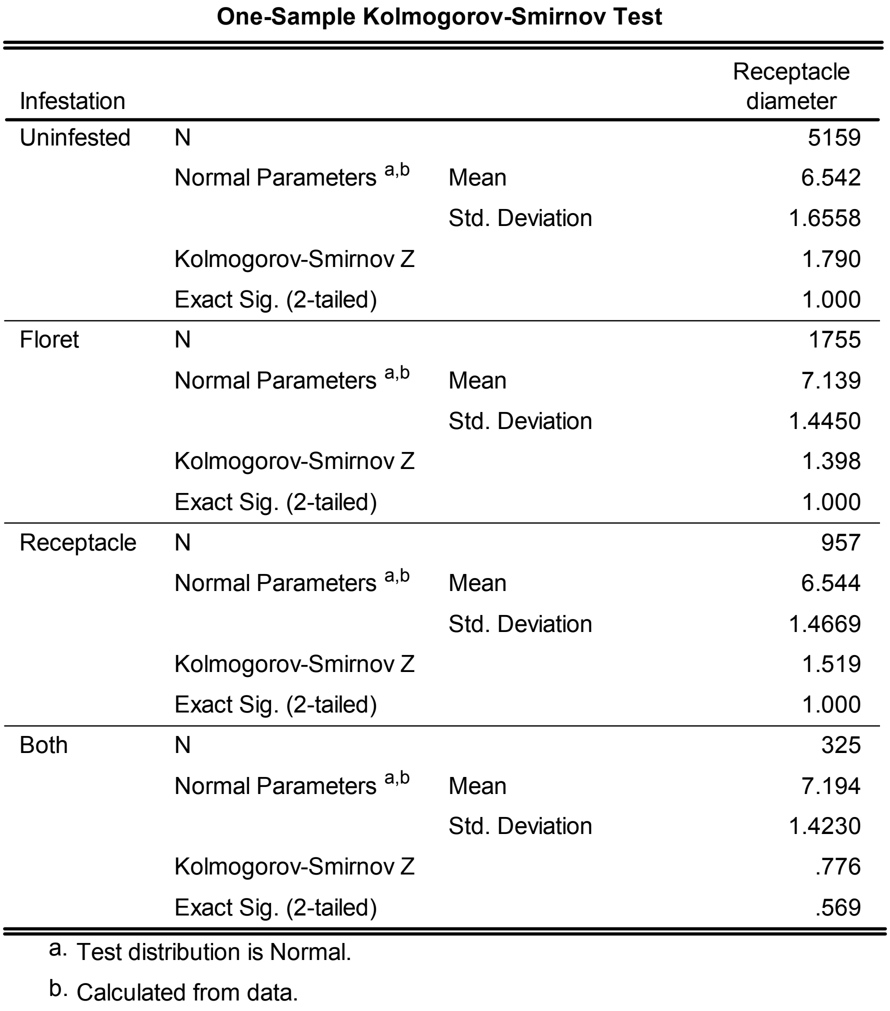
Test of Homogeneity of Variances

Diameter

Levene Statistic	df1	df2	Sig.
10.456	1	9187	.001

Leucanthemum vulgare (2003-2006)



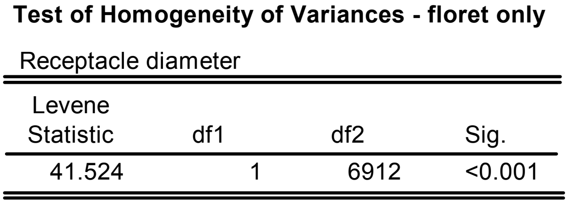


One-Sample Kolmogorov-Smirnov Test

Infestation			Receptacle diameter
Uninfested	N		5159
	Normal Parameters [a,b]	Mean	6.542
		Std. Deviation	1.6558
	Kolmogorov-Smirnov Z		1.790
	Exact Sig. (2-tailed)		1.000
Floret	N		1755
	Normal Parameters [a,b]	Mean	7.139
		Std. Deviation	1.4450
	Kolmogorov-Smirnov Z		1.398
	Exact Sig. (2-tailed)		1.000
Receptacle	N		957
	Normal Parameters [a,b]	Mean	6.544
		Std. Deviation	1.4669
	Kolmogorov-Smirnov Z		1.519
	Exact Sig. (2-tailed)		1.000
Both	N		325
	Normal Parameters [a,b]	Mean	7.194
		Std. Deviation	1.4230
	Kolmogorov-Smirnov Z		.776
	Exact Sig. (2-tailed)		.569

[a] Test distribution is Normal.

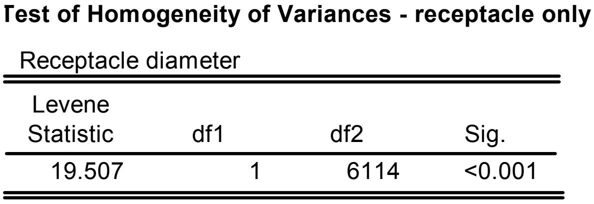
[b] Calculated from data.



Test of Homogeneity of Variances - floret only

Receptacle diameter

Levene Statistic	df1	df2	Sig.
41.524	1	6912	<0.001

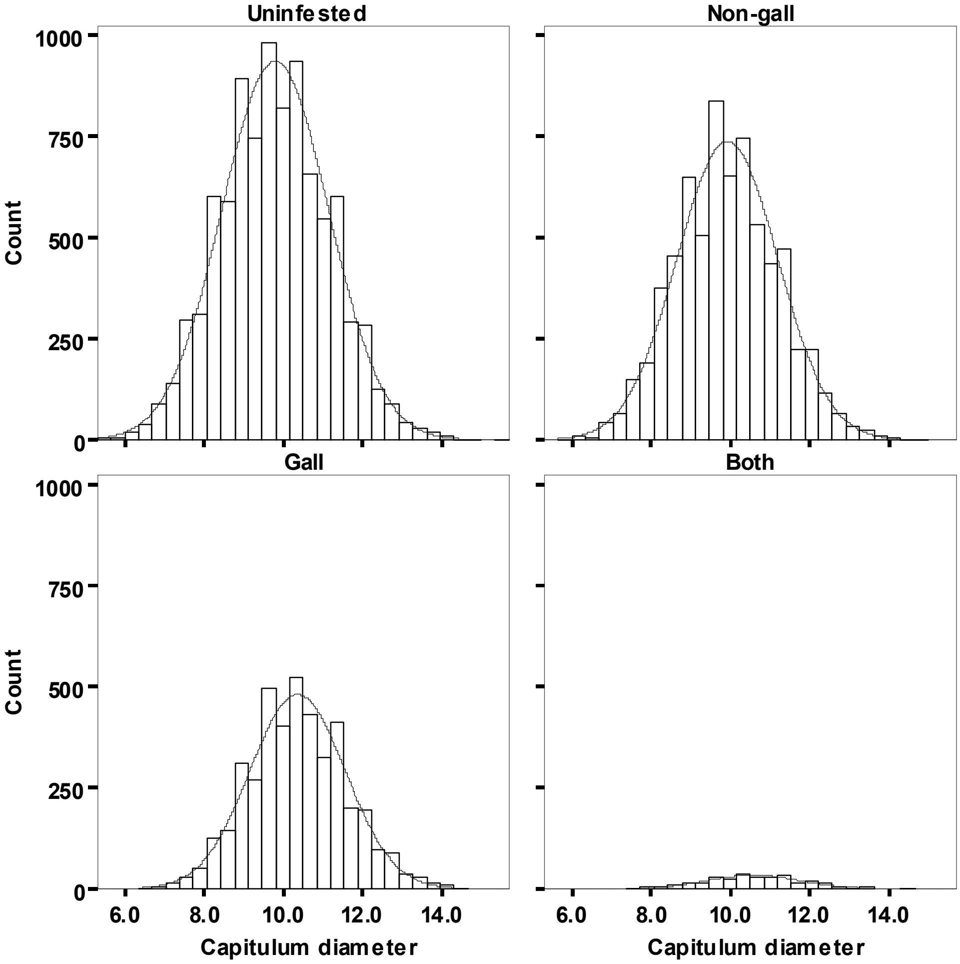


Test of Homogeneity of Variances - receptacle only

Receptacle diameter

Levene Statistic	df1	df2	Sig.
19.507	1	6114	<0.001

Centaurea nigra (2002-2006)



One-Sample Kolmogorov-Smirnov Test

Infestation			Capitulum diameter
Uninfested	N		9154
	Normal Parameters [a,b]	Mean	9.794
		Std. Deviation	1.3540
	Kolmogorov-Smirnov Z		2.110
	Exact Sig. (2-tailed)		1.000
Non-gall	N		6808
	Normal Parameters [a,b]	Mean	9.926
		Std. Deviation	1.2769
	Kolmogorov-Smirnov Z		2.075
	Exact Sig. (2-tailed)		1.000
Gall	N		4198
	Normal Parameters [a,b]	Mean	10.362
		Std. Deviation	1.2089
	Kolmogorov-Smirnov Z		1.863
	Exact Sig. (2-tailed)		1.000
Both	N		280
	Normal Parameters [a,b]	Mean	10.569
		Std. Deviation	1.1785
	Kolmogorov-Smirnov Z		.592
	Exact Sig. (2-tailed)		.862

a. Test distribution is Normal.

b. Calculated from data.

Test of Homogeneity of Variances - non gall only

Capitulum diameter

Levene Statistic	df1	df2	Sig.
14.622	1	20158	<0.001

Test of Homogeneity of Variances - gall only

Capitulum diameter

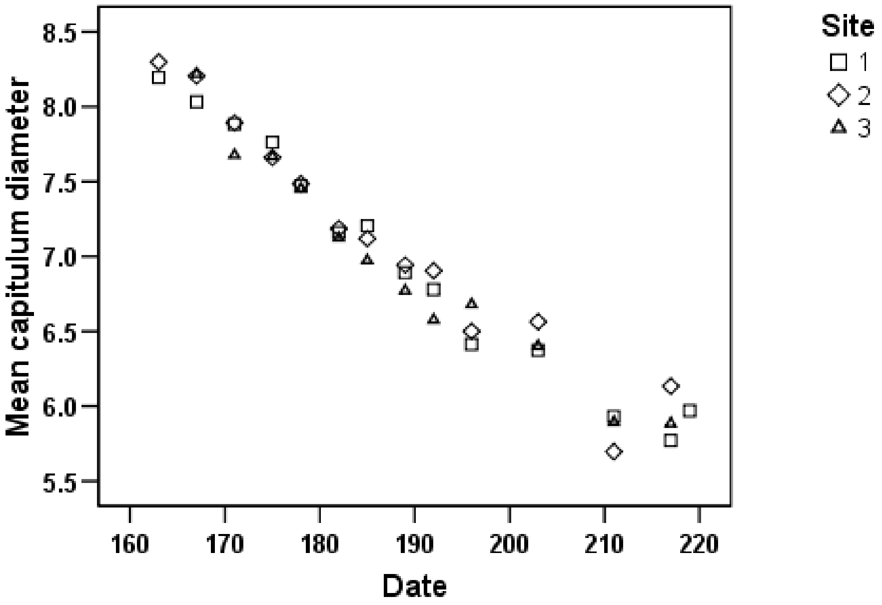
Levene Statistic	df1	df2	Sig.
52.921	1	20158	<0.001

With exact significance of 1.000, the main groupings of data show that acceptance of the nul hypothesis of normality can be accepted, with the exception of those where the number of cases is small in comparison with the total sample numbers, *Cirsium palustre* infested flowers, and flowers of *Leucanthemum vulgare* and *Centaurea nigra* that are infested by both of the predators identified.

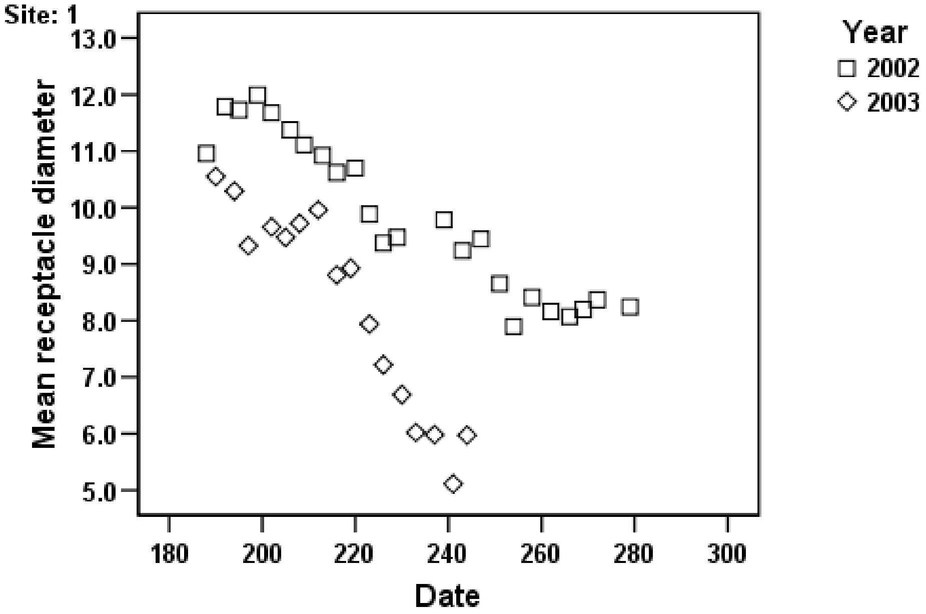
However, most of the samples fail the test for homogeneity of variance, indicating that ANOVA might give false results when testing for absolute size difference, althoth use of the t test, without homogenious variance would be appropriate. Although the data would be suitable for direct calculation using t in order to determine significance of size groups, for the purposes of a more general hypothesis as to the selective oviposition of predators, an analysis on the basis of normalised size would also be useful, and would also facilitate a general mathematical model to be determined for Asteraceae, based upon normalised size, rather than for individual plants of differing sizes. In addition there is a trend within the data towards smaller flowers over the course of the season as seen if Figure 2.

Figure 2 Variation of absolute flower size with day of sampling

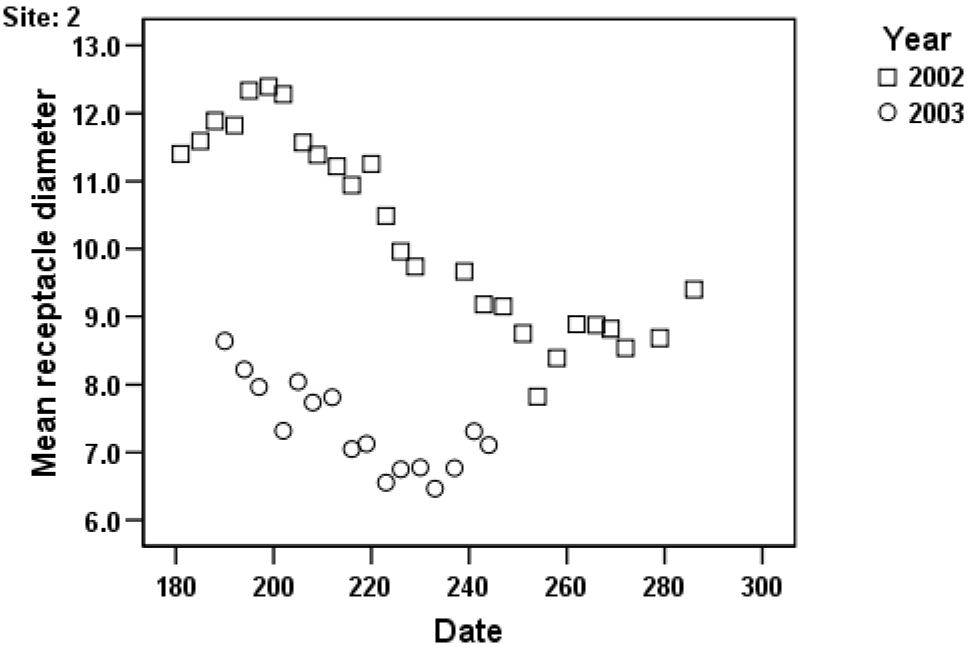
Cirsium palustre **(2001)**

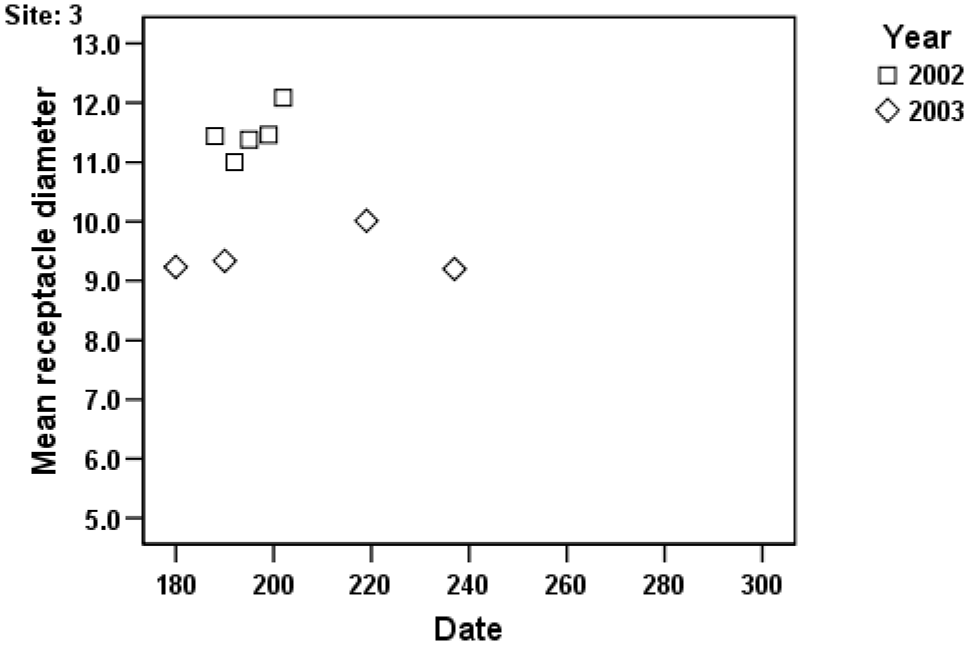


Tripleurospermum inodorum

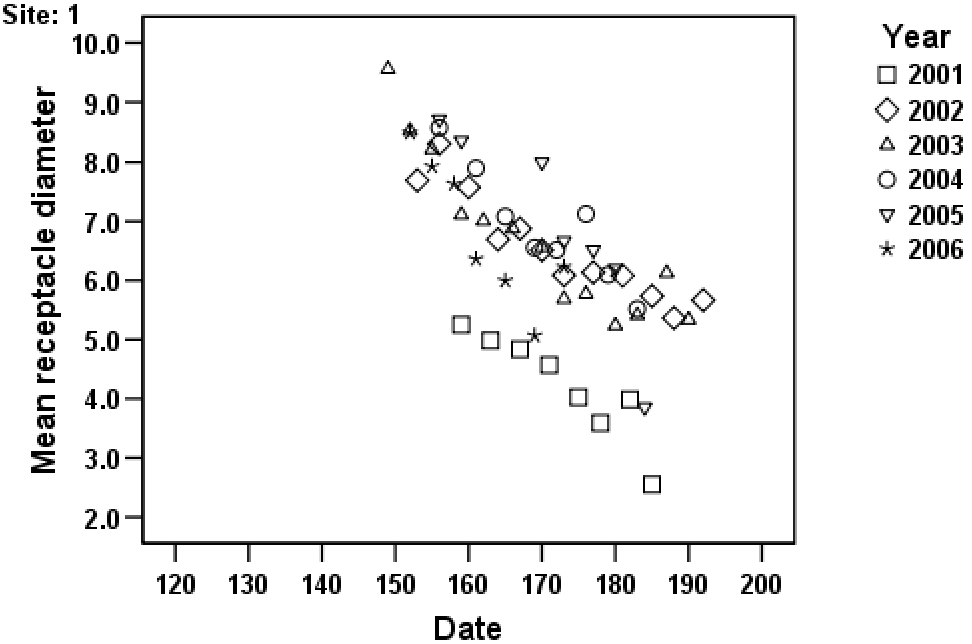


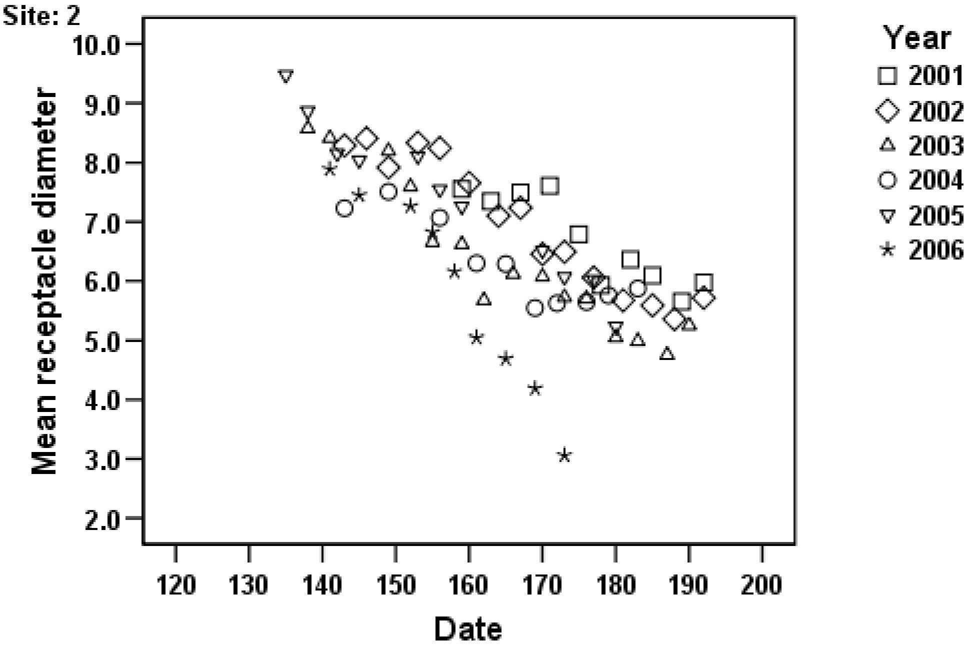
Tripleurospermum inodorum



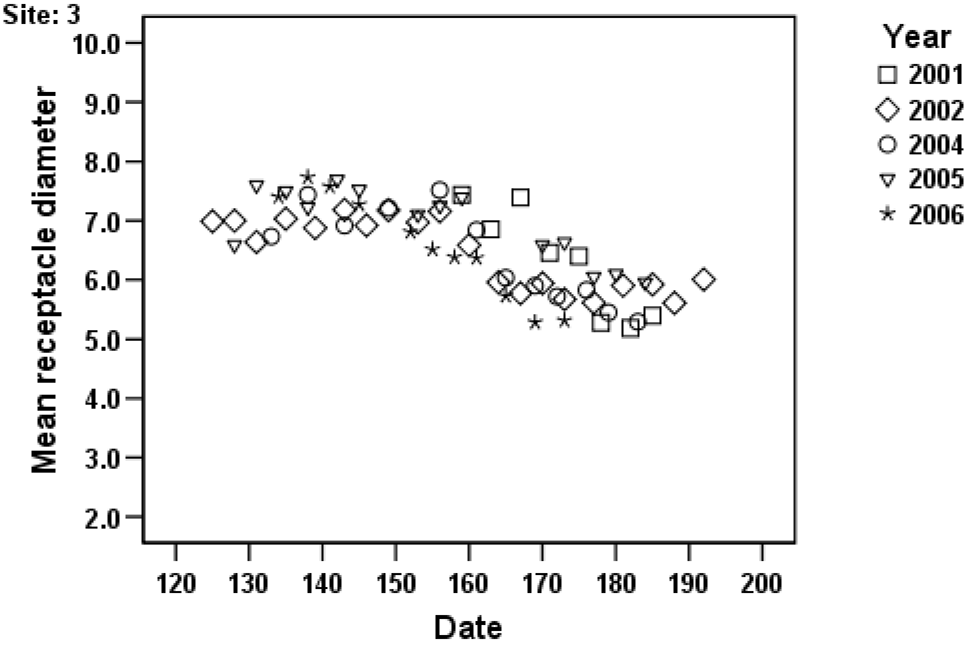


Leucanthemum vulgare

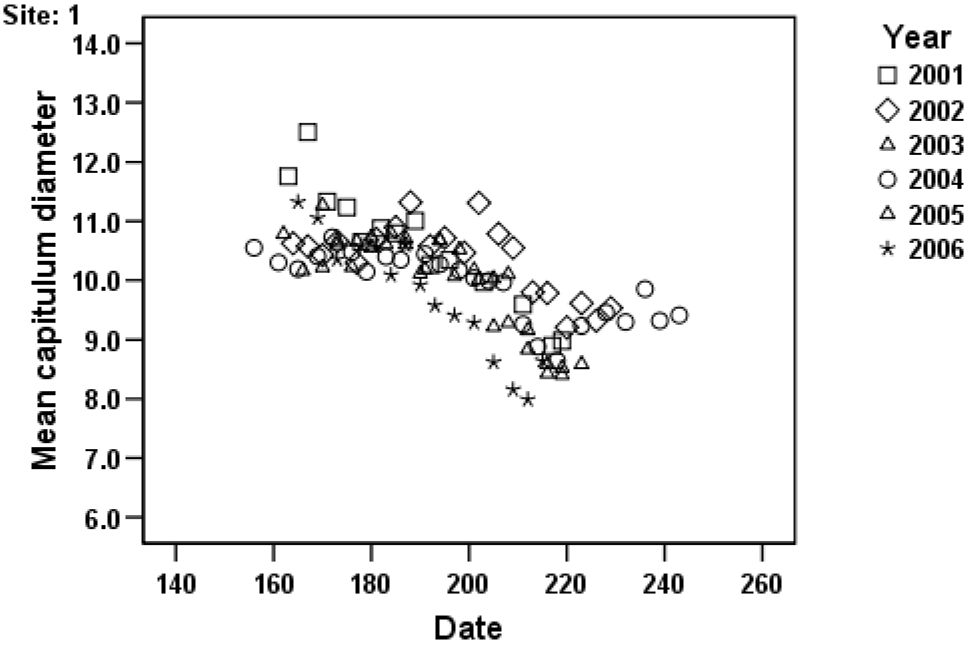


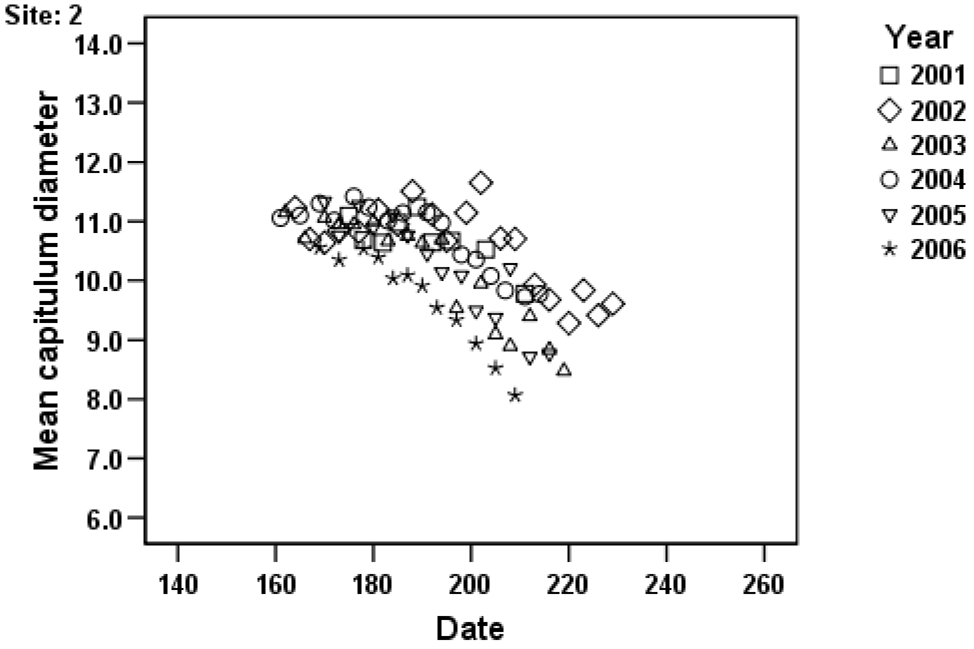


Leucanthemum vulgare

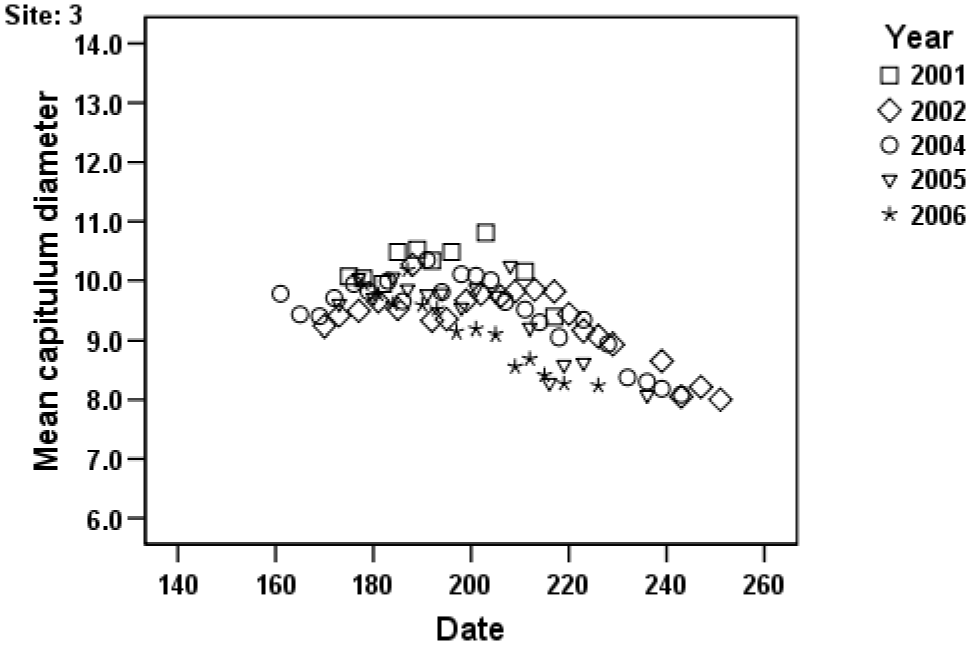


Centaurea nigra





Centaurea nigra



This tendency towards smaller flowers in late season is consistent over the various species under investigation, and appears to be independent of infestation.

In order to analyse these data for the possibility of predator selection on the basis of flower size, independent of the date of infestation, the data can be transformed simply by normalising on the basis of the mean capitulum size of the sampled heads on each day of sampling, provided that this does not affect the normality of the data. This has the further benefit that, in construction of a mathematical predation model, a predator choice by size function can be created with the single variable of relative flower size.

After this transposition, and ignoring flowers collected before correct identification of predation type, the data may be summarised as follows. Table 3 presents descriptive statistics on the normalised samples from each site and year, separated by infestation class. Data for *Leucanthemum vulgare* are only shown from 2003 onwards, and that for *Centaurea nigra* from 2002 onwards, to account for predator discrimination.

Table 3 Descriptive statistics by site and date

Cirsium palustre **(2001)**

Normalised capitulum size

Site		N	Mean	Std. Deviation
1	Uninfested	1440	.999	.104
	Infested	18	1.071	.107
	Total	1458	1.000	.104
2	Uninfested	1287	.999	.108
	Infested	23	1.034	.126
	Total	1310	1.000	.108
3	Uninfested	1012	.999	.102
	Infested	56	1.012	.090
	Total	1068	1.000	.101

Tripleurospermum inodorum

Normalised receptacle size

Site	Year		N	Mean	Std. Deviation
1	2002	Uninfested	1329	.978	.130
		Infested	1061	1.027	.120
		Total	2390	1.000	.128
	2003	Uninfested	1131	.995	.161
		Infested	274	1.019	.127
		Total	1405	1.000	.155
2	2002	Uninfested	1538	.977	.129
		Infested	1097	1.032	.131
		Total	2635	1.000	.132
	2003	Uninfested	831	.970	.166
		Infested	455	1.055	.161
		Total	1286	1.000	.169
3	2002	Uninfested	217	.974	.111
		Infested	257	1.022	.107
		Total	474	1.000	.111
	2003	Uninfested	601	.984	.123
		Infested	398	1.024	.122
		Total	999	1.000	.124

Leucanthemum vulgare

Descriptive Statistics

Normalised receptacle size

Site	Year	Infestation	N	Mean	Std. Deviation
1	2003	Uninfested	281	.941	.191
		Floret	608	1.027	.163
		Receptacle	5	.969	.251
		Both	6	1.011	.166
	2004	Uninfested	143	.964	.209
		Floret	139	1.032	.186
		Receptacle	29	.965	.173
		Both	35	1.048	.189
2	2003	Uninfested	577	.948	.195
		Floret	185	1.058	.208
		Receptacle	250	1.000	.211
		Both	115	1.167	.239
	2004	Uninfested	290	.943	.207
		Floret	63	1.036	.183
		Receptacle	179	1.028	.214
		Both	70	1.131	.191
3	2004	Uninfested	853	.981	.221
		Floret	75	1.032	.181
		Receptacle	144	1.079	.194
		Both	21	1.130	.189

Descriptive Statistics

Normalised receptacle size

Site	Year	Infestation	N	Mean	Std. Deviation
1	2005	Uninfested	216	.981	.178
		Floret	287	1.011	.149
		Receptacle	10	.982	.145
		Both	27	1.043	.163
	2006	Uninfested	211	.937	.211
		Floret	232	1.055	.170
		Receptacle	8	.966	.283
		Both	11	1.074	.222
2	2005	Uninfested	542	.985	.168
		Floret	31	1.036	.131
		Receptacle	108	1.041	.178
		Both	17	1.134	.162
	2006	Uninfested	426	.970	.217
		Floret	46	1.071	.186
		Receptacle	102	1.079	.240
		Both	9	1.170	.121
3	2005	Uninfested	962	.992	.195
		Floret	39	1.042	.152
		Receptacle	77	1.064	.195
		Both	8	1.190	.206
	2006	Uninfested	658	.987	.190
		Floret	50	1.076	.138
		Receptacle	45	1.075	.175
		Both	6	1.190	.133

Centaurea nigra

Descriptive Statistics

Normalised capitulum size

Site	Year	Infestation	N	Mean	Std. Deviation
1	2002	Uninfested	859	.990	.090
		Non-gall	724	1.004	.089
		Gall	84	1.068	.083
		Both	1	1.035	.
	2003	Uninfested	311	.979	.095
		Non-gall	897	.995	.100
		Gall	185	1.048	.094
		Both	34	1.068	.089
	2004	Uninfested	940	.984	.109
		Non-gall	609	1.001	.112
		Gall	245	1.054	.097
		Both	12	1.058	.075
2	2002	Uninfested	503	.985	.101
		Non-gall	456	.985	.097
		Gall	366	1.036	.094
		Both	25	1.047	.084
	2003	Uninfested	305	.976	.103
		Non-gall	740	.994	.098
		Gall	234	1.042	.094
		Both	47	1.038	.106
	2004	Uninfested	720	.983	.103
		Non-gall	427	.995	.106
		Gall	344	1.038	.104
		Both	12	1.077	.104
3	2002	Uninfested	622	.976	.097
		Non-gall	440	.980	.100
		Gall	545	1.044	.105
		Both	7	1.001	.180
	2004	Uninfested	1032	.979	.107
		Non-gall	512	.993	.104
		Gall	451	1.050	.117
		Both	44	1.074	.105

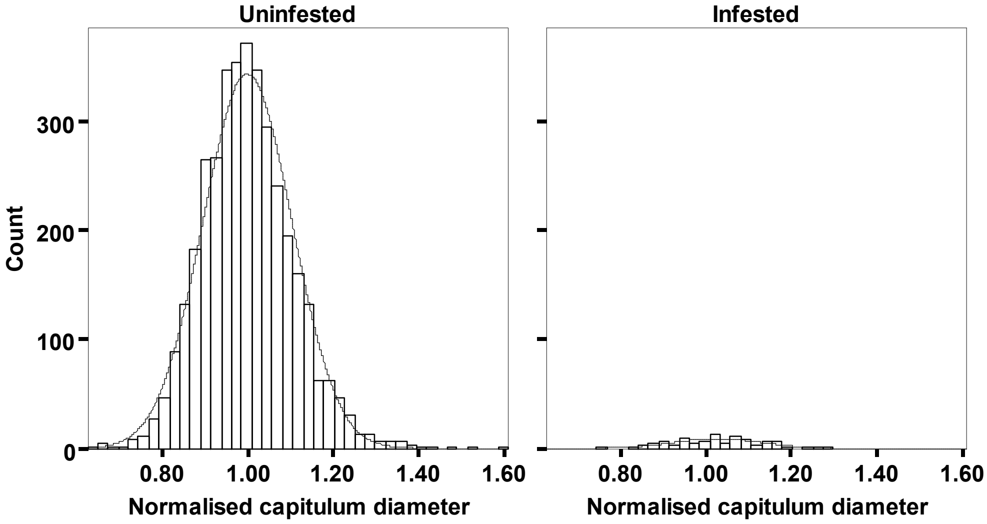
Descriptive Statistics

Normalised capitulum size

Site	Year	Infestation	N	Mean	Std. Deviation
1	2005	Uninfested	684	.988	.099
		Non-gall	389	.990	.095
		Gall	320	1.037	.084
		Both	15	1.020	.085
	2006	Uninfested	591	.995	.105
		Non-gall	418	.988	.097
		Gall	142	1.052	.086
		Both	9	1.068	.116
2	2005	Uninfested	577	.982	.104
		Non-gall	321	.988	.103
		Gall	307	1.045	.105
		Both	13	1.017	.133
	2006	Uninfested	481	.997	.107
		Non-gall	250	.987	.094
		Gall	203	1.022	.109
		Both	10	1.037	.096
3	2005	Uninfested	787	.978	.107
		Non-gall	373	.983	.107
		Gall	400	1.055	.105
		Both	18	1.082	.131
	2006	Uninfested	742	.987	.107
		Non-gall	252	.987	.106
		Gall	373	1.030	.100
		Both	32	1.045	.123

Figure 3 shows that the distribution of normalised sizes still maintains a normal distribution, and that there are unlikely to be sampling effects within the data.

Figure 3 Normalised size distribution of all sampled flower heads *Cirsium palustre* (2001)



One-Sample Kolmogorov-Smirnov Test

Infestation			Normalised capitulum diameter
Uninfested	N		3739
	Normal Parameters [a,b]	Mean	.999
		Std. Deviation	.105
	Kolmogorov-Smirnov Z		2.135
	Exact Sig. (2-tailed)		1.000
Infested	N		97
	Normal Parameters [a,b]	Mean	1.028
		Std. Deviation	.104
	Kolmogorov-Smirnov Z		.440
	Exact Sig. (2-tailed)		.985

[a] Test distribution is Normal.

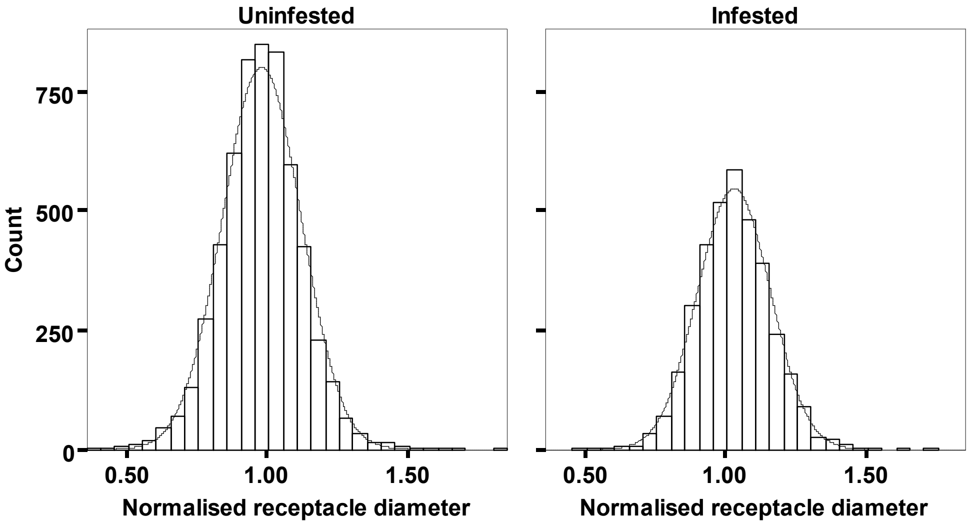
[b] Calculated from data.

Test of Homogeneity of Variances

Normalised capitulum diameter

Levene Statistic	df1	df2	Sig.
.042	1	3834	.838

Tripleurospermum inodorum (2002-2003)



One-Sample Kolmogorov-Smirnov Test

Infestation			Normalised receptacle diameter
Uninfested	N		5647
	Normal Parameters ^{a,b}	Mean	.981
		Std. Deviation	.141
	Kolmogorov-Smirnov Z		1.745
	Exact Sig. (2-tailed)		1.000
Infested	N		3542
	Normal Parameters ^{a,b}	Mean	1.031
		Std. Deviation	.130
	Kolmogorov-Smirnov Z		1.245
	Exact Sig. (2-tailed)		1.000

a. Test distribution is Normal.

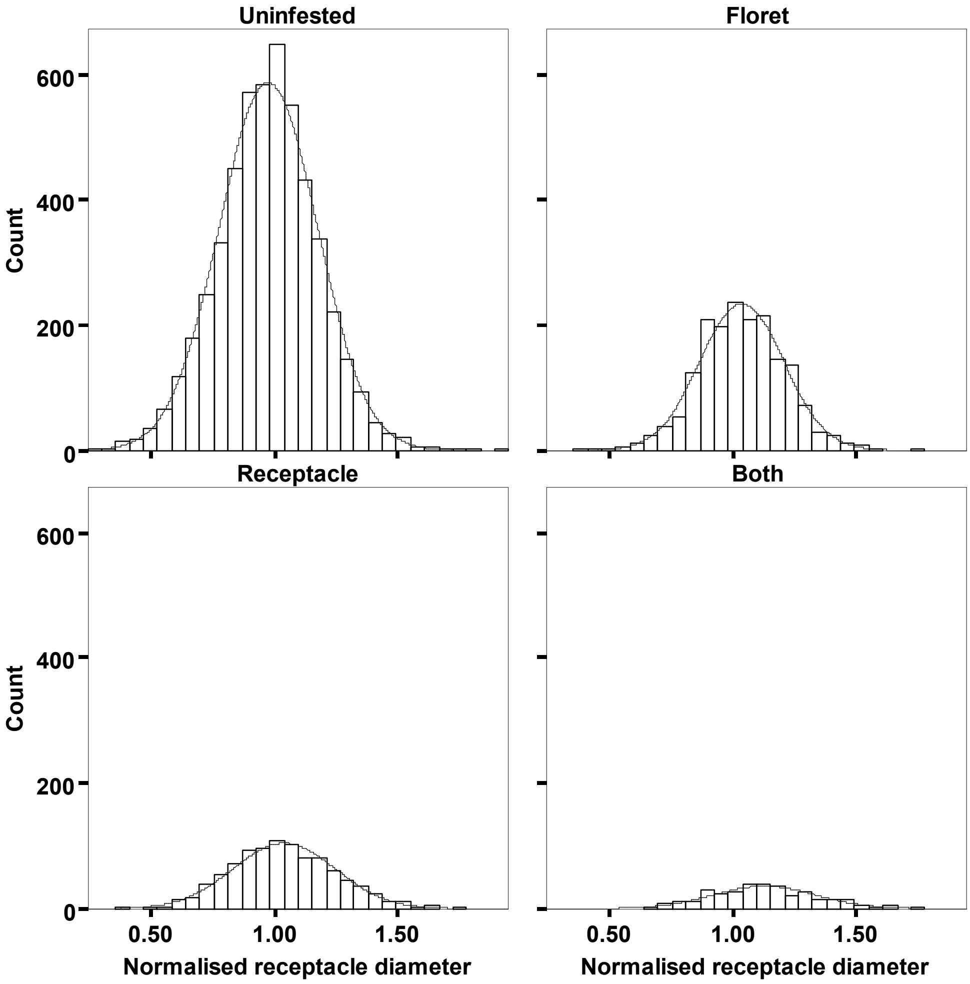
b. Calculated from data.

Test of Homogeneity of Variances

Normalised receptacle diameter

Levene Statistic	df1	df2	Sig.
13.393	1	9187	<0.001

Leucanthemum vulgare (2003-2006)



One-Sample Kolmogorov-Smirnov Test

Infestation			Normalised receptacle diameter
Uninfested	N		5159
	Normal Parameters [a,b]	Mean	.973
		Std. Deviation	.200
	Kolmogorov-Smirnov Z		1.226
	Exact Sig. (2-tailed)		1.000
Floret	N		1755
	Normal Parameters [a,b]	Mean	1.035
		Std. Deviation	.170
	Kolmogorov-Smirnov Z		.773
	Exact Sig. (2-tailed)		1.000
Receptacle	N		957
	Normal Parameters [a,b]	Mean	1.037
		Std. Deviation	.207
	Kolmogorov-Smirnov Z		.802
	Exact Sig. (2-tailed)		1.000
Both	N		325
	Normal Parameters [a,b]	Mean	1.127
		Std. Deviation	.208
	Kolmogorov-Smirnov Z		.835
	Exact Sig. (2-tailed)		.475

[a] Test distribution is Normal.

[b] Calculated from data.

Test of Homogeneity of Variances - floret only

Normalised receptacle diameter

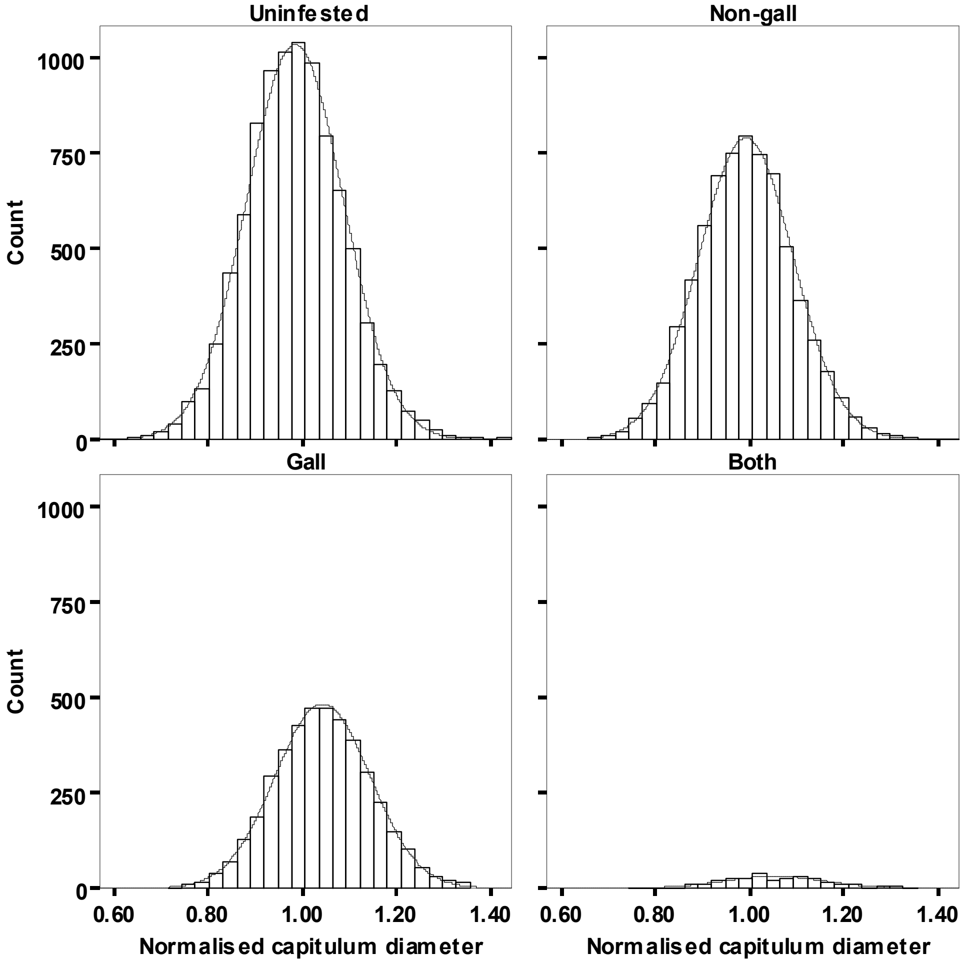
Levene Statistic	df1	df2	Sig.
42.840	1	6912	<0.001

Test of Homogeneity of Variances - receptacle only

Normalised receptacle diameter

Levene Statistic	df1	df2	Sig.
4.294	1	6114	.038

Centaurea nigra (2002-2006)



One-Sample Kolmogorov-Smirnov Test

Infestation			Normalised capitulum diameter
Uninfested	N		9154
	Normal Parameters [a,b]	Mean	.984
		Std. Deviation	.103
	Kolmogorov-Smirnov Z		1.362
	Exact Sig. (2-tailed)		1.000
Non-gall	N		6808
	Normal Parameters [a,b]	Mean	.992
		Std. Deviation	.100
	Kolmogorov-Smirnov Z		1.062
	Exact Sig. (2-tailed)		1.000
Gall	N		4198
	Normal Parameters [a,b]	Mean	1.043
		Std. Deviation	.101
	Kolmogorov-Smirnov Z		.866
	Exact Sig. (2-tailed)		1.000
Both	N		279
	Normal Parameters [a,b]	Mean	1.052
		Std. Deviation	.107
	Kolmogorov-Smirnov Z		.918
	Exact Sig. (2-tailed)		.355

[a] Test distribution is Normal.

[b] Calculated from data.

Test of Homogeneity of Variances - non gall only

Normalised capitulum diameter

Levene Statistic	df1	df2	Sig.
3.031	1	15960	.082

Test of Homogeneity of Variances - gall only

Normalised capitulum diameter

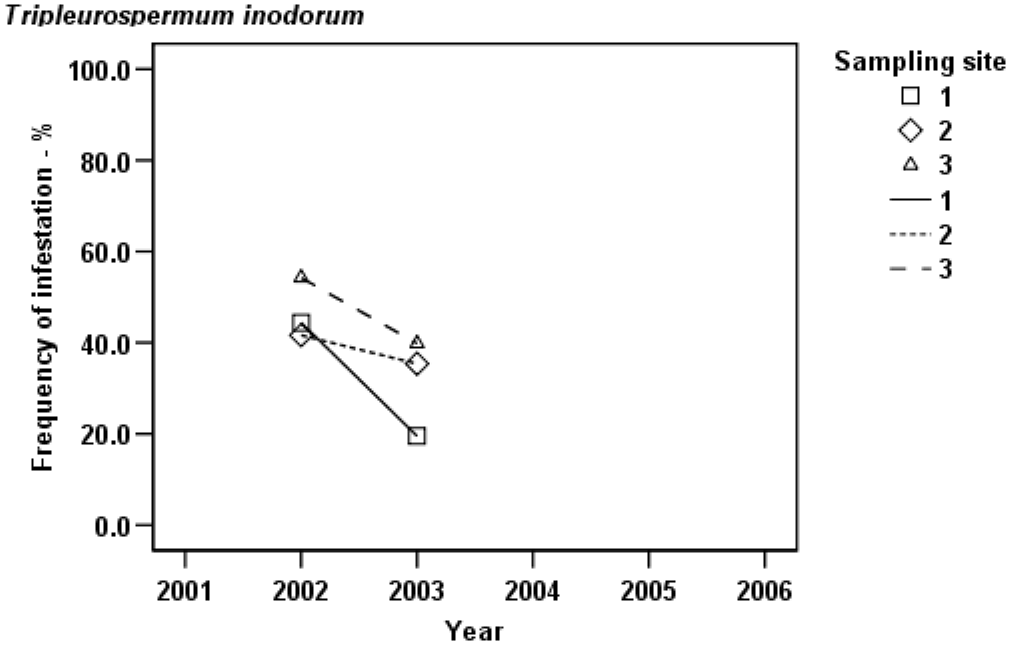
Levene Statistic	df1	df2	Sig.
.081	1	13350	.776

As with the full size data, the groups, other than the small subsets of dual infestation, exhibit a good fit with the normal distribution, but generally fail the homogeneity of variance. Analysis should therefore be carried out using the t test without assuming homogenious variance.

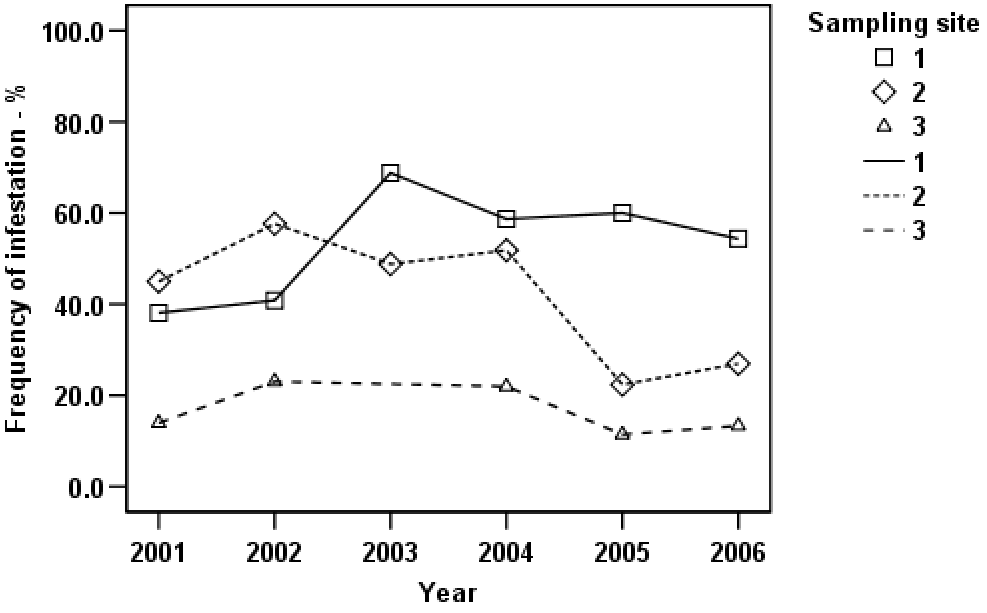
3.3 Patterns of infestation

The levels of overall seasonal infestation rates are shown as total infestation in Figure 4, and in Figure 5 determined by individual predator species. Although there is some annual variation, perhaps due to weather conditions during the season, or pupal survival rates over the previous winter, the general levels of infestation are relatively stable.

Figure 4 Variation of total infestation with year



Leucanthemum vulgare



Centaurea nigra

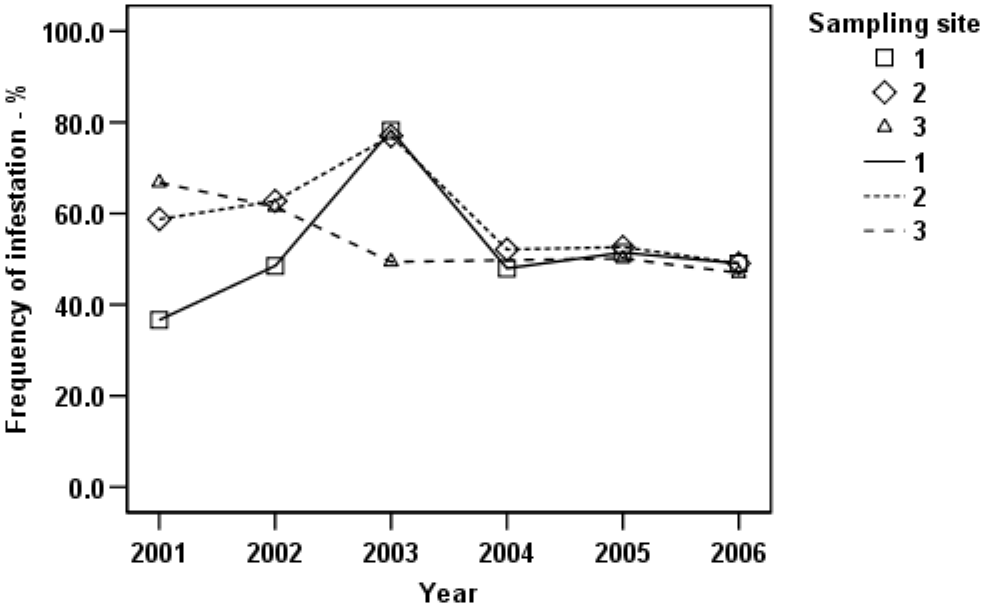
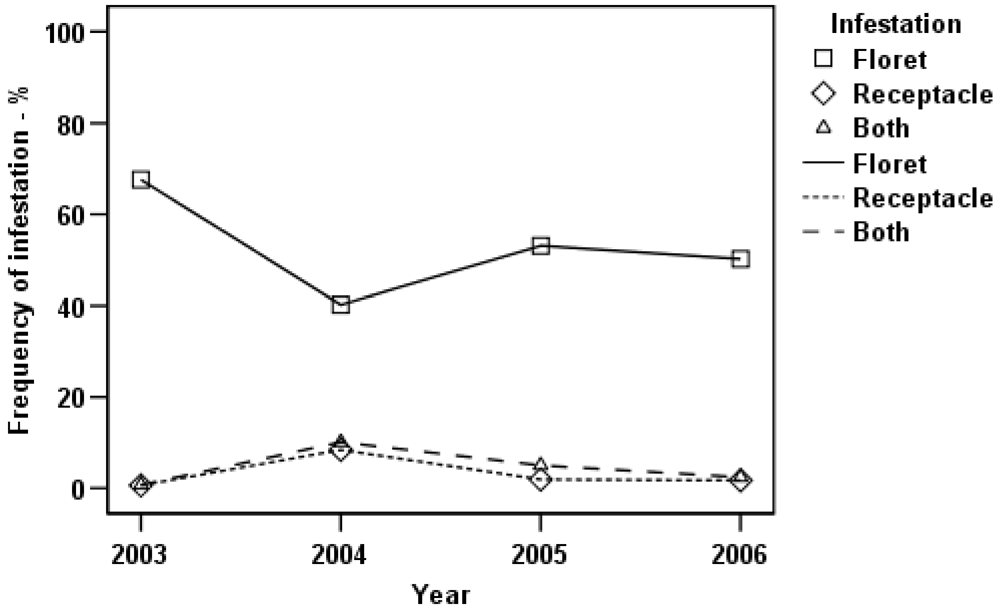


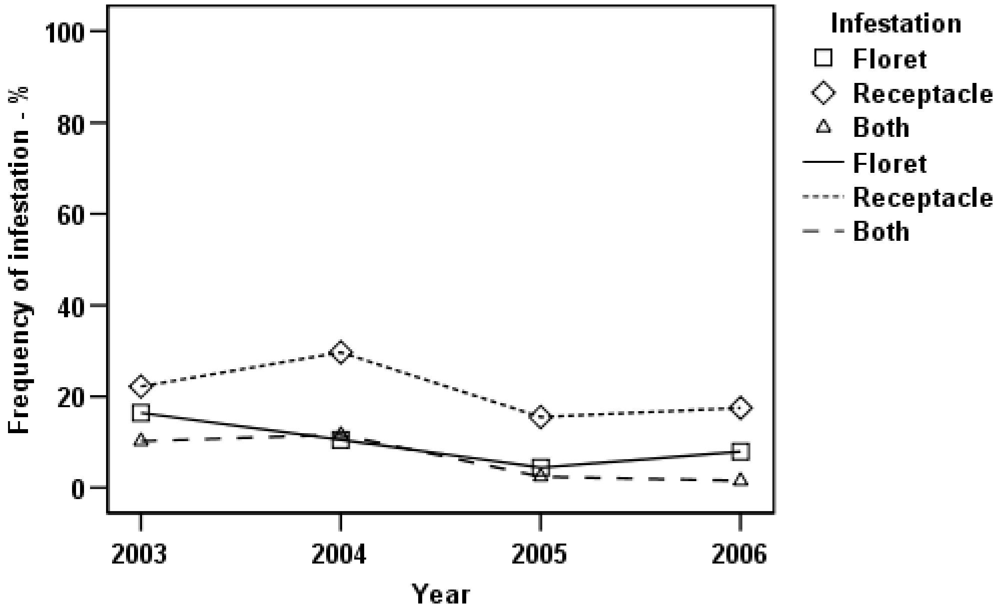
Figure 5 Variation of individual infestation frequency with year

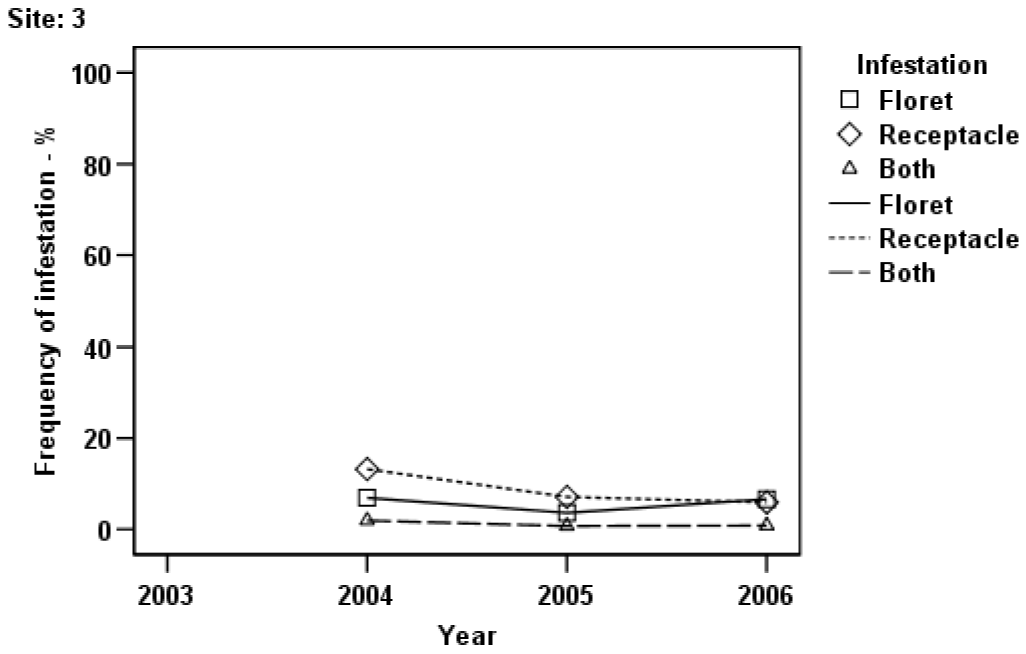
Leucanthemum vulgare

Site: 1

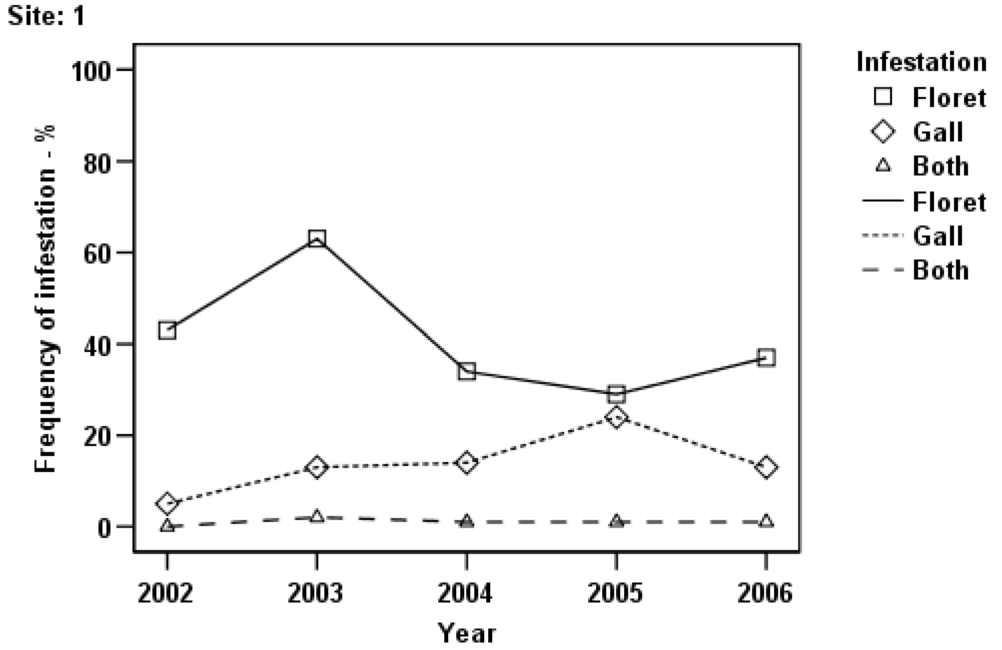


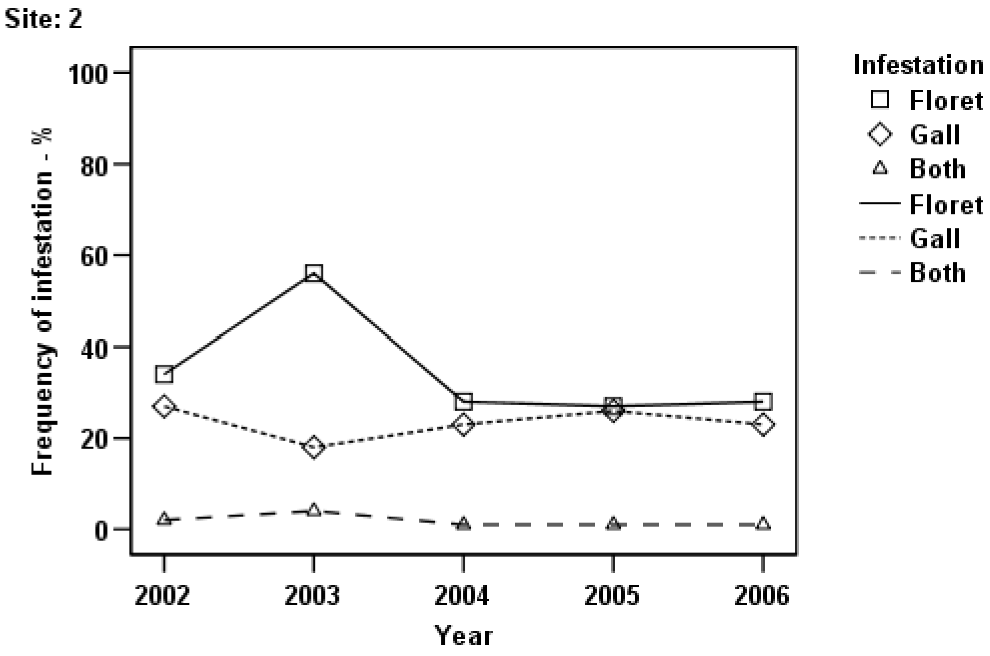
Site: 2

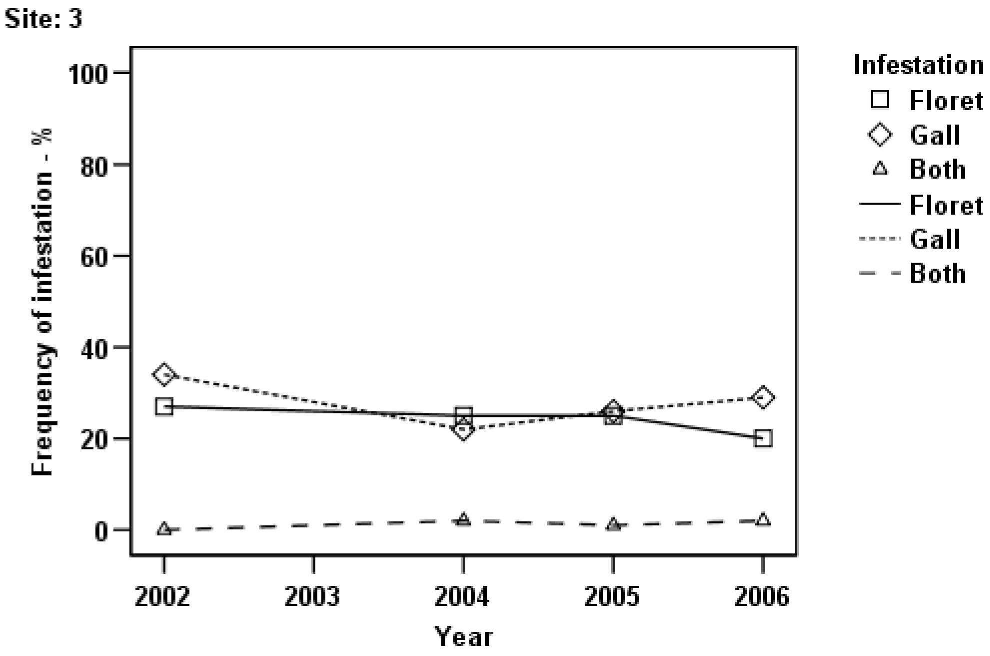




Centaurea nigra







3.4 Seed set

In 2003, 2004 and 2005, drying heads from each of the three species sampled were taken from random positions within the plots. In 2003 these were stored in a freezer, in 2004 and 2005 the heads were dried fully prior to observation.

In order to determine any effect on fecundity of predation, a means of determining the number of viable seeds in the heads is required. Viability of seed was taken to be indicated by the presence of an endosperm within the testa. An initial investigation of the seeds was carried out by means of dissection. This investigation identified four main characteristics that separated potentially viable seeds, with endosperm, from non-viable seeds, with no endosperm: integrity of testa, seed size, level of swelling and and colour. These four characteristics were common to seeds of both *Leucanthemum vulgare* and *Centaurea nigra*, and thus became the basis of a visual means of separating viable and non-viable seeds within a flower head.

The frozen or dried samples were removed and inspected for seed set. Each head was measured for its characteristic size, and cut open to reveal the presence of infestation, and the seeds. Seeds were removed from the head and, using visual inspection under magnification, with occasional confirmation by dissection, were separated into viable and non-viable categories. Finally the number of seeds in each category were counted and recorded for later analysis, together with the flower head size and type of infestation, where present.

The number of heads sampled, and totals for seeds sorted and counted are shown in Table 4.

Table 4 Seeds sampled

	Heads	Seeds	Seeds set	% Seeds set	% Infestation
T. inodorum	45	12174	10170	81%	16%
L. vulgare	259	67967	35161	52%	62%
C. nigra	449	17620	9005	51%	72%

3.5 Infesting species

In 2002, heads of infested flowers from each species were stored in aerated plastic bags until the infesting larvae hatched into adult flies. The adults were stored in marked plastic containers and sent for identification.

In both 2003 and 2004 selected heads of both *Leucanthemum vulgare* and *Centaurea nigra* were stored in plastic containers and the infested larvae hatched. Identification was undertaken of the species with free living larvae in both *Leucanthemum vulgare* and *Centaurea nigra* and the gall forming species in *Centaurea nigra* by microscopic inspection of the adults, using an identification key, White (1988).

Identification revealed that *Leucanthemum vulgare* florets were infested by *Tephritis neesii*, which White identifies as the only Tephritid predating the capitulum of *L. vulgare*, which is its principal host plant in the UK. *T. neesii* has a single generation per year, pupating within the capitulum and over wintering as an adult. The receptacle based predation appeared to be caused by a midge, although only a single specimen, unidentified species, was observed.

Centaurea nigra produced *Chaetostomella cylindrica* (free-living larvae), which White describes as a polyphagous predator, also infesting *Cirsium arvens, Cirsium palustre, Serratula tinctora* and *Centaurea montana*. *C. cylindrica* is variable in the number of generations per year, sometimes producing a second, which over winters within the dried heads of its host plant as a larva, emerging in May or June.

Two species of the genus *Urophora*, *U. jaceana* and *U. quadrifasciata* (gall forming), were also identified from *Centaurea nigra* heads. Both are specialist predators of *C. nigra*, *U. jaceana* producing a single generation per year, and *U. quadrifasciata* producing two, each over winters as a larva within the dried flower heads.

Freese and Gunther (1991) identify *Napomyza lateralis* (Agromyzidae) as a predator on *Tripleurospermum perforatum*, and partial identification of a hatched adult from *Tripleurospermum inodorum* was made as an agromyzid.

Figure 6 **Infesting species**

Tephritis neesii *Chaetostomella cylindrica*

Urophora jaceana *Urophora quadrifasciata*

3.6 Flowering phenology

Overall population phenology was recorded at selected sites by recording, at each visit throughout the season, the number of flowers suitable for collection within a marked portion of the sites. At the start of each season, a representative portion of the site was marked by bamboo canes. On each visit, before data collection began, these areas were thoroughly searched to determine the number of open flowers within them. These count records were translated into density of flower heads per square metre present on each collection date.

In 2002, an initial investigation into the average individual flowering phenology of each of the three sampled species was carried out at the sites where density of flowering was recorded. This study was undertaken at mid-season. Three flower heads were marked with looped labels at the point where florets were visible but unopened. At each subsequent visit the status of floret opening and floret drying was noted. For *L. vulgare* and *C. nigra* this process was undertaken twice, for *T. inodorum* only one set of heads was recorded.

3.7 Effect of plant density on infestation level

In 2003, five cleared plots, separated by gaps of 25 metres, in the Chilworth walled garden were planted with 20 *Tripleurospermum inodorum* seedlings, grown from seed in a greenhouse. Planting was made at a variety of seedling densities. The plots were sampled at five points during their growing season, and similar collection and inspection techniques used to determine the rates of infestation. Graphs of the rate of infestation against plant density were prepared for each of the five sampling visits.

Centaurea nigra plants, collected from naturally growing populations in Chilworth at the rosette stage of seasonal development, were also set out in prepared plots separated by 100m in the Chilworth meadow. Unfortunately these plots suffered heavy herbivory from rabbits. In 2005 therefore, a survey of the entire meadow allowed separate areas, suitably dispersed over the site, to be identified with a wide variety of plant densities, and the experiment repeated.

Leucanthemum vulgare sampling on the basis of local pant density was carried out in 2006, following a survey of the Chilworth meadow that identified suitable sampling points.

The results of the density experiments are summarised in terms of frequency of infestation in Table 5 and Figure 7.

Table 5 Frequencies of infestation at collection date

Tripleurospermum inodorum

Day of year	Minimum	Maximum	Mean	Std. Deviation
180	.00	.28	.0720	.11713
190	.22	.48	.3760	.11950
219	.72	.78	.7520	.02683
237	.35	.44	.3940	.03286

Centaurea nigra

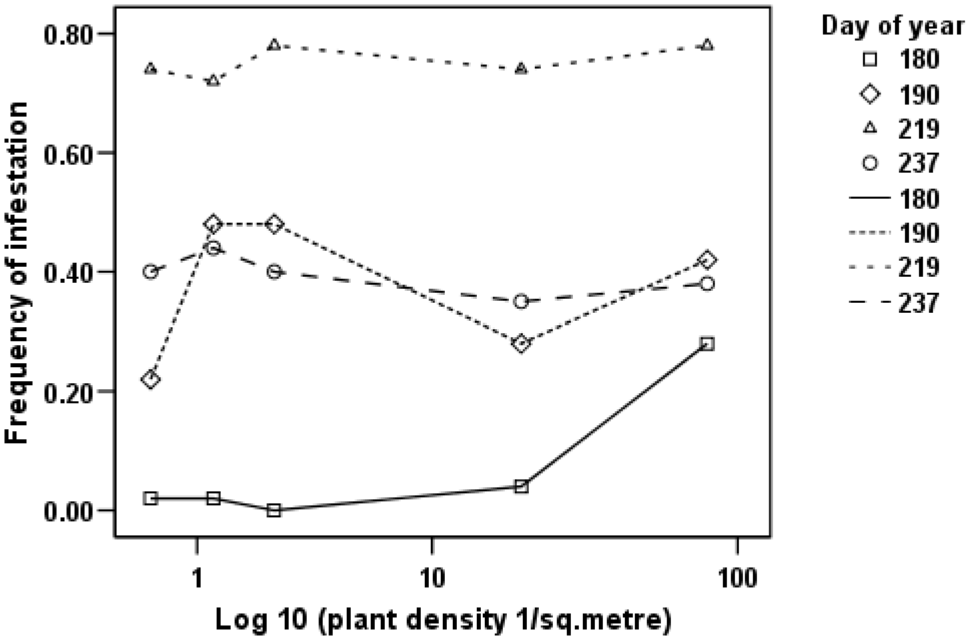
Day of year	Infestation	Minimum	Maximum	Mean	Std. Deviation
191	Floret	.10	.26	.1684	.06366
	Gall	.14	.45	.3340	.12116
208	Floret	.30	.46	.3540	.06229
	Gall	.26	.56	.3760	.12522

Leucanthemum vulgare

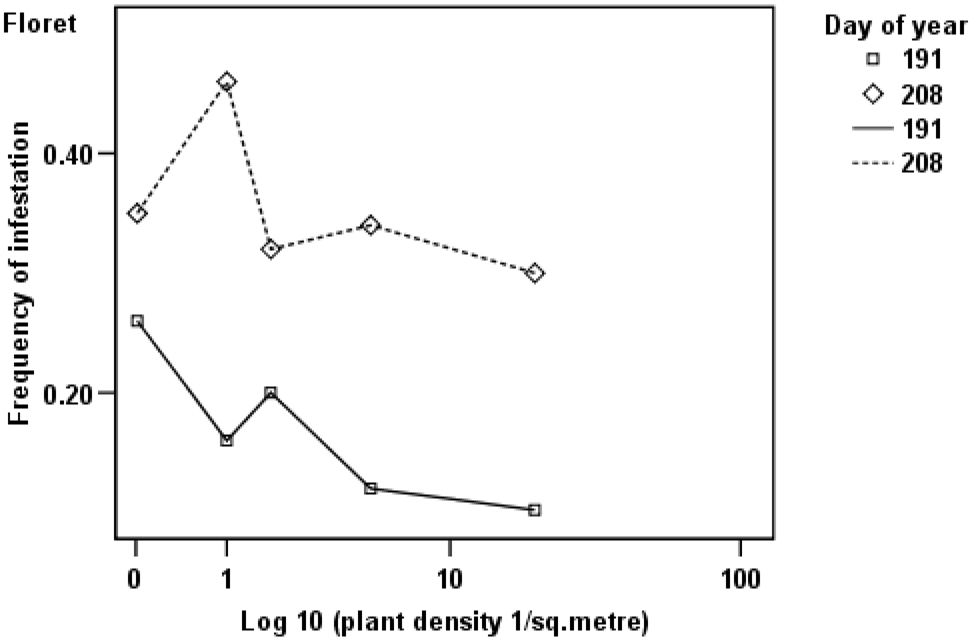
Day of year	Minimum	Maximum	Mean	Std. Deviation
155	.50	.74	.6600	.13856
161	.54	.80	.6733	.13013
169	.44	.78	.5600	.19079

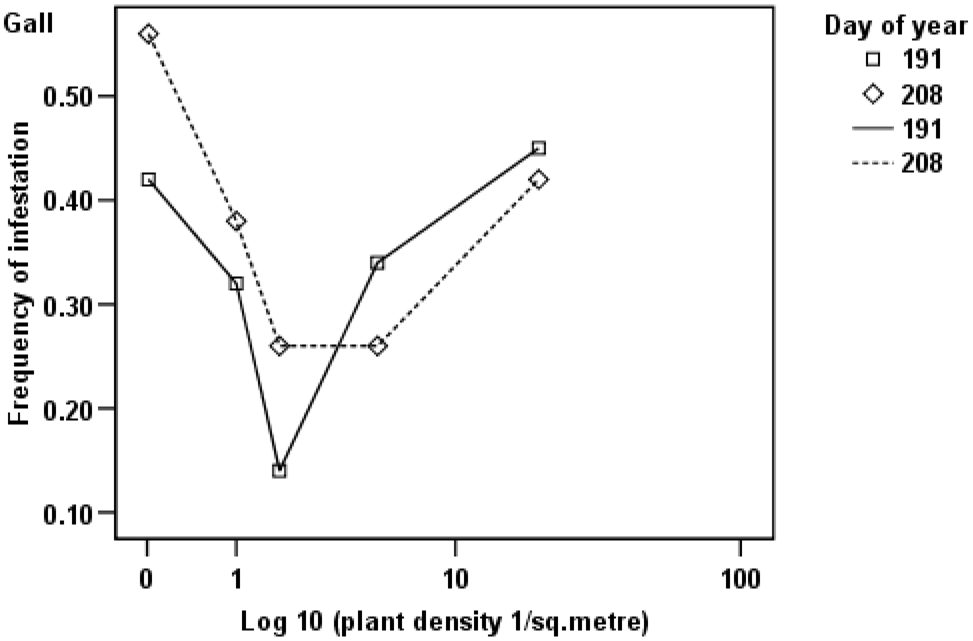
Figure 7 Density experiments

Tripleurospermum inodorum (2003)



Centaurea nigra **(2005)**





Leucanthemum vulgare **(2006)**

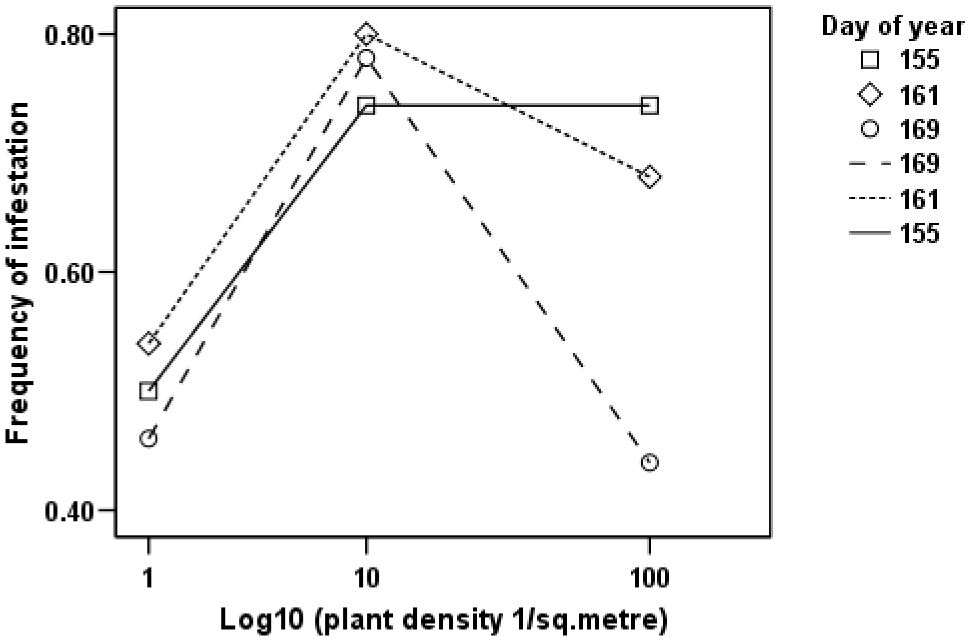


Table 6 shows the R-square values of a linear regression of frequency of infestation against plant density, and the partial correlations between frequency of infestation and density, controlling for day of the year.

Table 6 Regression statistics for density experiments

Tripleurospermum inodorum

Day of year	R Square
180	.973
190	.014
219	.307
237	.185

Correlations

Control Variables: Day of year

		Frequency of infestation	Density
Correlation	Frequency of infestation	1.000	.141
	Density	.141	1.000
Significance (2-tailed)	Frequency of infestation	.	.564
	Density	.564	.

Leucanthemum vulgare

Day of year	R Square
155	.324
161	.016
169	.225

Correlations

Control Variables: Day of year

		Density	Frequency of infestation
Correlation	Density	1.000	.010
	Frequency of infestation	.010	1.000
Significance (2-tailed)	Density	.	.981
	Frequency of infestation	.981	.

Centaurea nigra

Day of year	Infestation	R Square
191	Floret	.519
	Gall	.245
208	Floret	.287
	Gall	.001

Correlations

Control Variables: Day of year

Infestation			Density	Frequency of infestation
Floret	Correlation	Density	1.000	-.629
		Frequency of infestation	-.629	1.000
	Significance (2-tailed)	Density	.	.070
		Frequency of infestation	.070	.
Gall	Correlation	Density	1.000	.255
		Frequency of infestation	.255	1.000
	Significance (2-tailed)	Density	.	.507
		Frequency of infestation	.507	.

The data suggest some correlation for *T. inodorum* on the first date but little thereafter. For *C. nigra*, predated by *C. cylindrica*, frequency appears to reduce somewhat as density increases, whereas under infestation from *U. quadrifasciata* and *L. vulgare* infested with *T. neesii* there is little evidence of a correlation of infestation with local density, at the scale of patches distributed over 1 – 3 hectares.

The general lack of correlation between infestation rates and local plant density allows modelling to proceed without the need to include density effects. Clearly the populations sampled exist as part of a much larger meta-population in the local region. All of the plant species are common within S. Hampshire Brewis et al. (1996), and it is likely that the density plots would lie within the foraging area of the insect species involved, but clearly localised density is not a variable that significantly affects the probability of infestation. Halley & Dempster (1996) provide evidence of host plant density dependence over wider areas for populations of Tephritids, but this can be masked by immigration between neighbouring patches in the meta-population.

Chapter 4. Oviposition choice by flower size

The first hypothesis defined in order to determine the role of pre-dispersal seed predation in floral evolution is that:

> Pre-dispersal seed predators tend to select larger flower heads as oviposition sites in a repeatable pattern over time and population.

As described in Chapter 3, sampling of species over a six year period yielded a large amount of data on patterns of infestation. The initial description of these data indicate that both infested and uninfested samples appear to fall into normal distributions based upon a representative absolute or relative flower size, and that the mean sizes of these samples differ when grouped by predator selection for oviposition.

In order to determine the validity of the hypothesis it must be shown firstly that there is statistical significance in size difference in grouping samples by predator choice, and secondly that there is some pattern to the choice.

4.1 Size difference

Data from the sampling were entered into a Microsoft Excel spreadsheet. For each sampling date, the following data were calculated:

- mean flower size
- standard deviation of measured flower size
- minimum flower size
- maximum flower size
- number of infested flowers, as a percentage of the total, separated by infestation type where appropriate

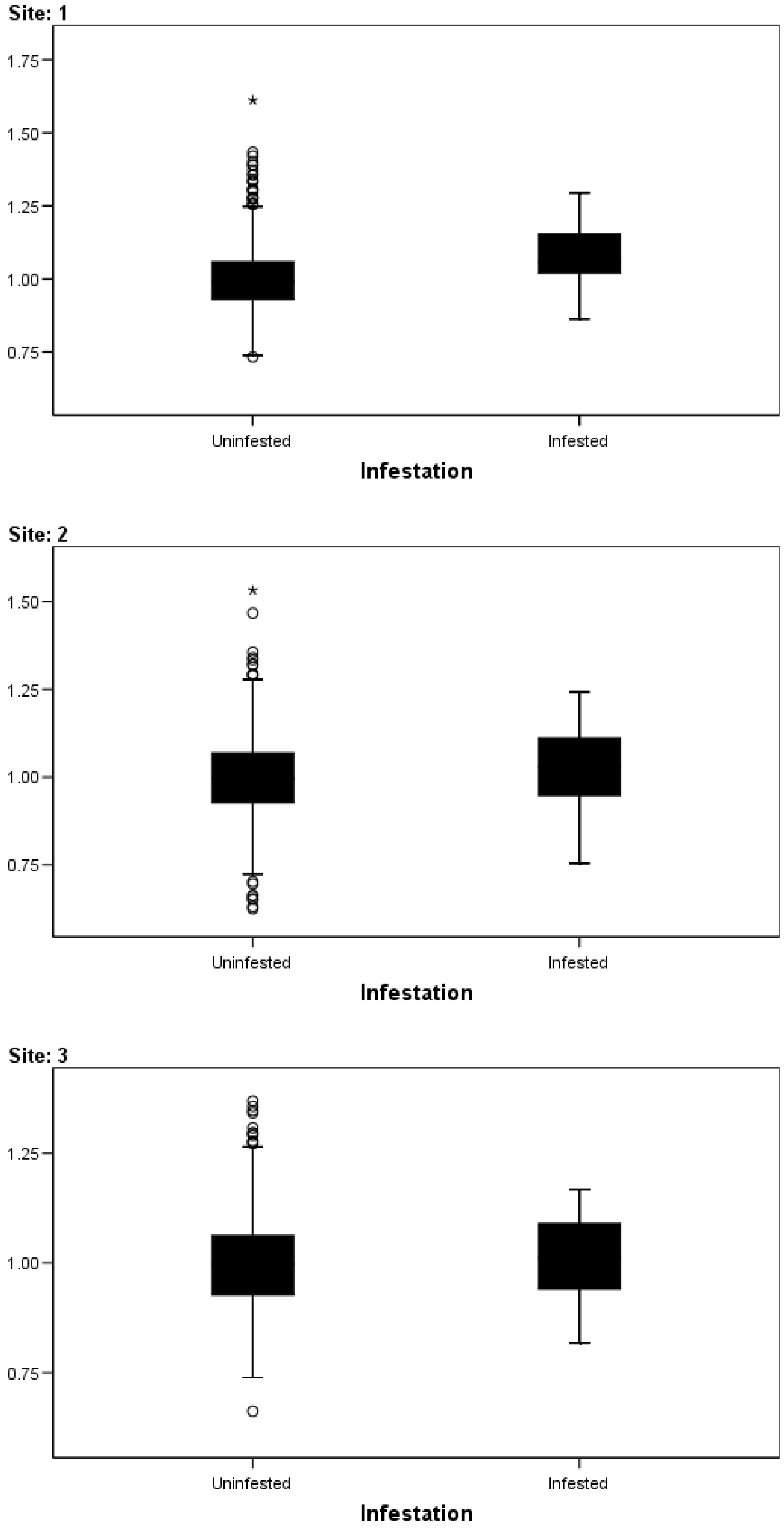
At the end of each season the following values were calculated at each site:

- the overall level of infestation
- mean normalised flower size for each of the total of uninfested and infested flower heads
- the significance of the difference between the mean normalised infested and uninfested values, using the t test.

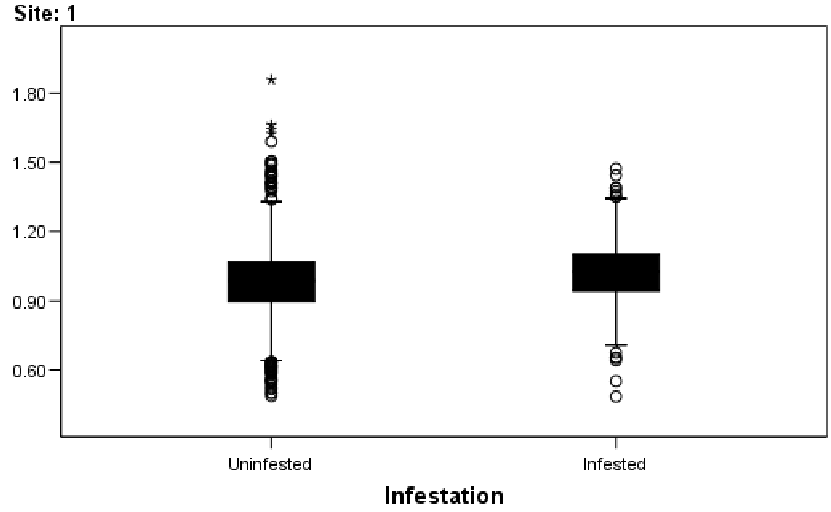
Figure 8 shows the relationships between the mean sizes of uninfested and infested flowers.

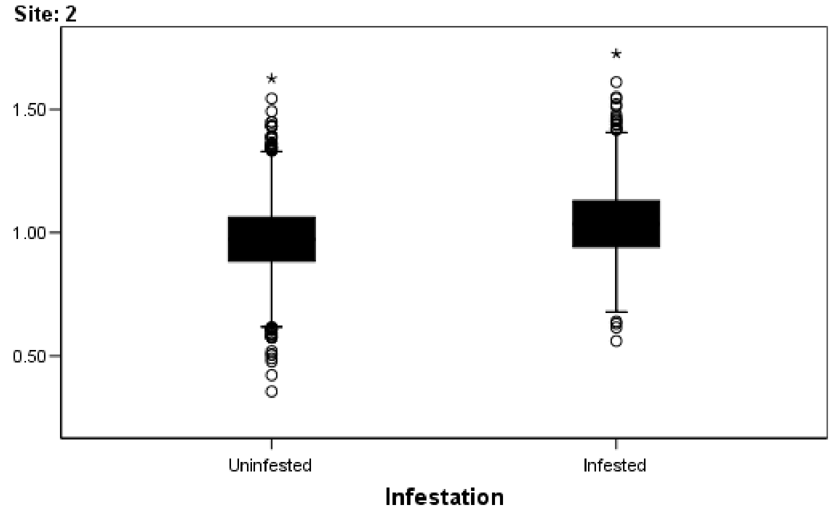
Figure 8 Boxplots of normalised size with infestation type

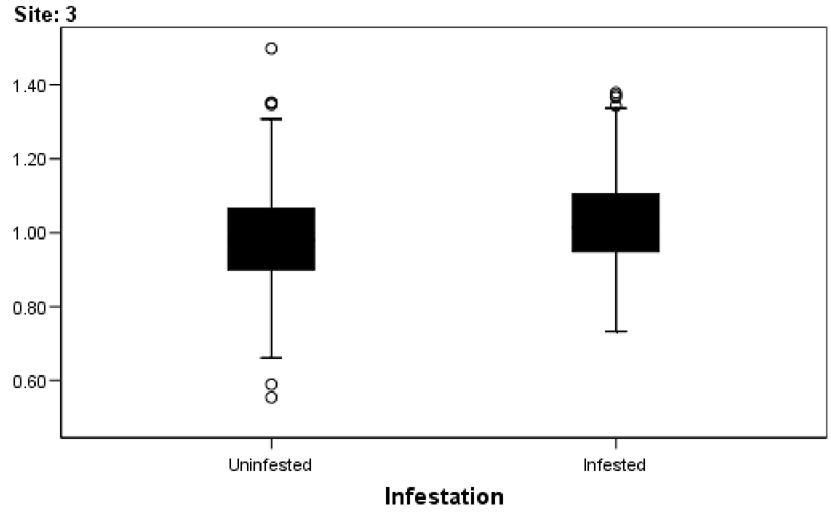
Cirsium palustre



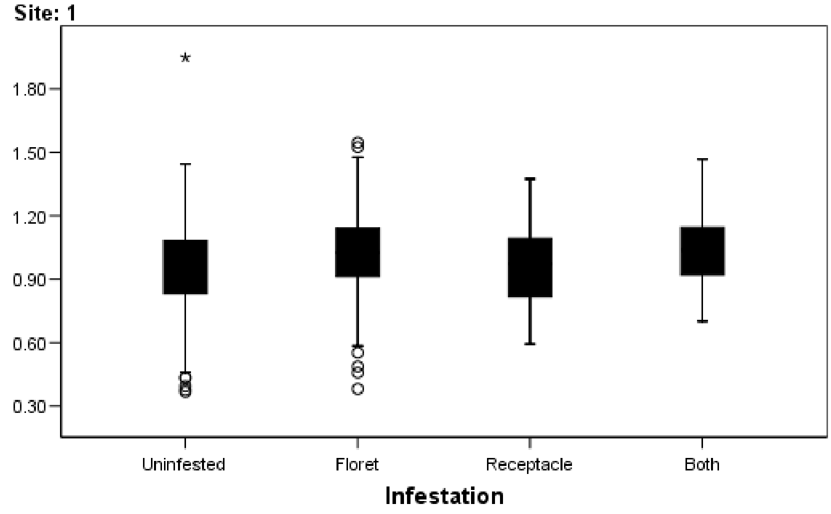
Tripeurospermum inodorum

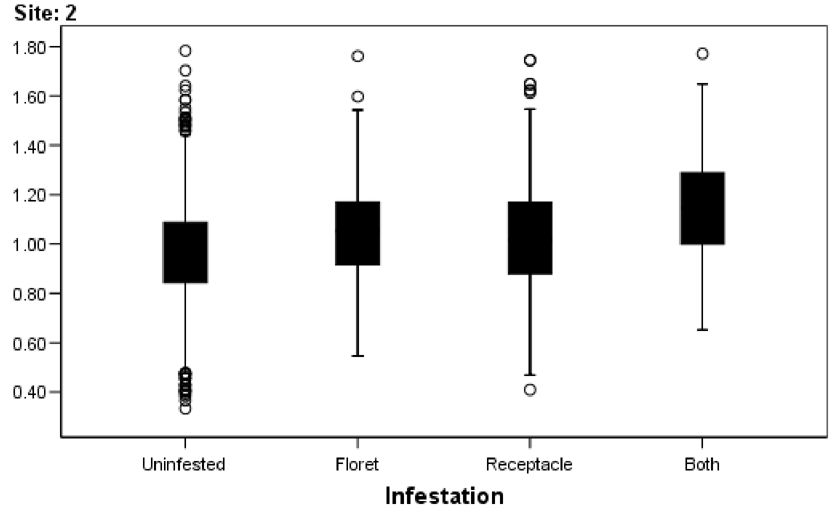


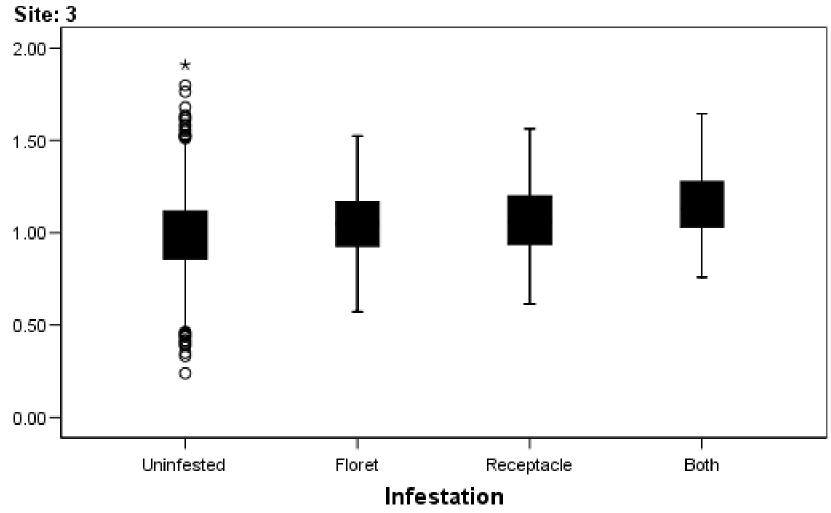




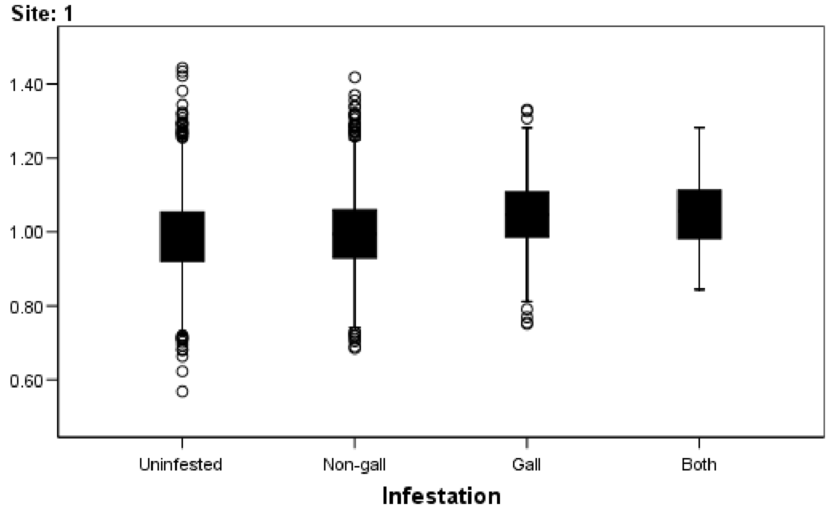
Leucanthemum vulgare

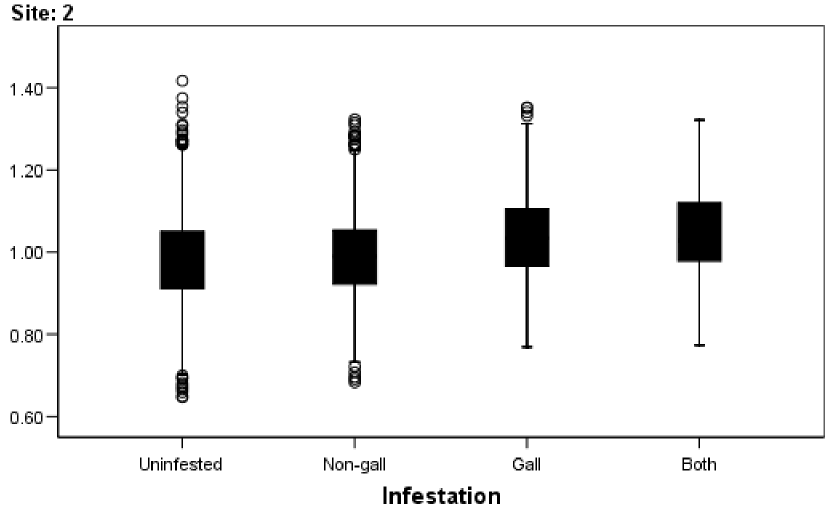


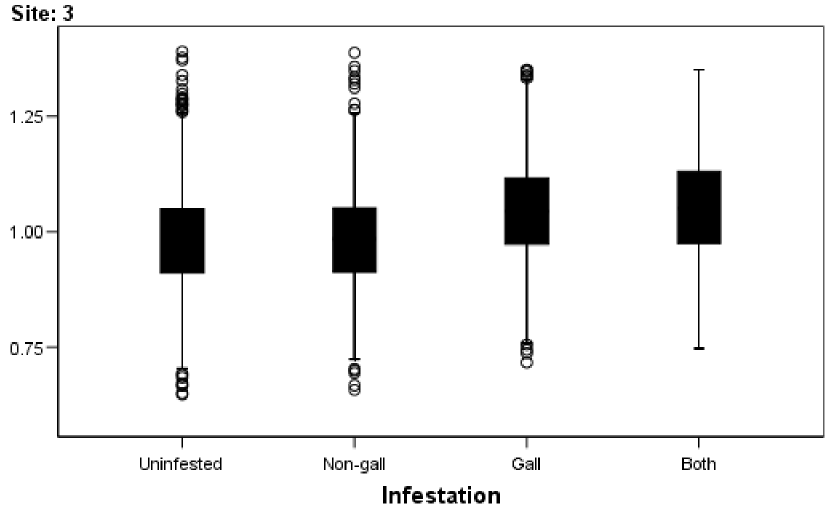




Centaurea nigra







Using the SPSS analysis package, these data were aggregated into a single data file covering all of the samples. T test analysis was carried out to determine levels of significance of the difference in mean size of infested and uninfested flower heads.

Table 7 presents the basic statistics for all of the sampled flowers considered in the analysis, and Table 8 the results of t testing on the mean size difference between each infestation type and the uninfested heads for all sampled flowers.

Table 7 Descriptive statistics of all sampled flowers

Cirsium palustre

Normalised capitulum diameter

Infestation	N	Mean	Std. Deviation	Skewness
Uninfested	3739	.999	.105	.425
Infested	97	1.028	.104	.063
Total	3836	1.000	.105	.415

Tripleurospermum inodorum

Normalised receptacle diameter

Infestation	N	Mean	Std. Deviation	Skewness
Uninfested	5647	.981	.141	.142
Infested	3542	1.031	.130	.241
Total	9189	1.000	.139	.128

Leucanthemum vulgare

Normalised receptacle diameter

Infestation	N	Mean	Std. Deviation	Skewness
Uninfested	5159	.973	.200	.058
Floret	1755	1.035	.170	.062
Receptacle	957	1.037	.207	.239
Both	325	1.127	.208	.291
Total	8196	1.000	.199	.080

Centaurea nigra

Normalised capitulum diameter

Infestation	N	Mean	Std. Deviation	Skewness
Uninfested	9154	.984	.103	.147
Non-gall	6808	.992	.100	.168
Gall	4198	1.043	.101	.095
Both	279	1.052	.107	.240
Total	20439	1.000	.105	.145

Table 8 **T test analysis on normalised size between infestation type for all flowers**

Cirsium palustre

Normalised capitulum diameter		
t-test for Equality of Means	t	-2.677
	df	101.10
	Sig. (2-tailed)	.009

Tripleurospermum inodorum

Normalised receptacle diameter		
t-test for Equality of Means	t	-17.483
	df	7985.832
	Sig. (2-tailed)	<0.001

Leucanthemum vulgare **receptacle infestation**

Normalised receptacle diameter		
t-test for Equality of Means	t	-12.666
	df	3517.469
	Sig. (2-tailed)	<0.001

Leucanthemum vulgare **floret infestation by** ***Tephritis neesii***

Normalised receptacle diameter		
t-test for Equality of Means	t	-8.846
	df	1305.981
	Sig. (2-tailed)	<0.001

Centaurea nigra **floret infestation by** ***Chaetostomella cylindrica***

Normalised capitulum diameter		
t-test for Equality of Means	t	-4.815
	df	14865.293
	Sig. (2-tailed)	<0.001

Centaurea nigra* gall infestation by *Urophora quadrifasciata

Normalised capitulum diameter

t-test for Equality of Means	t	-30.873
	df	8276.287
	Sig. (2-tailed)	<0.001

Viewed in the aggregate, all predated heads in each species, as selected by each predating species, show a significant difference in size from uninfested heads at a level of <0.01, and, with the exception of the data from *Cirsium palustre*, only sampled in 2001, at a level of <0.001. This is strong evidence for rejecting the nul hypothesis and accepting that the infested flower heads were indeed selected for size. However, in order to investigate the repeatability of these findings over different years and populations, the data can be analysed for differences within each year and sampling site, as shown in Table 9.

Table 9 T test on normalised size between infestation type by site and date

***Cirsium palustre* (2001)**

Normalised capitulum diameter

Site		
1	t	-2.840
	df	17.40
	Sig. (2-tailed)	.011
2	t	-1.302
	df	22.58
	Sig. (2-tailed)	.206
3	t	-.984
	df	62.96
	Sig. (2-tailed)	.329

Tripleurospermum inodorum

Normalised receptacle diameter

Site	Year		
1	2002	t	-9.572
		df	2338.180
		Sig. (2-tailed)	<0.001
	2003	t	-2.608
		df	508.390
		Sig. (2-tailed)	.009
2	2002	t	-10.669
		df	2333.443
		Sig. (2-tailed)	<0.001
	2003	t	-8.990
		df	957.633
		Sig. (2-tailed)	<0.001
3	2002	t	-4.786
		df	453.173
		Sig. (2-tailed)	<0.001
	2003	t	-4.935
		df	855.578
		Sig. (2-tailed)	<0.001

Leucanthemum vulgare floret infestation

Dependent variables: Normalised receptacle diameter

Site	Year	Statistics	
1	2003	t	-6.499
		df	474.815
		Sig. (2-tailed)	<0.001
	2004	t	-2.909
		df	277.934
		Sig. (2-tailed)	.004
	2005	t	-2.016
		df	414.783
		Sig. (2-tailed)	.044
	2006	t	-6.393
		df	403.911
		Sig. (2-tailed)	<0.001
2	2003	t	-6.369
		df	295.198
		Sig. (2-tailed)	<0.001
	2004	t	-3.558
		df	99.731
		Sig. (2-tailed)	.001
	2005	t	-2.043
		df	35.842
		Sig. (2-tailed)	.048
	2006	t	-3.455
		df	59.021
		Sig. (2-tailed)	.001
3	2004	t	-2.311
		df	94.416
		Sig. (2-tailed)	.023
	2005	t	-2.018
		df	43.258
		Sig. (2-tailed)	.050
	2006	t	-4.246
		df	63.945
		Sig. (2-tailed)	<0.001

Leucanthemum vulgare receptacle infestation

Normalised receptacle diameter

1	2003	t	-.241
		df	4.083
		Sig. (2-tailed)	.821
	2004	t	-.016
		df	46.281
		Sig. (2-tailed)	.987
	2005	t	-.015
		df	10.299
		Sig. (2-tailed)	.989
	2006	t	-.285
		df	7.298
		Sig. (2-tailed)	.784
2	2003	t	-3.358
		df	441.363
		Sig. (2-tailed)	.001
	2004	t	-4.211
		df	366.776
		Sig. (2-tailed)	<0.001
	2005	t	-3.010
		df	147.178
		Sig. (2-tailed)	.003
	2006	t	-4.192
		df	143.026
		Sig. (2-tailed)	<0.001
3	2004	t	-5.508
		df	210.626
		Sig. (2-tailed)	<0.001
	2005	t	-3.129
		df	88.617
		Sig. (2-tailed)	.002
	2006	t	-3.244
		df	51.344
		Sig. (2-tailed)	.002

Centaurea nigra floret infestation

Normalised capitulum diameter

Site	Year		
1	2002	t	-3.005
		df	1541.899
		Sig. (2-tailed)	.003
	2003	t	-2.448
		df	565.306
		Sig. (2-tailed)	.015
	2004	t	-3.000
		df	1268.109
		Sig. (2-tailed)	.003
	2005	t	-.207
		df	834.779
		Sig. (2-tailed)	.836
	2006	t	1.012
		df	935.946
		Sig. (2-tailed)	.312
2	2002	t	.095
		df	953.971
		Sig. (2-tailed)	.924
	2003	t	-2.635
		df	538.093
		Sig. (2-tailed)	.009
	2004	t	-1.839
		df	876.329
		Sig. (2-tailed)	.066
	2005	t	-.823
		df	667.999
		Sig. (2-tailed)	.411
	2006	t	1.304
		df	564.381
		Sig. (2-tailed)	.193
3	2002	t	-.662
		df	930.248
		Sig. (2-tailed)	.508
	2004	t	-2.446
		df	1045.365
		Sig. (2-tailed)	.015
	2005	t	-.664
		df	734.407
		Sig. (2-tailed)	.507
	2006	t	.035
		df	438.170
		Sig. (2-tailed)	.972

Centaurea nigra gall infestation

Normalised capitulum diameter

Site	Year		
1	2002	t	-8.198
		df	103.336
		Sig. (2-tailed)	<0.001
	2003	t	-7.800
		df	388.652
		Sig. (2-tailed)	<0.001
	2004	t	-9.802
		df	418.836
		Sig. (2-tailed)	<0.001
	2005	t	-8.039
		df	727.627
		Sig. (2-tailed)	<0.001
	2006	t	-6.774
		df	249.883
		Sig. (2-tailed)	<0.001
2	2002	t	-7.677
		df	817.566
		Sig. (2-tailed)	<0.001
	2003	t	-7.749
		df	522.107
		Sig. (2-tailed)	<0.001
	2004	t	-8.055
		df	669.814
		Sig. (2-tailed)	<0.001
	2005	t	-8.492
		df	622.461
		Sig. (2-tailed)	<0.001
	2006	t	-2.783
		df	372.854
		Sig. (2-tailed)	.006
3	2002	t	-11.437
		df	1116.723
		Sig. (2-tailed)	<0.001
	2004	t	-11.016
		df	791.612
		Sig. (2-tailed)	<0.001
	2005	t	-11.750
		df	821.993
		Sig. (2-tailed)	<0.001
	2006	t	-6.555
		df	793.624
		Sig. (2-tailed)	<0.001

Looked at in terms of repeatability the data are less clear cut. For *Cirsium palustre* the differences measured between the groups of flowers selected by oviposition are significantly different at a $p<0.02$ only at site 1. However, for *Tripleurospermum inodorum*, the significance is always less than $p<0.01$ at all sites and years between the uninfested and infested groups.

In *Leucanthemum vulgare* the situation is more complicated as two types of infestation are recorded; within the florets Tephritid based and within the receptacle, midge based. For the Tephritid floret infestation, all three sites generally show levels of significance $p<0.05$ and more usually $p<0.01$, except for all sites in 2005, and site 3 in 2004. For the receptacle based infestation, however, at site 1 there is no significant difference between uninfested and infested flowers in any of the years, however the levels of infestation shown in Table 2 reveal very low levels for this type at Site 1, typically 2% - 8%, compared with levels at Site 2 of 15% - 30% and Site 3 between 6% and 13%. At the other sites significance is always $p<0.01$.

Like *L. vulgare*, *Centaurea nigra* demonstrated two predators, free living, non-gall *Chaetostomella cylindrica*, and gall making *Urophora quadrifasciata*. In all cases studied, the significance of size difference between heads selected by gall making predators was less than $p<0.001$, whereas the non-gall predation showed only five out of fourteen combinations of site and year where significance level was better then $p<0.05$.

These levels of significance are sufficient to have a high level of confidence in rejecting a nul hypothesis, and accepting that, in the specific host-predator interactions examined, host choice does include an element of selection by means of flower size in a repeatable manner over space and time for the interactions between *Tripleurospermum inodorum* and its agromyzid predator, and also for *Leucanthemum vulgare* and *Tephritis neesii*, and *Centaurea nigra* and *Urophora quadrifasciata.*

This reflects several other workers data, such as Fenner et al. (2002) and Hemborg and Despres (1999). However this study has the advantage of an extended period of sampling that extends the information over the full course of several seasons, allowing good estimates to be made of overall infestation rates and significant differences from the total sampling period, and discriminating between sampling sites.

4.2 Predation probability

Having accepted the basic hypothesis that predators do discriminate in oviposition sites by flower size, it is logical to extend the question to look for a progressive pattern in this choice. Discrimination should lead to an increasing probability of choosing a flower head for oviposition the larger the flower.

Straw (1989a) presents a review suggesting a general trend towards predator choice of larger flowers, and in (1989b) presents experimental evidence to show a plausible mechanism for predator choice by flower size, based upon the size of ovipositor and length of involucral bracts on the buds.

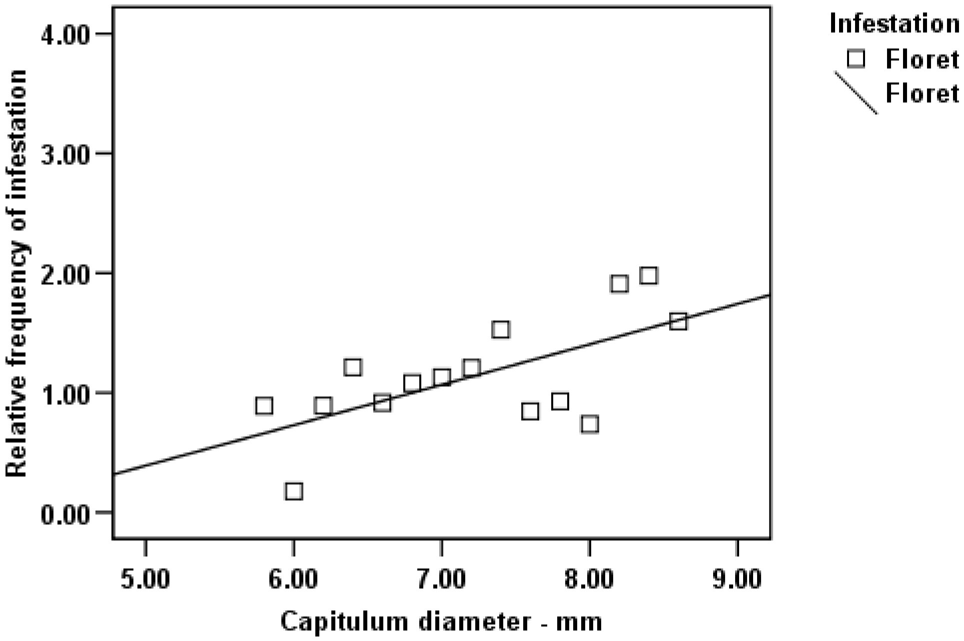
A logistical regression was tried in order to predict the presence if each infestation type on the basis of either actual or normalised flower size, but in no cases was the model significantly successful.

A different approach was devised. Aggregate data for each plant-predator interaction was spilt into capitulum size categories, and the total frequency of predation in each size range determined by dividing the number of infested heads by the total number of sampled heads in that size range. These values were themselves normalised by dividing them by the mean predation frequency of the specific interaction. This gave a measure of the relative probability of attack in relation to normalised capitulum size for each of the interactions investigated.

The relative probability of infestation is presented in Figures 9 and 10 plotted respectively against absolute and normalised flower size. Linear regression statistics are included with each graph

Figure 9 Variation of the relative probability of infestation with absolute size

Cirsium palustre



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.646[a]	.417	.373	.37047

a. Predictors: (Constant), Relative size

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	1.278	1	1.278	9.315	.009[a]
	Residual	1.784	13	.137		
	Total	3.063	14			

a. Predictors: (Constant), Relative size

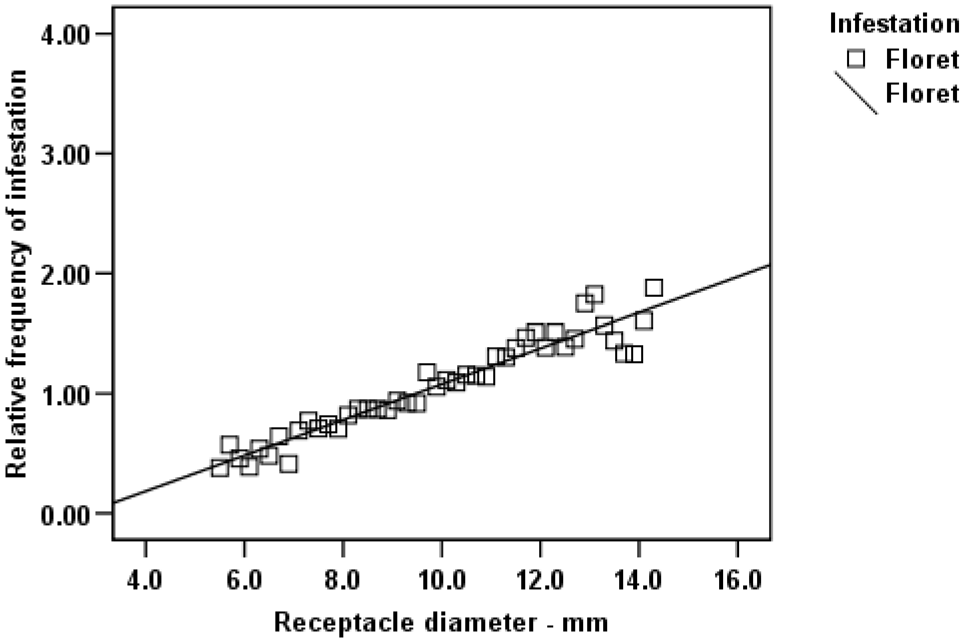
b. Dependent Variable: Relative frequency of predation

Coefficients[a]

		Unstandardized Coefficients		Standardized Coefficients		
Model		B	Std. Error	Beta	t	Sig.
1	(Constant)	-1.298	.803		-1.616	.130
	Relative size	.338	.111	.646	3.052	.009

a. Dependent Variable: Relative frequency of predation

Tripleurospermum inodorum



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.958[a]	.918	.917	.11780

a. Predictors: (Constant), Relative size

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	6.724	1	6.724	484.564	<0.001[a]
	Residual	.597	43	.014		
	Total	7.321	44			

a. Predictors: (Constant), Relative size

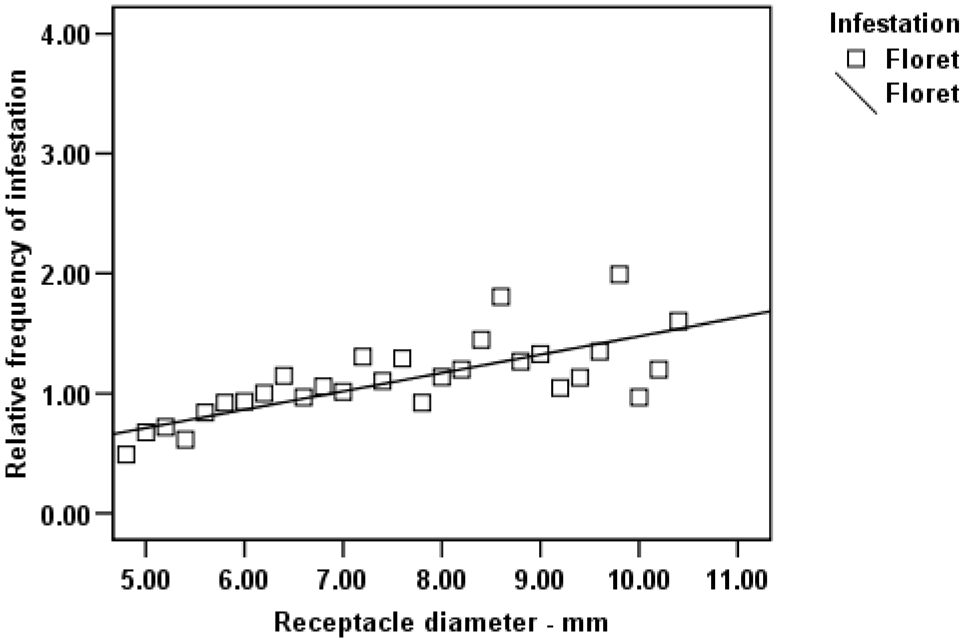
b. Dependent Variable: Relative frequency of predation

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	-.409	.069		-5.917	<0.001
	Relative size	.149	.007	.958	22.013	<0.001

a. Dependent Variable: Relative frequency of predation

Leucanthemum vulgare



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.789[a]	.622	.610	.22749

a. Predictors: (Constant), Receptacle diameter

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	2.559	1	2.559	49.444	<0.001[a]
	Residual	1.553	30	.052		
	Total	4.111	31			

a. Predictors: (Constant), Receptacle diameter

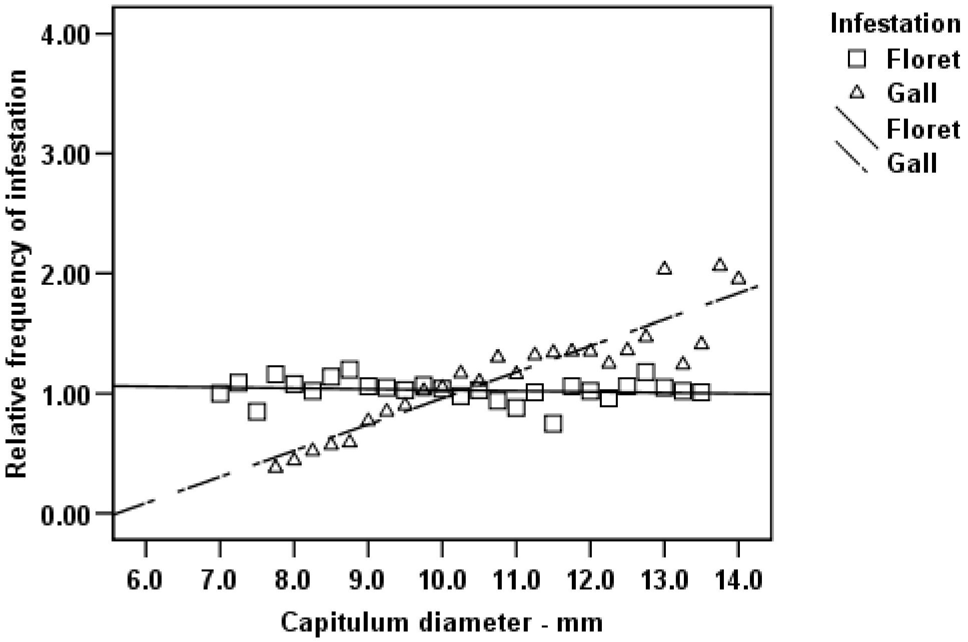
b. Dependent Variable: Relative frequency of predation

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	-.053	.164		-.322	.750
	Receptacle diameter	.153	.022	.789	7.032	<0.001

a. Dependent Variable: Relative frequency of predation

Centaurea nigra



Model Summary

Infestation	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
Floret	1	.155[a]	.024	-.015	.09727
Gall	1	.924[a]	.854	.848	.17631

a. Predictors: (Constant), Capitulum diameter

ANOVA[b]

Infestation	Model		Sum of Squares	df	Mean Square	F	Sig.
Floret	1	Regression	.006	1	.006	.612	.441[a]
		Residual	.237	25	.009		
		Total	.242	26			
Gall	1	Regression	4.367	1	4.367	140.480	<0.001[a]
		Residual	.746	24	.031		
		Total	5.113	25			

a. Predictors: (Constant), Capitulum diameter

b. Dependent Variable: Relative frequency of predation

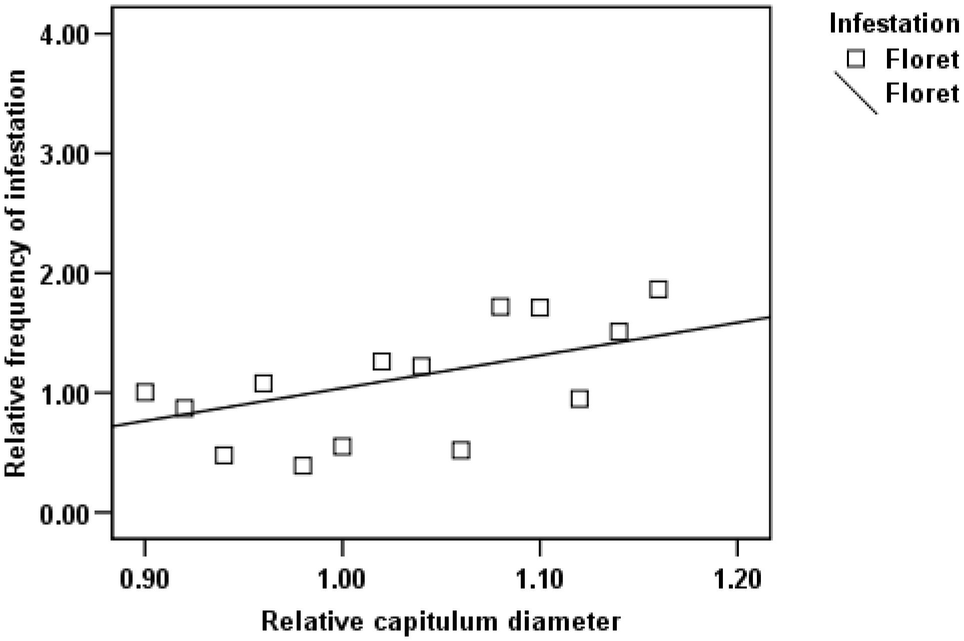
Coefficients[a]

			Unstandardized Coefficients		Standardized Coefficients		
Infestation	Model		B	Std. Error	Beta	t	Sig.
Floret	1	(Constant)	1.105	.100		11.012	<0.001
		Capitulum diameter	-.008	.010	-.155	-.782	.441
Gall	1	(Constant)	-1.225	.204		-6.020	<0.001
		Capitulum diameter	.219	.018	.924	11.852	<0.001

a. Dependent Variable: Relative frequency of predation

Figure 10 Variation of relative probability of infestation with relative flower size

Cirsium palustre



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.591[a]	.349	.302	.41761

a. Predictors: (Constant), Relative size

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	1.308	1	1.308	7.501	.016[a]
	Residual	2.442	14	.174		
	Total	3.750	15			

a. Predictors: (Constant), Relative size

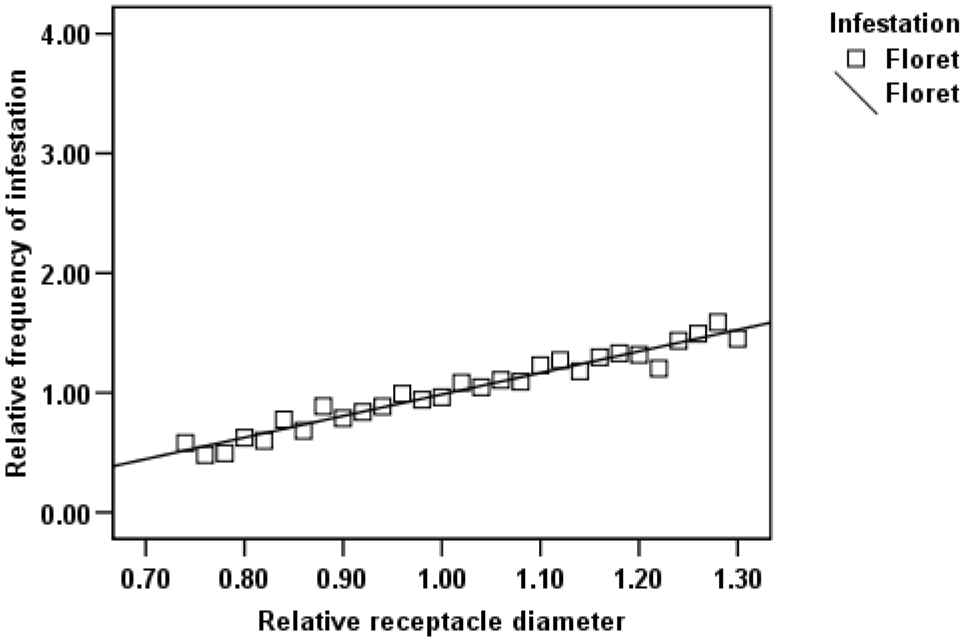
b. Dependent Variable: Relative frequency of predation

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	-2.086	1.148		-1.817	.091
	VAR00001	3.101	1.132	.591	2.739	.016

a. Dependent Variable: Relative frequency of predation

Tripleurospermum inodorum



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.978[a]	.956	.954	.06708

a. Predictors: (Constant), Relative size

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	2.630	1	2.630	584.503	<0.001[a]
	Residual	.121	27	.004		
	Total	2.751	28			

a. Predictors: (Constant), Relative size

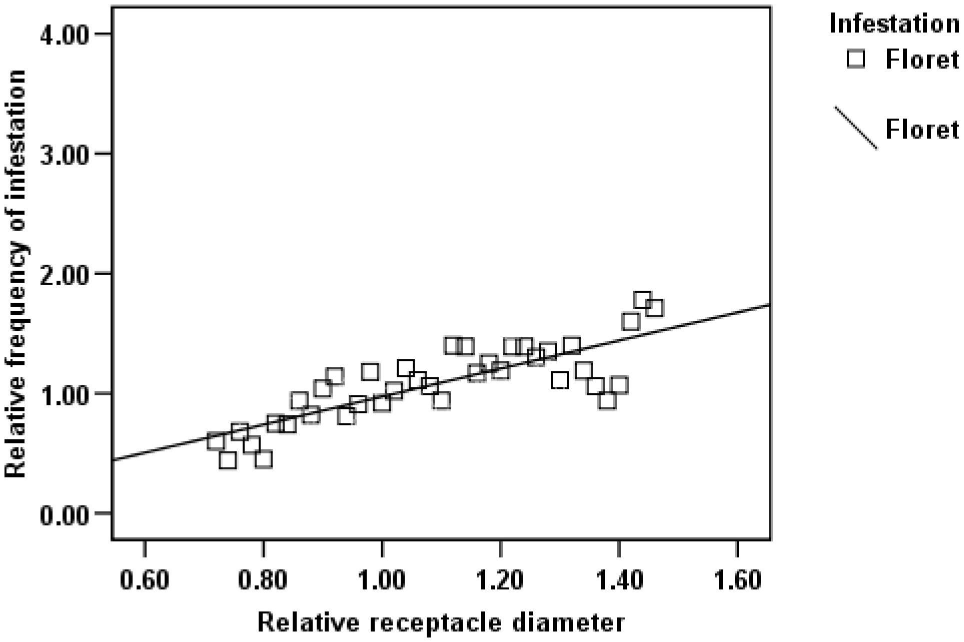
b. Dependent Variable: Relative frequency of predation

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	-.813	.077		-10.569	<0.001
	Relative size	1.800	.074	.978	24.177	<0.001

a. Dependent Variable: Relative frequency of predation

Leucanthemum vulgare



Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.810[a]	.656	.647	.19069

a. Predictors: (Constant), Relative frequency of predation

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	2.498	1	2.498	68.687	<0.001[a]
	Residual	1.309	36	.036		
	Total	3.807	37			

a. Predictors: (Constant), Relative receptacle diameter

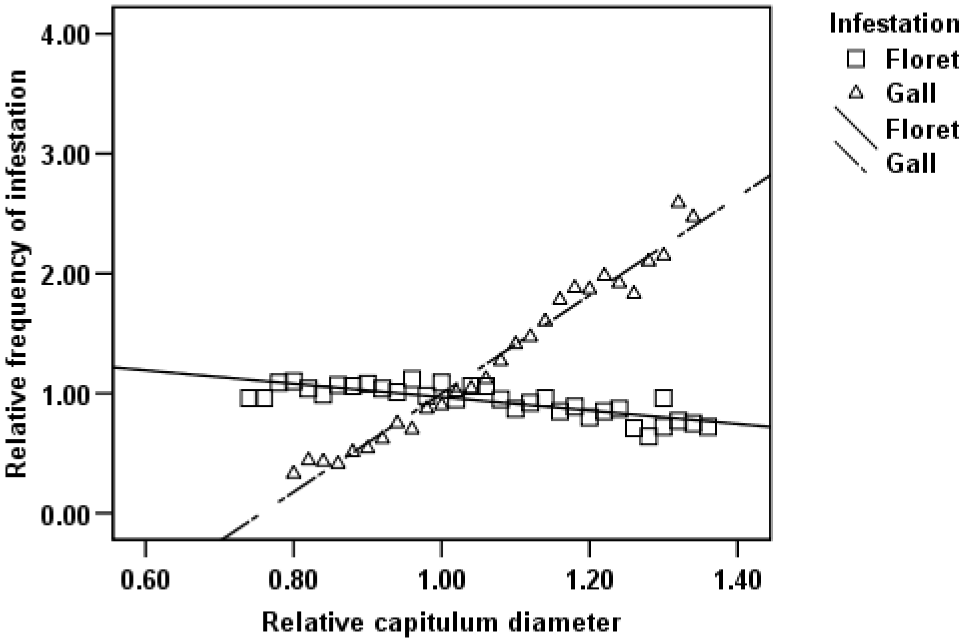
b. Dependent Variable: Relative frequency of infestation

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	-.195	.157		-1.241	.222
	Relative receptacle diameter	1.169	.141	.810	8.288	<0.001

a. Dependent Variable: Relative frequency of infestation

Centaurea nigra



Model Summary

Infestation	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
Floret	1	.797[a]	.636	.624	.08156
Gall	1	.987[a]	.974	.972	.11341

a. Predictors: (Constant), Relative size

ANOVA[b]

Infestation	Model		Sum of Squares	df	Mean Square	F	Sig.
Floret	1	Regression	.360	1	.360	54.117	<0.001[a]
		Residual	.206	31	.007		
		Total	.566	32			
Gall	1	Regression	12.291	1	12.291	955.610	<0.001[a]
		Residual	.334	26	.013		
		Total	12.626	27			

a. Predictors: (Constant), Relative size

b. Dependent Variable: Relative frequency of predation

Coefficients[a]

			Unstandardized Coefficients		Standardized Coefficients		
Infestation	Model		B	Std. Error	Beta	t	Sig.
Floret	1	(Constant)	1.527	.082		18.714	<0.001
		Relative size	-.559	.076	-.797	-7.356	<0.001
Gall	1	(Constant)	-3.103	.144		-21.613	<0.001
		Relative size	4.101	.133	.987	30.913	<0.001

a. Dependent Variable: Relative frequency of predation

From these results it is clear that there is a positive correlation between the probability of infestation and flower size in the interactions of the predator and *Tripleurospermum inodorum* p<0.005, between *Tephritis neesii* and *Leucanthemum vulgare* p<0.005, and between *Urophora* spp. and *Centaurea nigra* p<0.005, a trait also found even at the low infestation rates in *Cirsium palustre* p<0.02. The interaction between *Chaetostomella cylindrica* and *Centaurea nigra* showed a negative correlation p<0.005.

The slopes of these curves can be viewed as a measure of the strength of association between predator choice and flower size, and Table 7 presents their values for the various species.

Table 10 Association slopes all species, based on normalised size

Species	**Year**	**Infestation**	**t statistic**	**Significance**	**Association Slope**
Cirsium palustre	All	Floret	2.739	p<0.02	3.10
Tripleurospermum inodorum	All	Receptacle	24.177	p<0.001	1.55
Leucanthemum vulgare	All	Floret	8.288	p<0.001	1.17
Leucanthemum vulgare	All	Receptacle	4.647	p<0.001	1.10
Leucanthemum vulgare	2003	Floret	7.165	p<0.001	1.35
Leucanthemum vulgare	2004	Floret	4.773	p<0.001	1.31
Leucanthemum vulgare	2005	Floret	3.200	p<0.004	0.86
Leucanthemum vulgare	2006	Floret	3.333	p<0.003	1.31
Centaurea nigra	All	Floret	-7.356	p<0.001	-0.56
Centaurea nigra	2002	Floret	-3.076	p<0.007	-0.60
Centaurea nigra	2003	Floret	-2.169	p<0.05	-0.30
Centaurea nigra	2004	Floret	0.207	p<0.84	0.04
Centaurea nigra	2005	Floret	-4.312	p<0.001	-1.02
Centaurea nigra	2006	Floret	-3.911	p<0.003	-1.19
Centaurea nigra	All	Gall	30.913	p<0.001	4.10
Centaurea nigra	2002	Gall	13.601	p<0.001	4.69
Centaurea nigra	2003	Gall	7.883	p<0.001	4.40
Centaurea nigra	2004	Gall	14.725	p<0.001	4.62
Centaurea nigra	2005	Gall	13.637	p<0.001	4.01
Centaurea nigra	2006	Gall	10.231	p<0.001	3.90

As might be expected, from the differing specificity of predator and host plant, the more specialised gall formers exhibit a greater strength of association than the more generalist feeders. In the case of *Tephritis neesii* and *Leucanthemum vulgare* the slope may be reduced by the extended individual phenology exhibited by the plant. The oligophagous *Chaetostomella cylindrica* has a slope of association close to zero, indicating that there is no appreciable size choice involved in its selection. Clearly each individual predator host interaction has its own character, which impacts significantly upon their ability to form selective pressures.

As the two predators of *C. nigra* exhibit different attack patterns, its growing season can be viewed as seasons of low and high competition for oviposition sites. Taking the low season as days up to 190, and greater than 210, and high season as between those dates, based upon the average date that infestation of *U. quadrifasciata* equals of exceeds that of *C. cylindrica*, the analysis reveals the following values.

Table 11 **Association slopes *C. nigra* by date**

Species	**Season**	**Infestation**	**t statistic**	**Significance**	**Association Slope**
Centaurea nigra	Low	Floret	-5.886	p<0.001	-0.55
Centaurea nigra	High	Floret	-4.458	p<0.001	-1.07
Centaurea nigra	Low	Gall	19.149	p<0.001	4.48
Centaurea nigra	High	Gall	22.799	p<0.001	3.83

The differences between periods of low and high competition for oviposition open the possibility of demonstrating both intra and inter specific competition. During the period of high competition, *Chaetostomella cylindrica* appears less likely to choose larger flowers than during low competition with *Urophora* spp. Similarly *Urophora* spp. appear to reduce their preference for larger flowers during the period of high competition.

Having established the statistical significance of the difference between flower sizes chosen for oviposition, the analysis of relative probability of oviposition demonstrates a consistent pattern for each predator-host interaction, indicating that for both *Tephritis neesii/Leucanthemum vulgare* and *Urophora spp/.Centaurea nigra* interactions there is a positive correlation between flower size and the relative probability of infestation. Taken together, these two results should allow the overall hypothesis that predators tend to select larger flower heads as oviposition sites in a repeatable pattern to be accepted.

4.3 Sampling effects

Despite this result, however, there remains one question that has as yet proved difficult to address, the possibility of increased flower size as an artefact of the presence of predators. Despite Straw's direct measurement of predator choice for larger flowers, this remains a significant obstacle to proving the effect as a probable selective pressure on flower size.

It is obviously in the interest of insect larvae to increase the delivery of food resource to their location, and there is no reason to suppose that this is limited to the building of galls structures. However, the common view of any plant gall implies swelling of the material, so the *Urophora*/*Centaurea* interaction is most open to question regarding its strength of association.

In 2005 a small experiment within the location of one collection site, involving application of insecticide, effectively excluded predator larvae for several weeks, and the resulting sample of flower heads actually showed a slightly greater mean size than those close by where predation continued, however this could have been the result of different resource availability within the separate patches, which had not been separately sampled before. However, the experiment was inconclusive, and might have affected other insects that could affect the result, so its findings are not accepted.

As a further check statistical information on populations of flowers where oviposition is random, but results in an increase in flower size might be gathered. In order to match this, a normally distributed population was modelled, and a sample of 40% was randomly selected for treatment. Two types of treatment were applied, firstly a simple absolute increase in capitulum size, and secondly a size increase relative to original size.

Based upon an original capitulum diameter of 10mm, representative of the values found among *C. nigra*, a 2mm absolute size increase or a 20% size increase was necessary in order to achieve relative probability association slopes of 4.2 and 4.8 respectively. However, where the normalised mean of galled *Centaurea nigra* flowers in the field was 1.045, in both simulations the normalised mean was measured at 1.11, more than twice the difference from the overall mean.

Such a population model also shows highly variable association slope values for different infestation rates. Using a 2mm absolute capitulum diameter increase on a normally distributed population with a mean capitulum size of 10mm, consisting of 5000 heads as an example, the association slopes for different infestations were as follows:

Table 12 Association slopes

Modelled infestation	Association Slope
10%	18.48
20%	10.10
30%	6.17
40%	4.19
50%	2.70
60%	1.65

In the real populations, however, the association slope was seen to be independent of the rate of infestation over infestation rates for *C. nigra* by Urophora spp. ranging between 13% and 33%.

Although this discrepancy between the field data and a simple model cannot discount the possibility of there being some size increase due to the presence of galls, it adds significant weight to the conclusion that the effect is real.

In summary, despite the inherent difficulty of proving a negative, there are three arguments in favour of the measured data representing a real effect

> Straw (1989b) has demonstrated a mechanism for oviposition choice by means of direct measurement by the insect of the depth between capitulum and bract.
>
> Several authors, Hainsworth et al. (1984), Campbell et al. (2002), Petersen (2000) and Fenner et al. (2002) have presented data showing oviposition preference for larger flowers.
>
> The statistical differences between the measured data and those that would mimic the relative probability data by random selection of flower followed by capitulum swelling are large.

Chapter 5. The effects of infestation and flower size on fecundity

Two hypotheses defined in order to determine the role of pre-dispersal seed predation and capitulum size in floral evolution are:

The presence of seed predators within flower heads reduces their fecundity.

The size of flower heads is correlated with successful seed set.

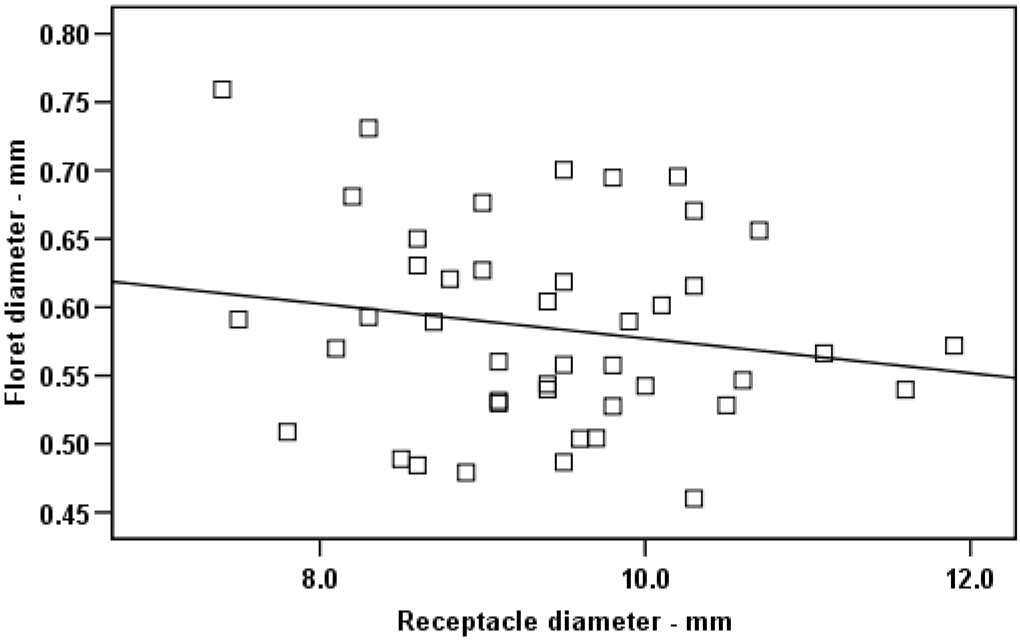
As described in Chapter 3, sampling of the dried heads of *T. inodorum*, *L. vulgare* and *C. nigra* was undertaken in order to assess the effects of predation and size on fecundity. All three species are described as primarily insect pollinated and self-incompatible, Kay (1994), Howarth (1968) and Lack (1982). These data allow calculation not only of the effects of predation, but also of pollination success, provided that the number of set seeds is related to the potential seed load per head, which is the number of florets.

5.1 Individual floret size

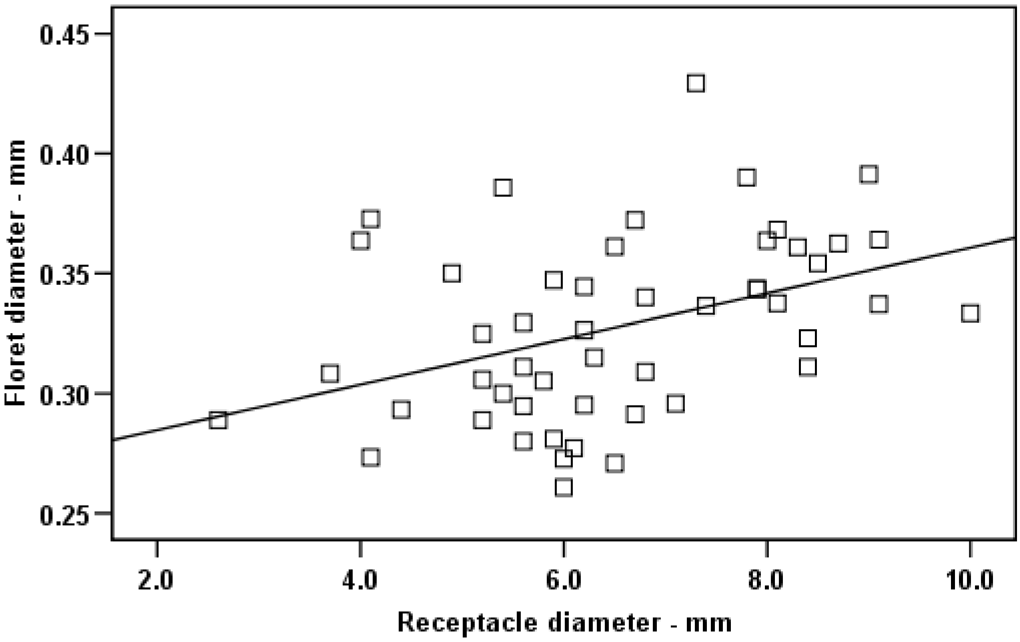
In order to investigate pollination success as a function of flower size, a number of the flowering heads recorded in the standard infestation sampling were further investigated by counting the number of florets across their diameter, following the first cut in their dissection. Dividing the flower size measure by the number of florets gave a measure of effective floret diameter (a value relative to the flower size measure rather than an absolute floret size). Plotting the resulting data and fitting a second order polynomial to the data allowed an equation to be developed for each species that predicted the potential number of seeds for any flower head based upon its characteristic size. The variation of floret size with flower size, used in order to calculate a mean level of seed potential if shown in Figure 11.

Figure 11 Variation of floret size with flower size

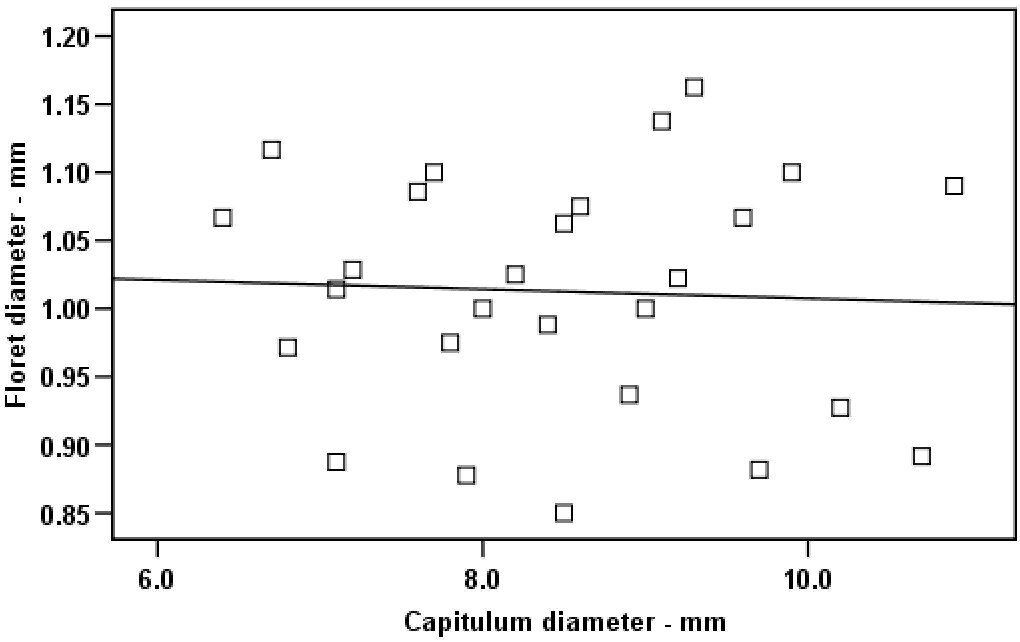
Tripleurospermum inodorum



Leucanthemum vulgare



Centaurea nigra



These data show a relatively stable relationship between flower size and seed potential for the three species. However, although the linear regressions might be reasonable estimates to use in the mathematical modelling, only that for *L. vulgare* shows a significance $p<0.05$ for its linearity, as shown in Table 13.

Table 13 Regression coefficients for floret diameter as a function of flower size

Tripleurospermum inodorum

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	.757	.090		8.377	<0.001
	Receptacle diameter - mm	-.018	.010	-.276	-1.885	.066

a. Dependent Variable: Floret diameter - mm

Leucanthemum vulgare

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	.266	.021		12.770	<0.001
	Diameter	.009	.003	.405	3.064	.004

a. Dependent Variable: Floret_diamter

Centaurea nigra

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	1.041	.122		8.535	<0.001
	Capitulum diameter - mm	-.003	.014	-.048	-.238	.814

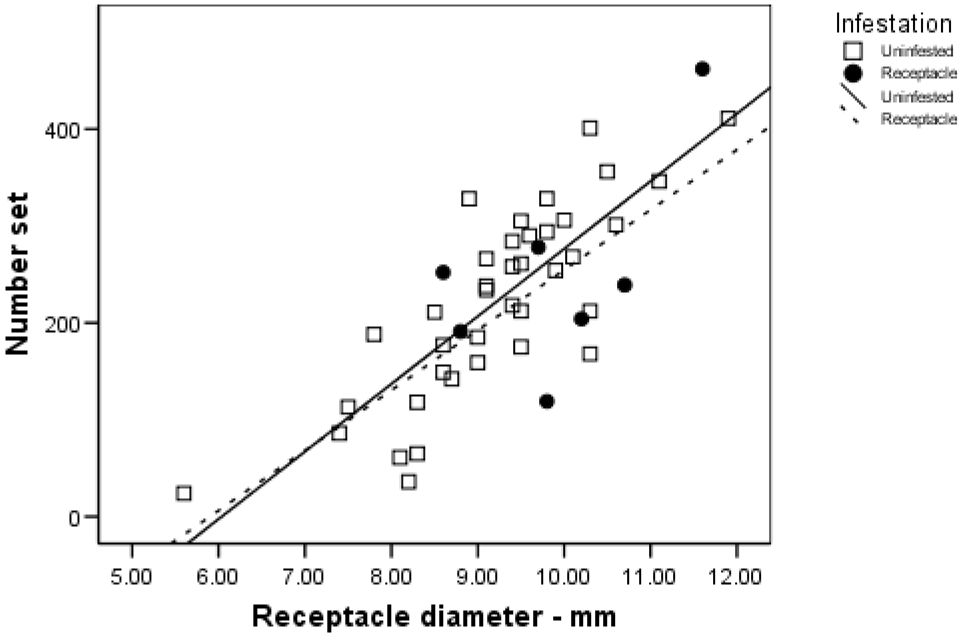
a. Dependent Variable: Floret diameter - mm

5.2 Viable seeds per head and the effect of predation

The results of the seed counting of viable seeds are shown in Figure 12.

Figure 12 Variation of seed set with flower size

Tripleurospermum inodorum

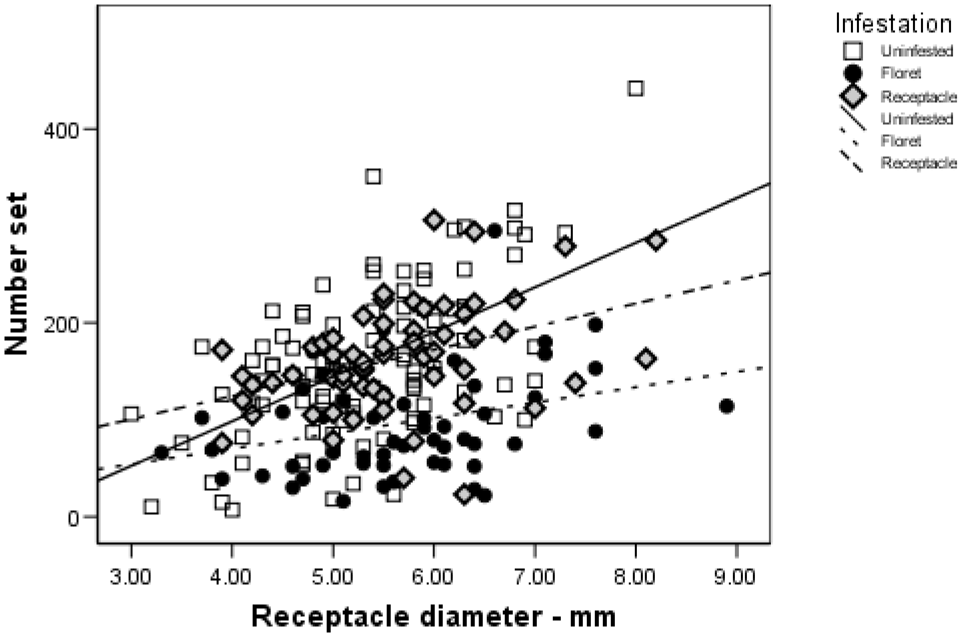


Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	-412.469	79.335		-5.199	<0.001
	Receptacle diameter - mm	68.823	8.537	.796	8.062	<0.001
	Infestation	-20.576	26.617	-.076	-.773	.444

a. Dependent Variable: Set

Leucanthemum vulgare



Coefficients for floret based regression

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	-16.277	27.476		-.592	.554
	Receptacle diameter - mm	32.955	5.042	.437	6.536	<0.001
	Infestation	-74.405	10.470	-.475	-7.106	<0.001

a. Dependent Variable: Set

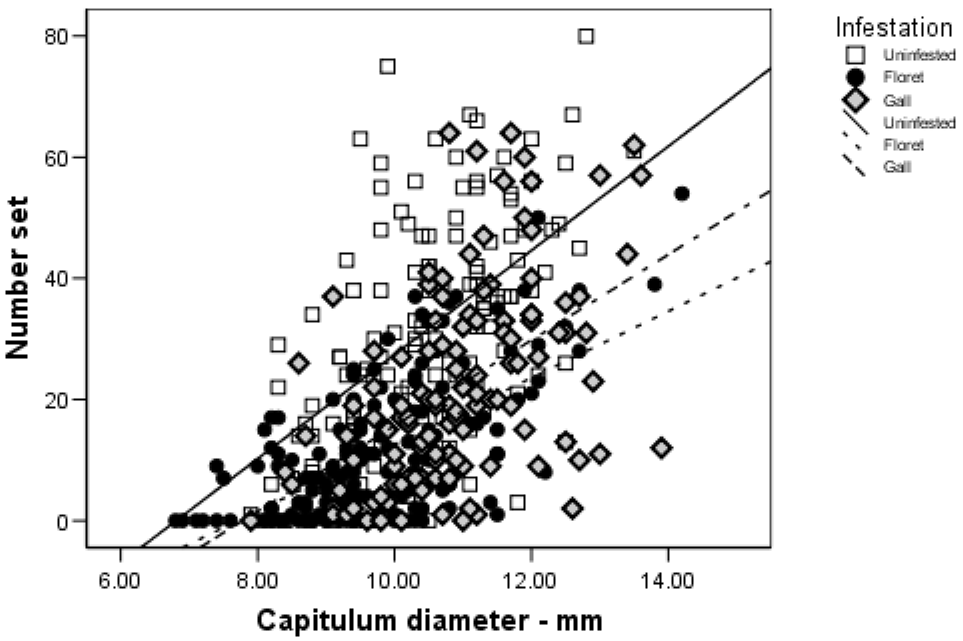
Coefficients for receptacle based regression

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	-42.404	28.853		-1.470	.144
	Receptacle diameter - mm	37.881	5.309	.504	7.135	<0.001
	Infestation	-3.555	5.186	-.048	-.685	.494

a. Dependent Variable: Set

Centaurea nigra



Coefficients for floret based regression

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	-38.325	6.372		-6.015	<0.001
	Capitulum diameter - mm	6.672	.597	.471	11.177	<0.001
	Infestation	-15.600	1.537	-.427	-10.147	<0.001

a. Dependent Variable: Set

Coefficients for gall based regression

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	-50.631	8.941		-5.663	<0.001
	Capitulum diameter - mm	7.844	.841	.500	9.327	<0.001
	Infestation	-6.413	.994	-.346	-6.450	<0.001

a. Dependent Variable: Set

Considering only the data relating to uninfested flowers, all three flowers show a correlation of seed set and flower size significant to $p<0.001$. This illustrates a seemingly obvious point that, within the Asteraceae, larger flowers tend to be significantly more fecund than smaller ones, irrespective of predation and may thus be inherently advantageous in an evolutionary sense. This is a result connected with the physiology of Asteraceae, where flower heads are composites of smaller florets, each of which is relatively insensitive to the overall flower size.

In *T. inodorum*, there in very little difference between the regression lines for uninfested and infested heads and the regression shows no level of significance, p<0.445, for including the effect of infestation. From this it can be concluded that infestation has no significant effect on fecundity, which, considered against the other examples, argues against any general familial wide hypothesis for the effect of predation on fecundity. This conclusion led to no further sampling of *T. inodorum*, although is important in coming to a view that each plant-insect interaction is a unique phenomenon.

For *L. vulgare*, there are two interactions to consider. The regression coefficient for flowers infested by the floret based *T. neesii* show a significance of p<0.001, whilst that for those infested by the receptacle based midge is not significant at p<0.495. It appears therefore, that the midge has little impact on fecundity, whilst *T. neesii* does, and is therefore a candidate for inclusion in a mathematical model that might illustrate the evolutionary impact of predation. Again, the individuality of different insect-plant interactions is highlighted.

In *C. nigra*, however, both the floret based *C. cylindrica* and the gall forming *U. quadrifasciata* show a significant effect on fecundity, at p<0.001, which makes both interactions suitable for modelling.

5.3 Relative seed set

For each dried head investigated, the number of viable seeds was converted into a value of relative seed set success by dividing by the calculated value of seed potential, Seed set potential is calculated by using the regression equations developed in section 5.1 to calculate the mean floret diameter for each flower based upon its characteristic size, and, using simple geometry the expected number of florets. Expressed as percentage success, the results for each flower are shown in Table 14.

Table 14 Seed set success as a percentage

Tripleurospermum inodorum

Success %

Infestation	Mean	N
Uninfested	81.6	38
Receptacle	81.1	7
Total	81.5	45

Leucanthemum vulgare

Success %

Infestation	Mean	N
Uninfested	56.9	98
Floret	32.1	59
Receptacle	55.0	59
Total	49.6	216

Centaurea nigra

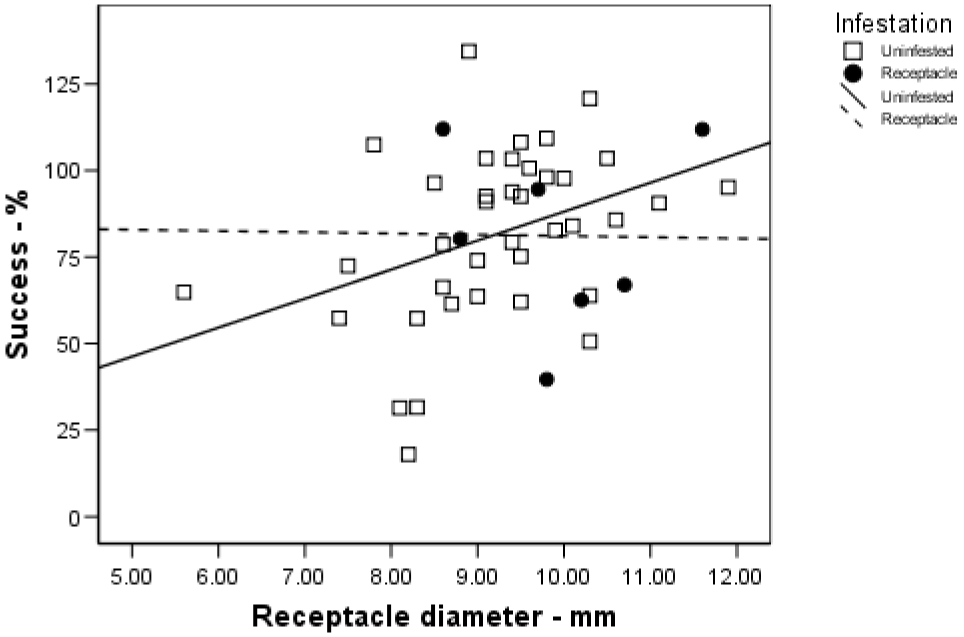
Success %

Infestation	Mean	N
Uninfested	46.7	127
Floret	30.2	173
Gall	30.3	121
Total	35.2	421

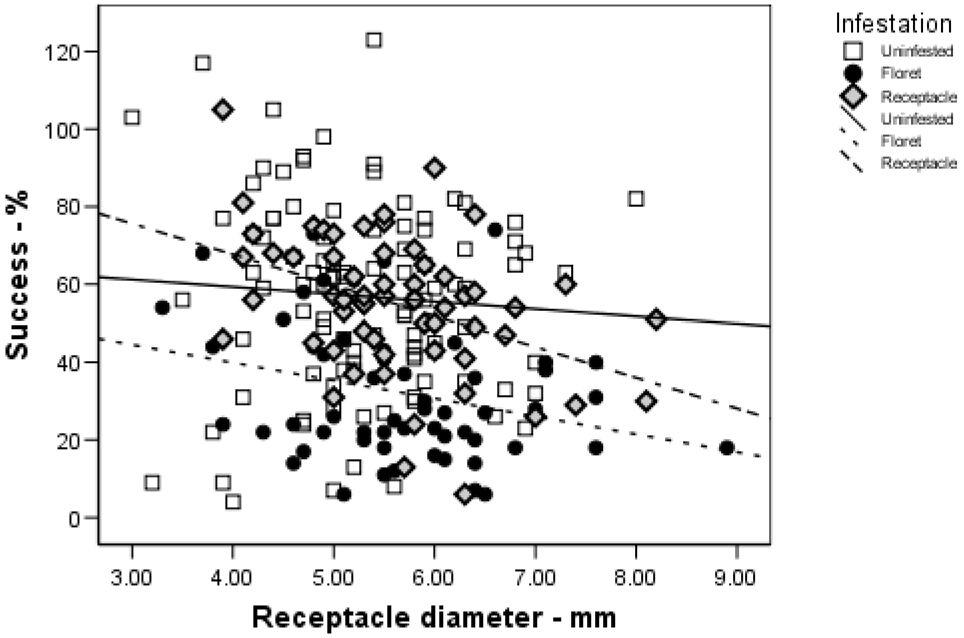
Graphs of success against flower size were then created for uninfested and infested flowers and the resulting variation of seed set success is shown in Figure 13, shown by infestation class.

Figure 13 Seed set variation with flower size

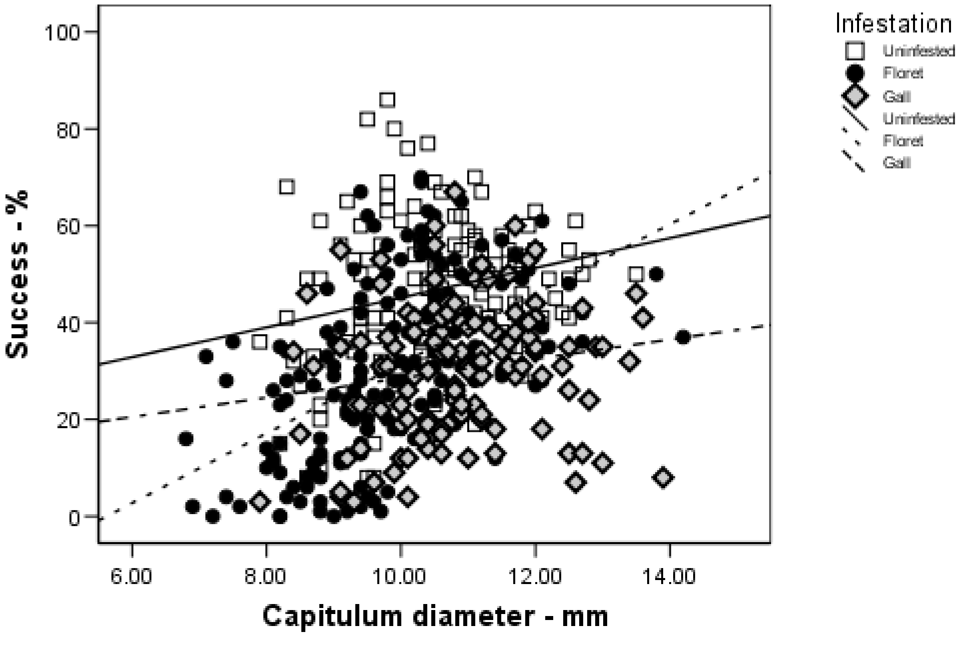
Tripleurospermum inodorum



Leucanthemum vulgare



Centaurea nigra



In order to test for significant differences between levels of success in uninfested and infested flower heads, a t test, without assuming homogeneity of variance was applied to the frequency of success value, as shown in Table 15.

Table 15 T test of success frequency for each plant insect interaction

Tripleurospermum inodorum **by Agromyzid**

Success %		
t-test for Equality of Means	t	.040
	df	8.027
	Sig. (2-tailed)	.969

Leucanthemum vulgare **by** ***Tephritis neesii***

Success %		
t-test for Equality of Means	t	7.356
	df	152.382
	Sig. (2-tailed)	<0.001

Leucanthemum vulgare **by midge**

Success %		
t-test for Equality of Means	t	.547
	df	149.535
	Sig. (2-tailed)	.585

Centaurea nigra **by** ***Chaetostomella cylindrica***

Success %		
t-test for Equality of Means	t	8.630
	df	288.303
	Sig. (2-tailed)	<0.001

Centaurea nigra* by *Urophora quadrifasciata

Success %		
t-test for Equality of Means	t	8.821
	df	245.349
	Sig. (2-tailed)	<0.001

The results on seed set success show similar results to the effects of including infestation as a term in the regression for the number of set seeds in section 5.2. For *Tripleurospermum inodorum* it is clear that there is no significant effect on fecundity due to infestation, and thus that for this particular plant-insect interaction the hypothesis of predation reducing fecundity is not shown. In these terms, the Agromyzid infestation of receptacles in *T. inodorum* would appear to be commensal rather than predatory, and is unlikely to provide selective pressure on the floral traits under consideration.

For *Leucanthemum vulgare* receptacle infestation, the midge species, there does not appear to be any reduction in fecundity, but when infested by *Tephritis neesii* the success rate falls from and average 57% to 32% with a level of significance for frequency of success of p<0.001.

Similarly fecundity of *Centaurea nigra* is reduced from a level of 47% of potential seeds to about 30% due to predation by both *Chaetostomella cylindrica* and *Urophora* spp again with significance levels for success frequency of p<0.001.

The assessment of the success of seed set has been an important method of determining the insect plant interactions that might play a part in the evolution of floral traits. An inspection of the data shows immediately that in the *Tripleurospermum inodorum*, the presence of *agromyzid* larvae does not affect levels of fecundity, nor does the receptacle based midge larva alter the success of *Leucanthemum vulgare*.

By calculating the significance of a linear regression for seed set success with flower size, on uninfested flowers, it may also be possible to comment upon the effect of flower size on pollination success. The values are shown in Table 16.

Table 16 Linear regression of seed set success with flower size

Tripleurospermum inodorum

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	.044	.314		.140	.889
	Receptacle diameter - mm	.084	.034	.382	2.477	.018

a. Dependent Variable: Success

Leucanthemum vulgare

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	.669	.145		4.608	<0.001
	Receptacle diameter - mm	-.019	.027	-.071	-.701	.485

a. Dependent Variable: Success

Centaurea nigra

Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.
1	(Constant)	.145	.121		1.191	.236
	Capitulum diameter - mm	.031	.011	.233	2.673	.009

a. Dependent Variable: Success

Both *T. inodorum* and *C. nigra* display a positive correlation with significance of $p<0.02$ and $p<0.01$ respectively. This would imply that larger flowers are more successful in attracting pollinators than smaller ones, and thus might demonstrate a selective pressure for increased floral size. *L. vulgare*, however, shows no significant correlation between flower size and relative success of seed set. As noted above, larger flowers do achieve a higher numerical level of seed set, due to their increased number of available florets, however, for *L. vulgare* it would appear that similar fecundity might be achieved as easily by increasing the number of flower heads per plant rather than increasing flower size.

It is also clear that overall fecundity where predation by tephritid species is absent, at some 57% and 47% respectively for *Leucanthemum vulgare* and *Centaurea nigra*, is significantly reduced from the number of florets available within the capitulum. Whether this is due to differing pollination success, predation from other insect sources, fertility failure within individual achenes, or lack of resource supply, each of which is likely to play a part, is not possible to determine from these data. However, from the perspective of selective pressures applied by Tephritid predation all of these sources can be taken together as determining a base level of seed set against which to measure the effect of predation. It is also noted that very few heads of any species showed no seed set at all.

For both *T. inodorum* and *L. vulgare* all measured and uninfested heads had some level of seed set, and for *C. nigra* less than 5% of measured uninfested heads had no seed set, which indicates that there is little evidence to show lack of any pollination visits at all to any flower.

Using this approach, Tephritid predation results in significant reductions in fecundity in both *L. vulgare* and *C. nigra*, but not in *T. inodorum*. There is some evidence to show that for *Leucanthemum vulgare* the damage done by infestation is greater in larger flower heads, whilst in *Centaurea nigra* the opposite trend, towards more relative damage in smaller flowers, is found.

This result should allow the overall hypothesis that the presence of seed predators within flower heads reduces their fecundity to be accepted for both *L. vulgare* infested by *T. neesii*, and *C. nigra* infested by either *C. cylindrica* or *U. quadrifasciata*.

Overall fecundity is also shown to be significantly correlated to absolute flower size, although for relative pollination success this is not the case for *L. vulgare*.

Chapter 6. Phenology of oviposition

The fourth hypothesis defined in order to determine the role of pre-dispersal seed predation in floral evolution is that:

> The pattern of seed predation over the course of a season is repeatable over several seasons, and locations

The requirement for repeatability, in terms of levels of infestation and phenological pattern, over time is necessary to support an hypothesis of sustained local selective pressure due to predation, especially for plants other than annuals. To extend this to a more general case a geographical element is also required. Clearly this study, although extending over sampling areas separated by up to ten miles, cannot answer either question definitively, but might be able to suggest the likelihood of a wider pattern.

6.1 Pattern of infestation levels across locations and years

The overall seasonal totals of infestation levels recorded in the data are shown in Table 17.

Table 17 Variation of overall predation rates over location and time

Cirsium palustre

	Predation - %
Site	2001
1	1.2
2	1.8
3	5.2

Tripleurospermum inodorum

	Predation - %	
Site	2002	2003
1	44.4	19.5
2	41.6	35.4
3	54.2	39.8

Leucanthemum vulgare

	Floret predation - %			
Site	2003	2004	2005	2006
1	67.6	40.2	53.1	50.2
2	16.4	10.5	4.4	7.9
3		6.9	3.6	6.6

Centaurea nigra

	Floret predation - %				
Site	2002	2003	2004	2005	2006
1	43.4	62.9	33.7	28.7	36.8
2	33.8	55.8	28.4	27.4	27.5
3	27.3		25.1	24.8	20.3

	Gall predation - %				
Site	2002	2003	2004	2005	2006
1	5.0	13.0	13.6	23.8	13.0
2	27.1	17.6	22.9	26.3	22.6
3	33.8		22.1	26.5	28.9

Although these data are not extensive, it is possible to apply a linear regression across the years of sampled data for both *Leucanthemum vulgare* and *Centaurea nigra*, in order to see if the levels are relatively constant, or if there is any association with time for the overall infestation levels. Table 18 presents the results

Table 18 ANOVA of regression for frequency of infestation with year of sampling

***Leucanthemum vulgare*[b]**

Model: 1
Regression

Infestation	Site	F	Sig.
Floret	1	.502	.552[a]
	2	3.713	.194[a]
	3	.007	.948[a]
Receptacle	1	.028	.884[a]
	2	1.009	.421[a]
	3	6.658	.235[a]

a. Predictors: (Constant), Year

b. Dependent Variable: Percent infested

***Centaurea nigra*[b]**

Model: 1
Regression

Infestation	Site	F	Sig.
Floret	1	1.308	.336[a]
	2	1.159	.361[a]
	3	6.002	.134[a]
Gall	1	1.990	.253[a]
	2	.000	1.000[a]
	3	.518	.547[a]

a. Predictors: (Constant), Year

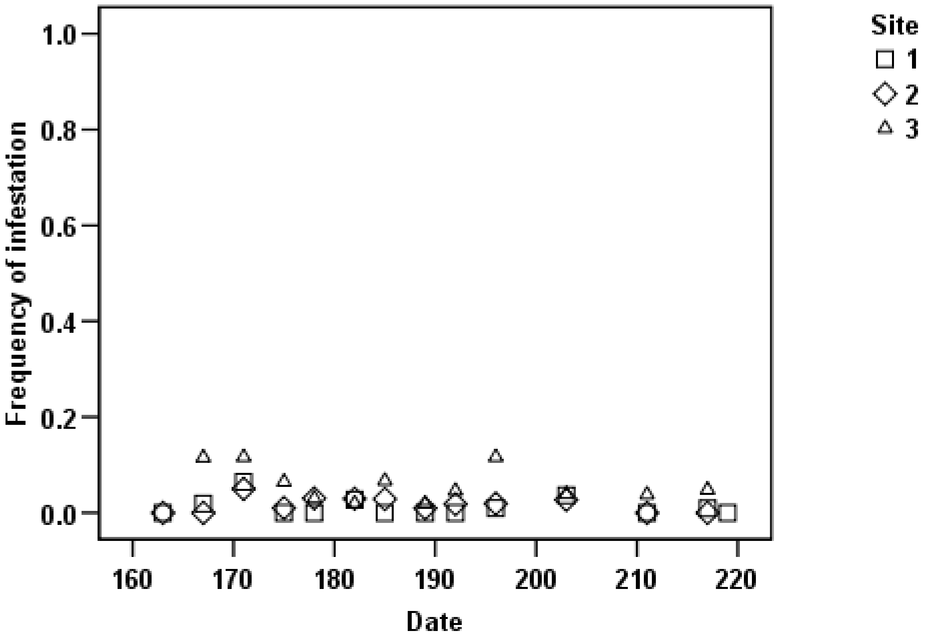
b. Dependent Variable: Frequency of infestation - %

Although the number of data are small, these results do not show a significant correlation between levels of infestation and year of sampling, perhaps this is not surprising, as annual climatic variation will, inevitably have a significant effect on these interactions. Clearly there are random effects, both temporal and spatial, that result in predation levels varying over a range of levels in real populations. In order to show an evolutionary effect on overall populations therefore, it will be necessary to demonstrate it over a range of total infestation levels, and accept significant differences in that effect over seasons and localities.

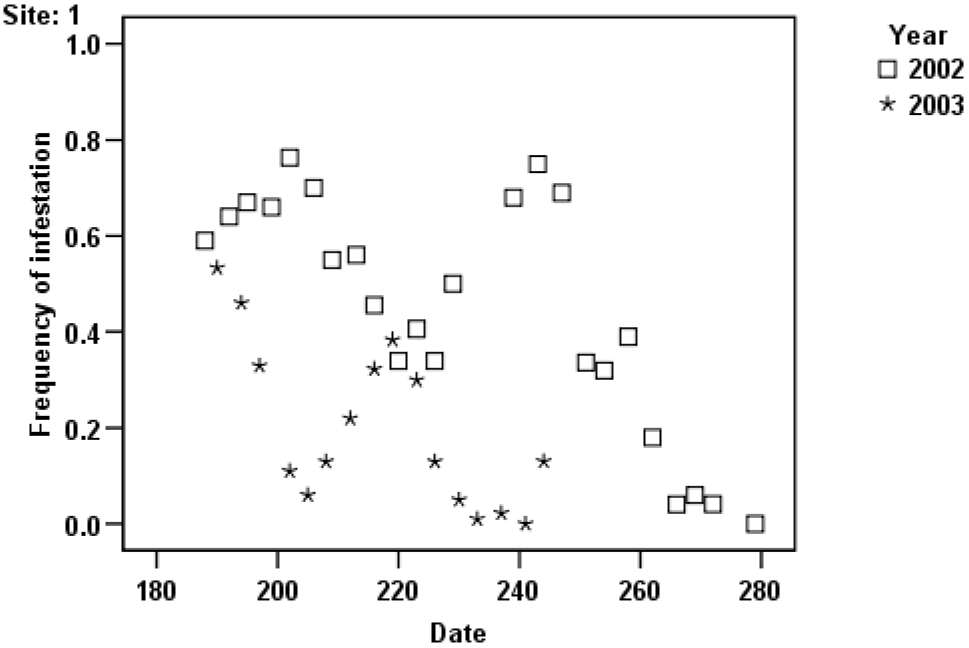
6.2 Variation of infestation through the course of seasons

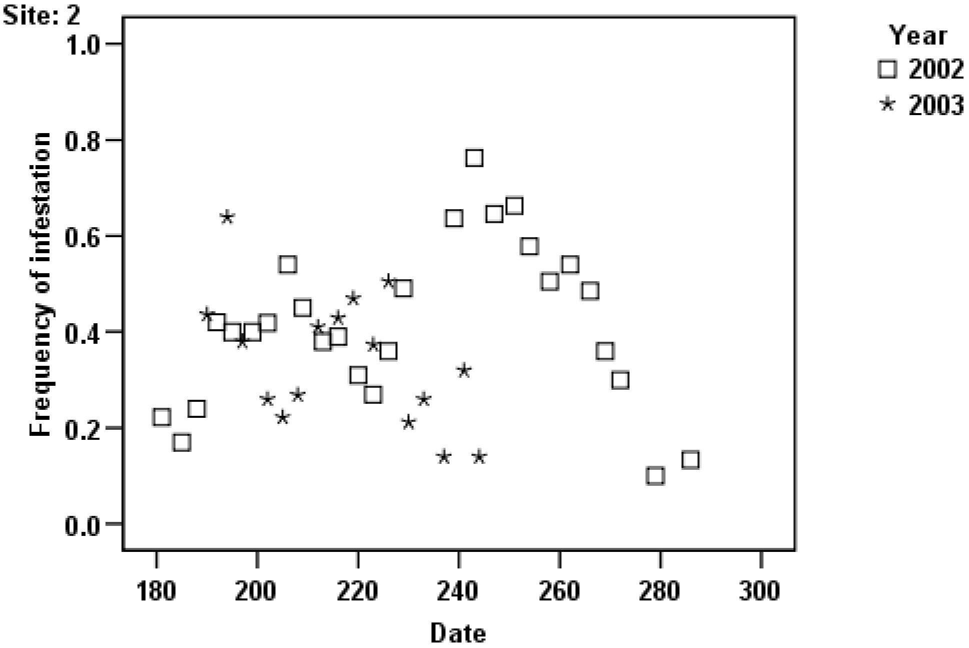
The variation of relative infestation frequency with day of sampling is shown in Figure 14 for each species, year and site.

Figure 14 Variation of infestation frequency with day of sampling
***Cirsium palustre* (2001)**

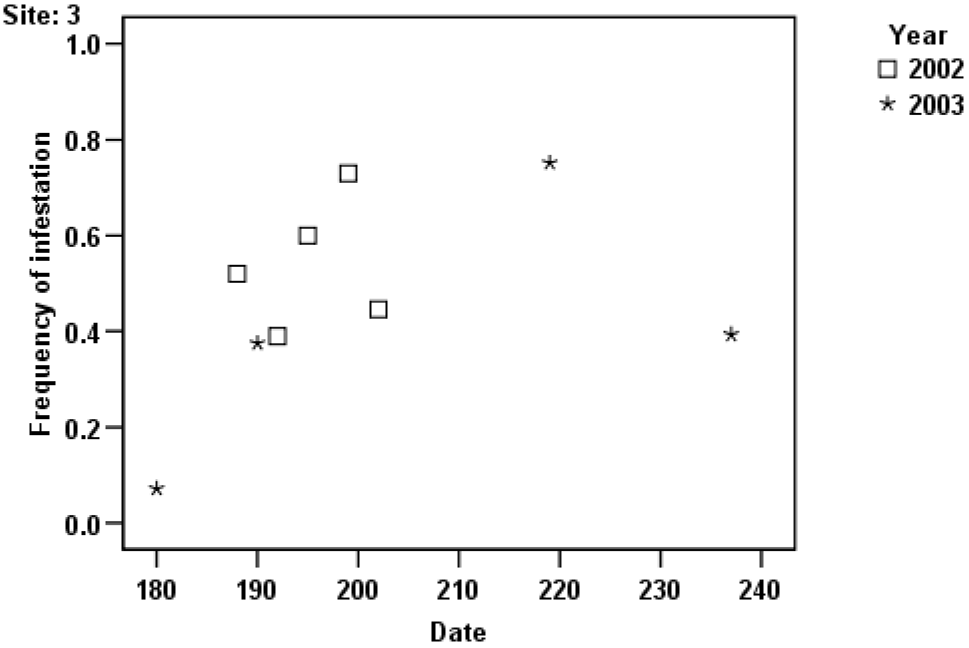


Tripleurospermum inodorum

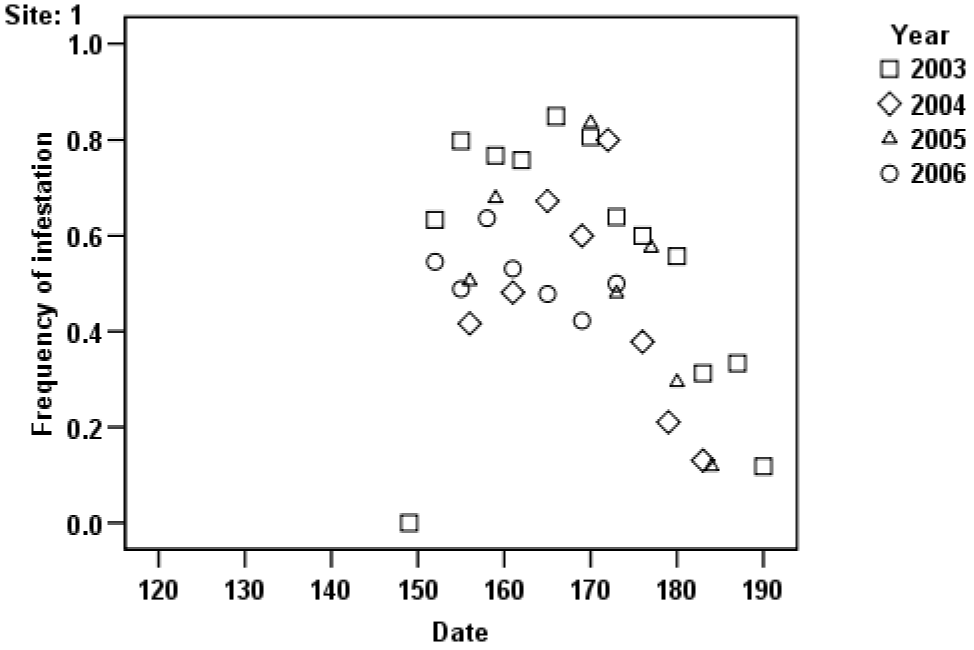


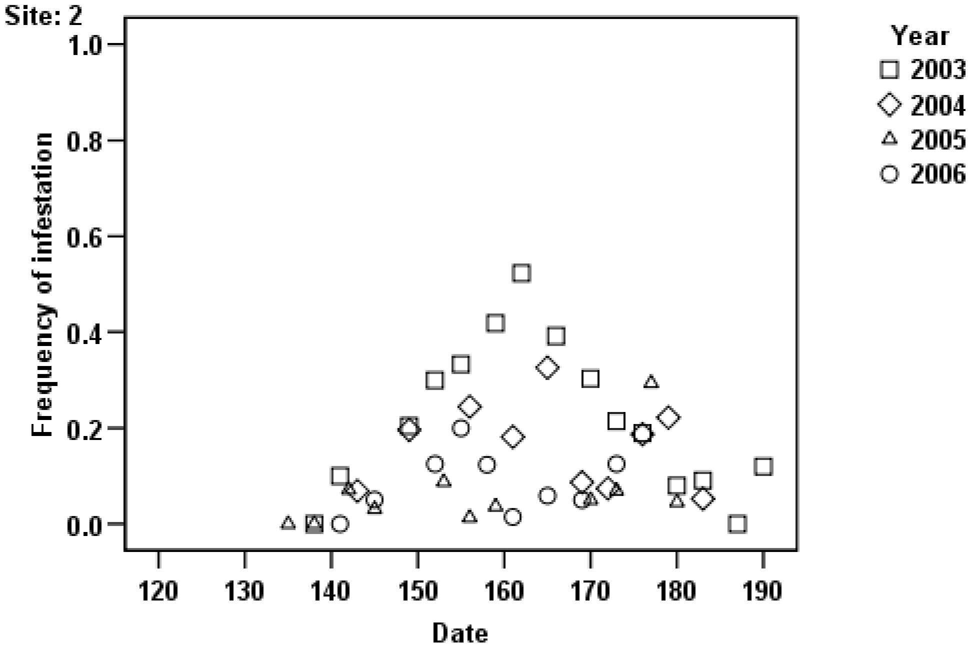


Tripleurospermum inodorum

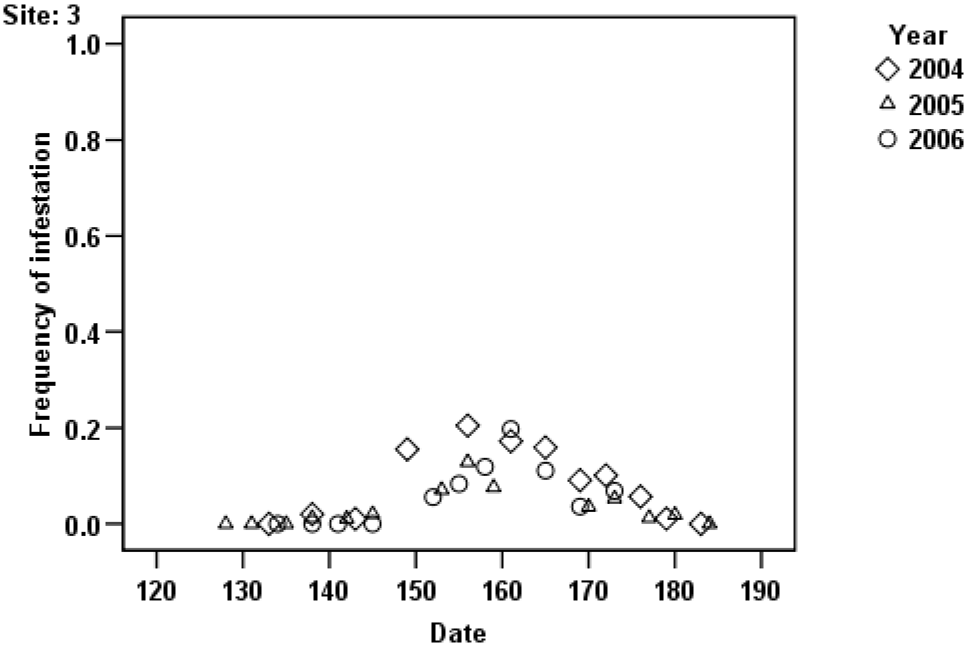


Leucanthemum vulgare* by *Tephritis neesii

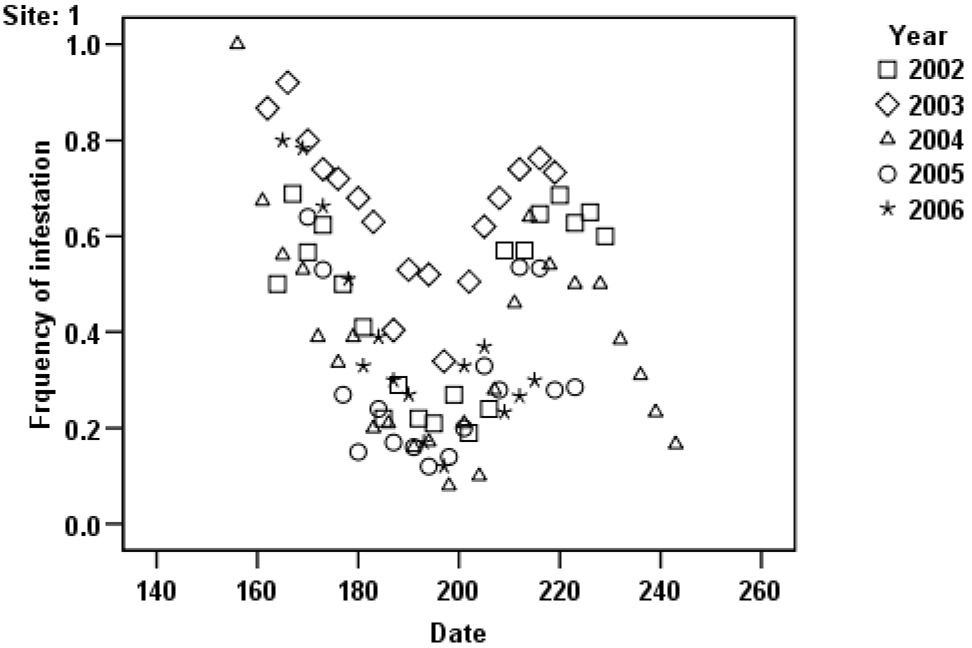


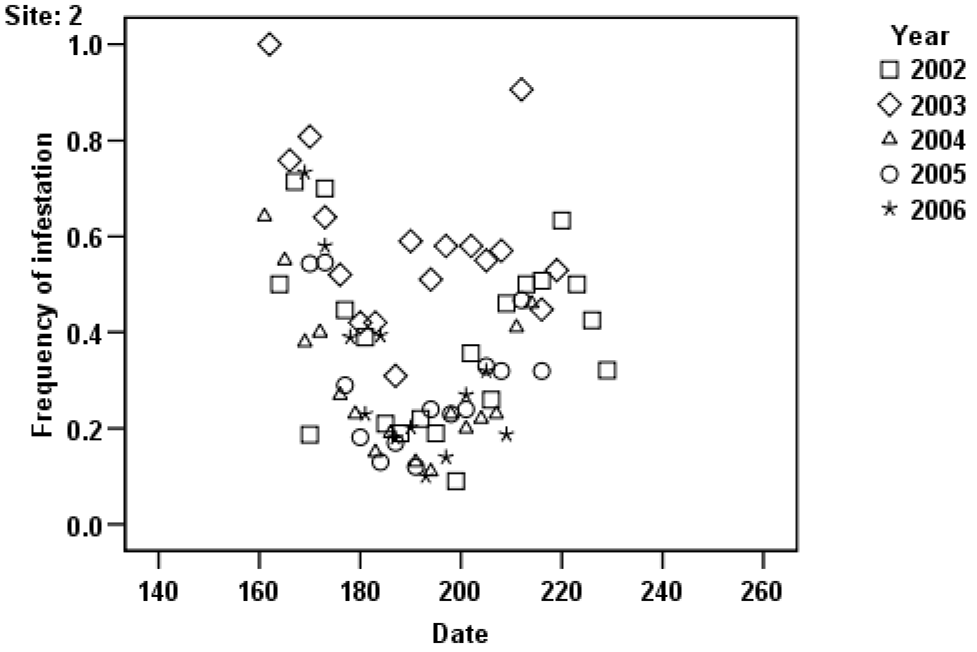


Leucanthemum vulgare* by *Tephritis neesii

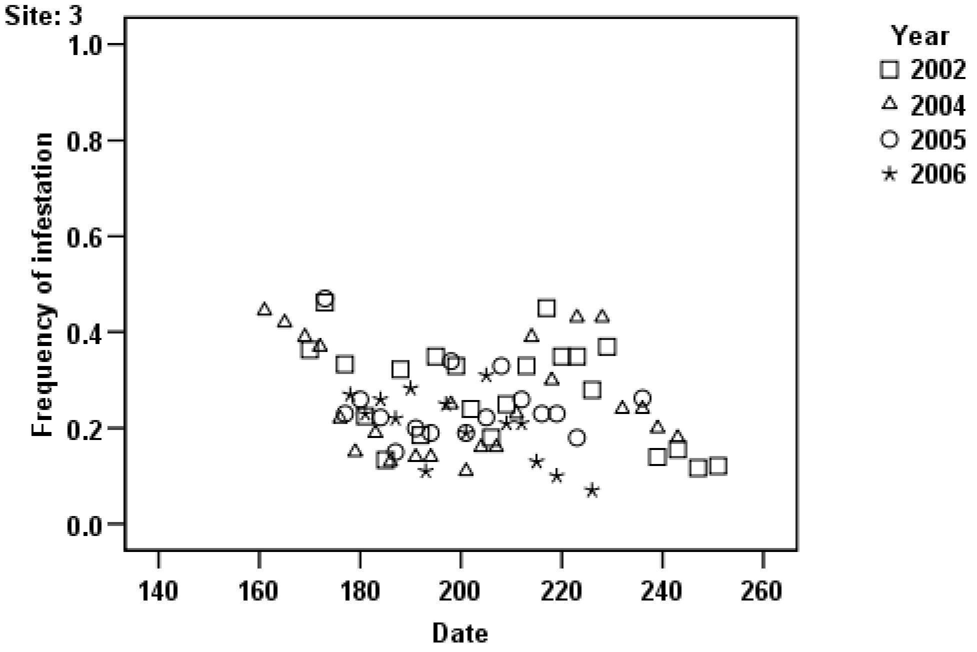


Centaurea nigra* by *Chaetostomella cylindrica

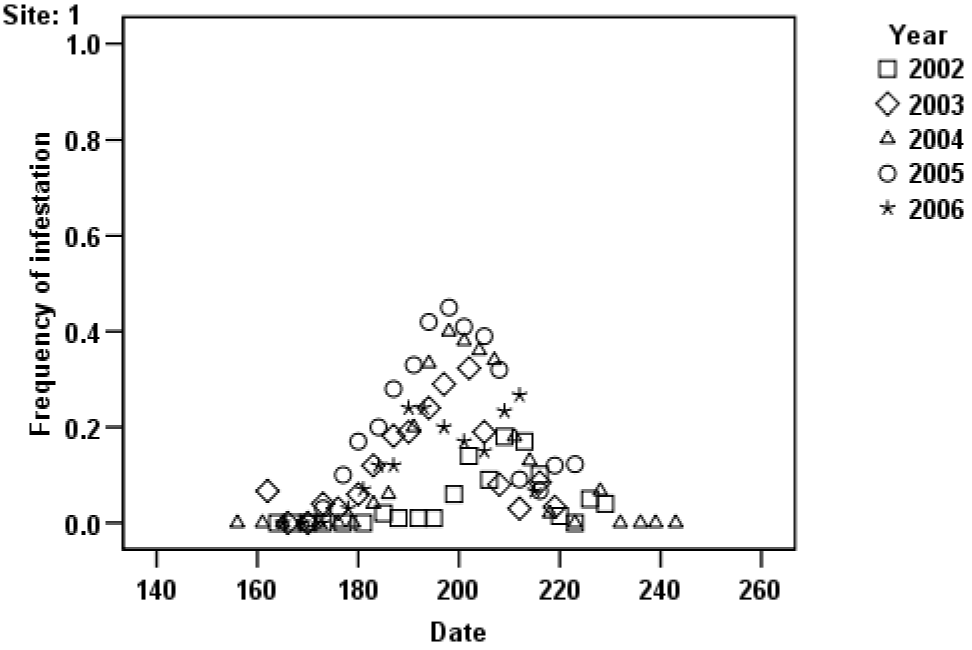


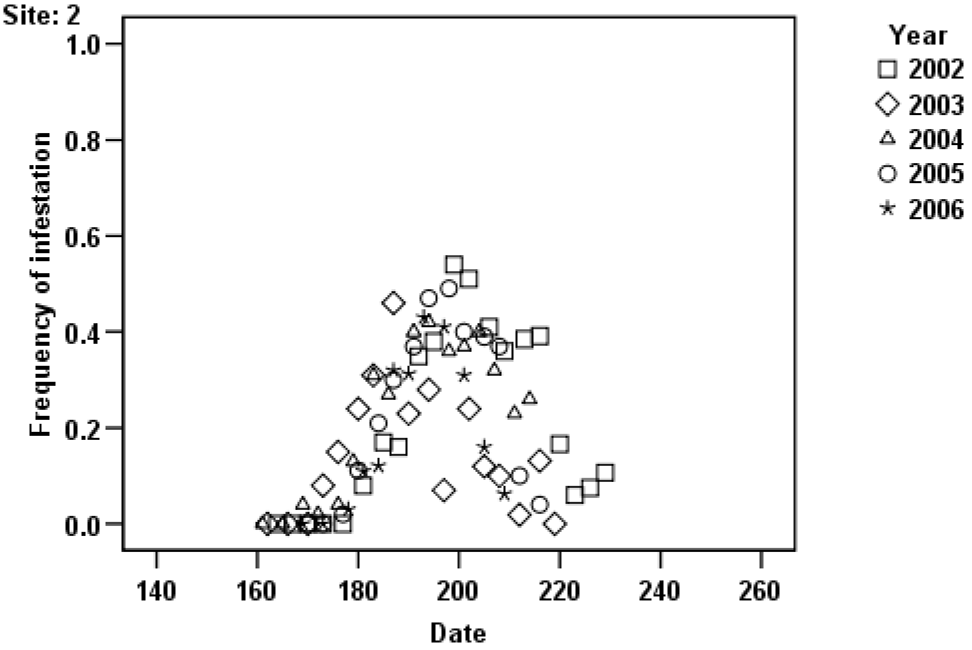


Centaurea nigra by *Chaetostomella cylindrica*

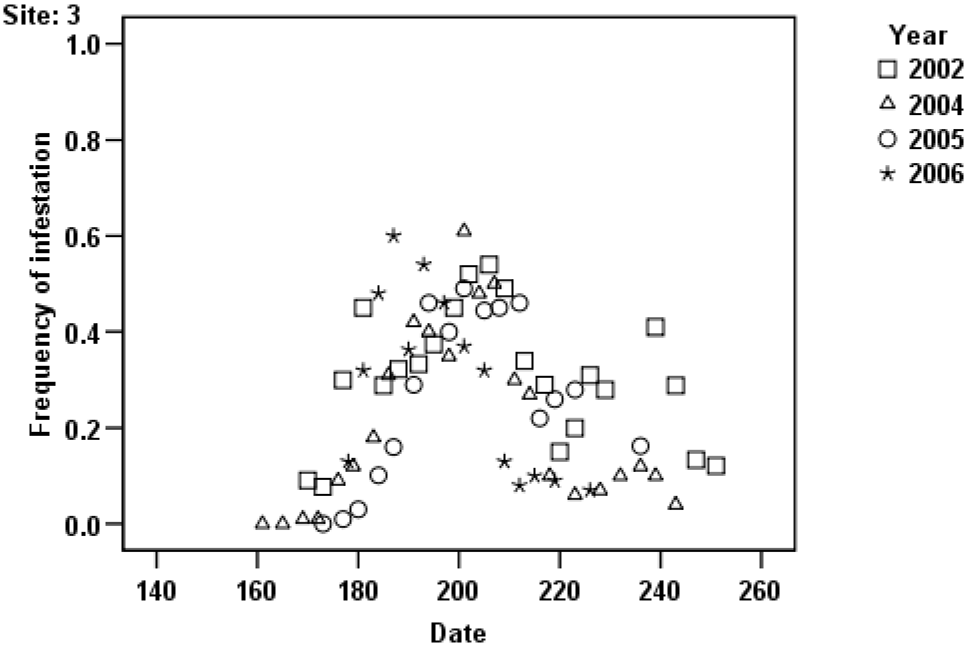


***Centaurea nigra* by *Urophora spp* (gall forming)**





***Centaurea nigra* by *Urophora spp* (gall forming)**



The low infestation rates exhibited in *Cirsium palustre* are evident, and demonstrate well that any selective effect that might apply from competing pressures of predation and pollination is quite specific to the individual interactions between plants and insects. However, as the main thrust of the project was to seek out such interactions as may be significant, *C. palustre* was removed from the project after 2001.

For the remaining species, two types of pattern are generally distinguishable. In the interactions between *Tripleurospermum inodorum* and its non-tephritid predator, and those between *Leucanthemum vulgare* and *Tephritis neesii* as well as *Centaurea nigra* and *Urophora* spp. the relative infestation frequency tends to peaks in mid-season. However, between *Centaurea nigra* and *Chaetostomella cylindrica* the infestation frequency is higher at the start and end of season, and dips in mid-season, although as the number of flower heads available in mid-season is much greater the actual number of attacks is still highest at the flowers phenological maximum.

Looking at total frequency of infestation, however, it is clear that each interaction shows the expected distribution of attack pattern that peaks in mid-season, as shown in Table 18 and Figure 15 below.

Table 19 Mean peak dates of predation

Cirsium palustre

Date

Site		Year 2001
1	N	18
	Mean	183.44
	Median	176.50
	Mode	171
	Std. Deviation	16.169
	Skewness	.740
2	N	23
	Mean	184.30
	Median	182.00
	Mode	171
	Std. Deviation	10.810
	Skewness	.412
3	N	56
	Mean	190.71
	Median	192.00
	Mode	196
	Std. Deviation	14.483
	Skewness	.191

Tripleurospermum inodorum

Date

Site		Year 2002	2003
1	N	1061	274
	Mean	220.48	209.74
	Median	216.00	212.00
	Mode	243	194
	Std. Deviation	22.546	13.298
	Skewness	.303	.195
2	N	1097	455
	Mean	231.18	213.05
	Median	239.00	212.00
	Mode	243	194
	Std. Deviation	26.857	14.461
	Skewness	-.115	.125
3	N	257	398
	Mean	195.16	214.82
	Median	195.00	219.00
	Mode	199	219
	Std. Deviation	4.709	18.181
	Skewness	-.214	-.469

Leucanthemum vulgare - Tephritis neesii

Date

Site		Year 2003	2004	2005	2006
1	N	608	139	287	232
	Mean	165.94	167.06	167.55	160.56
	Median	166.00	165.00	170.00	161.00
	Mode	166	165	170	158
	Std. Deviation	8.675	6.141	8.171	4.808
	Skewness	.189	.351	-.207	.571
2	N	185	63	31	46
	Mean	162.05	161.46	162.35	155.33
	Median	162.00	161.00	159.00	155.00
	Mode	152	165	153	155
	Std. Deviation	10.048	9.612	12.387	5.457
	Skewness	.429	.183	-.181	.779
3	N		75	39	50
	Mean		160.59	158.59	159.66
	Median		161.00	156.00	161.00
	Mode		156	156	161
	Std. Deviation		9.604	9.442	4.826
	Skewness		-.161	.420	.646

Centaurea nigra - Chaetostomella cylindrica

Date

		Year				
Site		**2002**	**2003**	**2004**	**2005**	**2006**
1	N	724	897	609	404	427
	Mean	197.27	190.38	193.19	195.01	185.00
	Median	199.00	190.00	191.00	198.00	184.00
	Mode	216	173[a]	214	173[a]	173
	Std. Deviation	20.552	16.438	23.096	17.211	13.458
	Skewness	-.123	.077	.217	-.079	.450
2	N	456	740	427	334	.091
	Mean	199.47	190.19	185.04	191.40	185.40
	Median	202.00	190.00	179.00	194.00	184.00
	Mode	209	173	165	173	178[a]
	Std. Deviation	17.508	15.130	17.977	14.643	10.825
	Skewness	-.200	-.025	.254	-.020	.369
3	N	440		512	391	284
	Mean	212.22		200.91	198.60	196.54
	Median	213.00		204.00	198.00	197.00
	Mode	217		223[a]	173	205
	Std. Deviation	17.358		24.022	17.833	13.117
	Skewness	-.215		-.106	.219	.277

a. Multiple modes exist. The smallest value is shown

Centaurea nigra - Uropphora quadrafasciata

Date

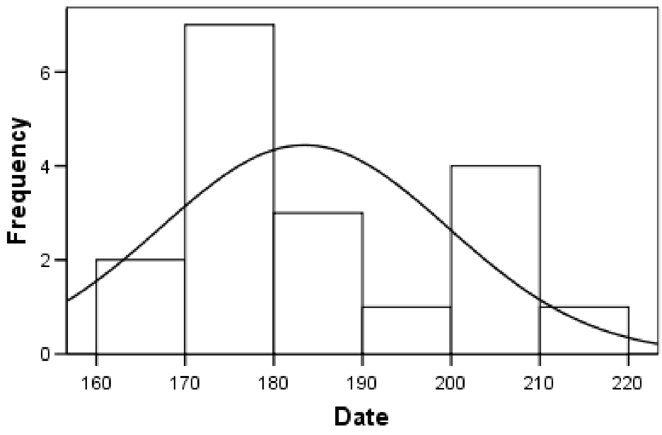
		Year				
Site		**2002**	**2003**	**2004**	**2005**	**2006**
1	N	84	185	245	335	151
	Mean	208.26	195.26	201.08	196.84	195.14
	Median	209.00	197.00	201.00	198.00	193.00
	Mode	209	202	198	198	190[a]
	Std. Deviation	8.258	10.026	7.551	10.364	8.880
	Skewness	-.249	-.433	.191	.066	.281
2	N	366	234	344	320	213
	Mean	201.92	190.09	196.27	196.61	192.78
	Median	202.00	187.00	198.00	198.00	193.00
	Mode	199	187	194	198	193
	Std. Deviation	9.879	10.071	10.158	8.230	6.406
	Skewness	.125	.511	-.309	-.175	-.065
3	N	545		451	418	405
	Mean	211.28		201.52	204.95	194.06
	Median	209.00		201.00	205.00	193.00
	Mode	206		201	201	187
	Std. Deviation	17.693		13.405	11.817	10.662
	Skewness	.271		.566	.385	.835

a. Multiple modes exist. The smallest value is shown

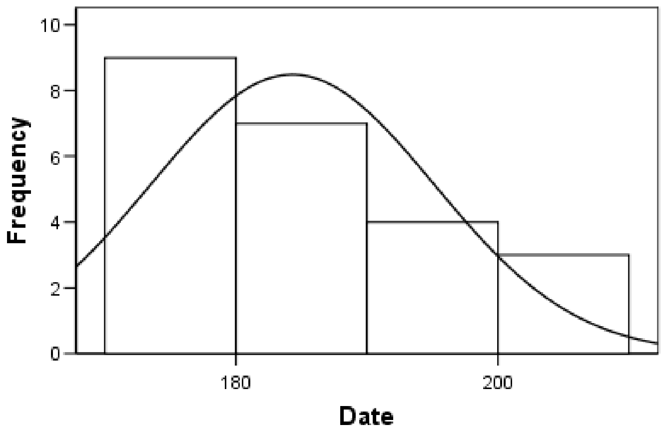
Figure 15 Variation of total infestation level with date, averaged over all years

Cirsium palustre

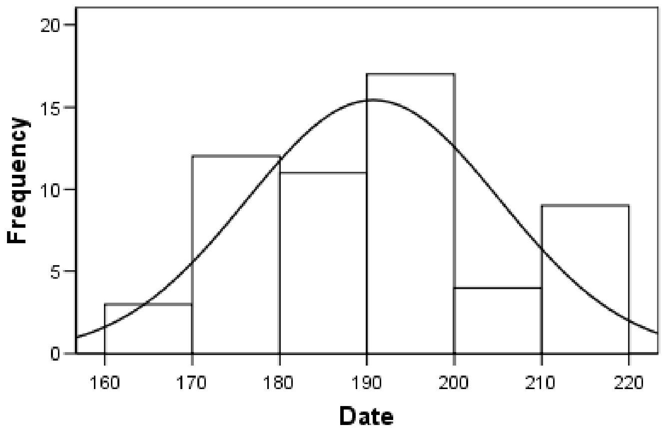
Site: 1



Site: 2

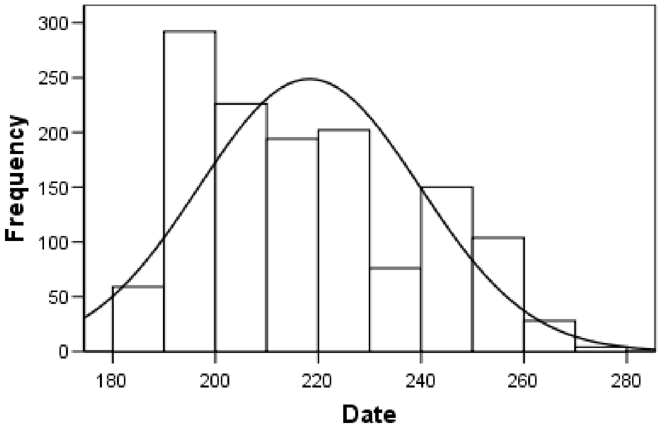


Site: 3



Tripleurospermum inodorum

Site: 1



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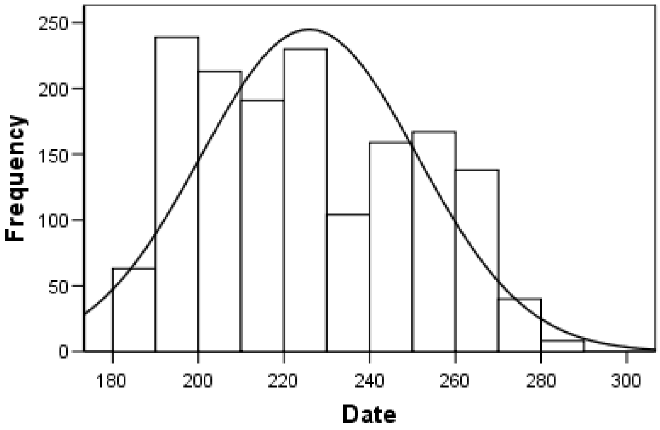
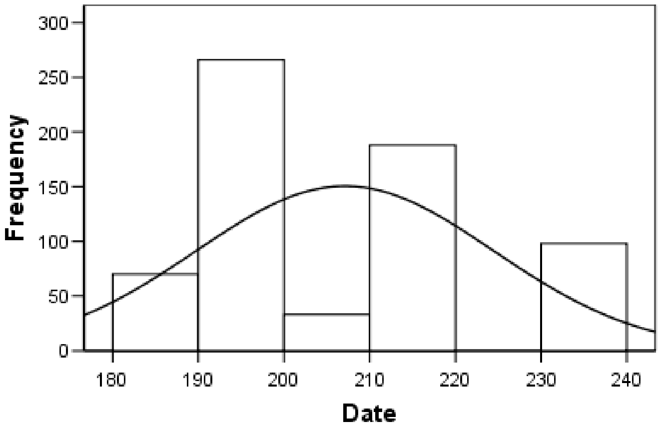
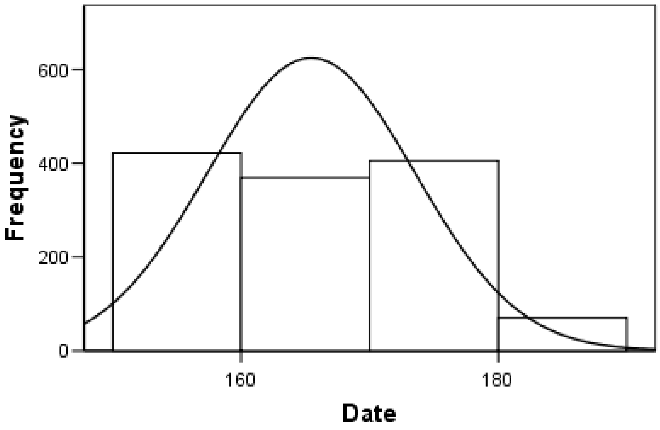


Site: 3

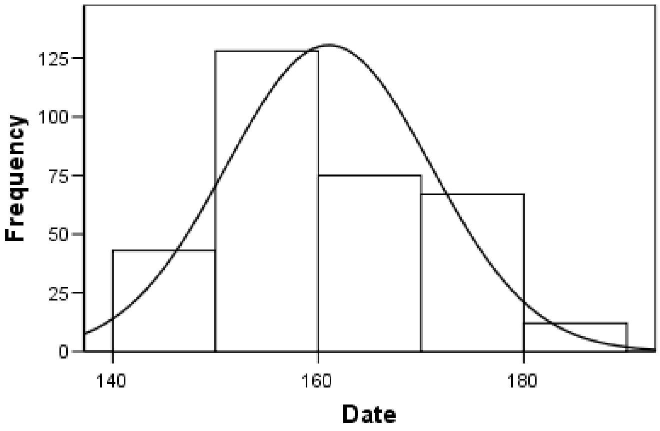


Leucanthemum vulgare – Tephritis neesii

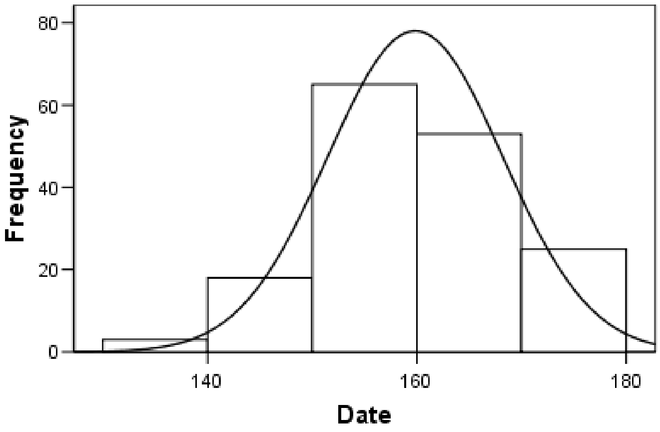
Site: 1



Site: 2

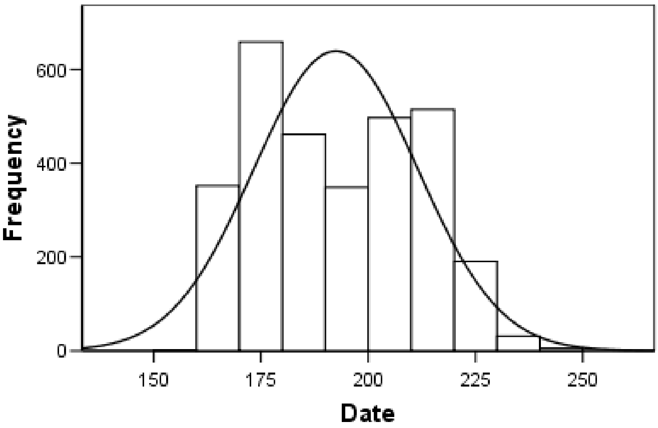


Site: 3

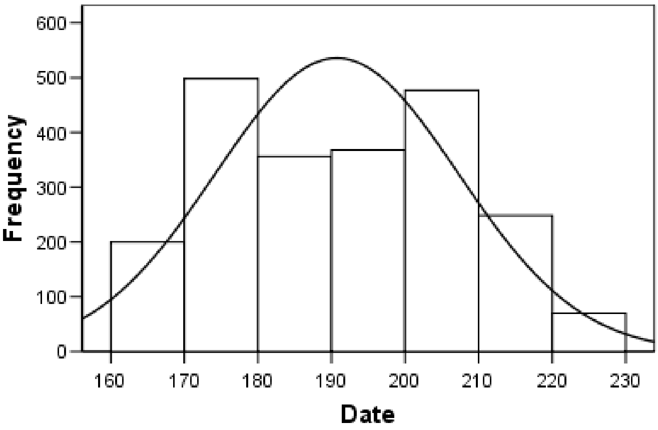


Centaurea nigra – Chaetostomella cylindrica

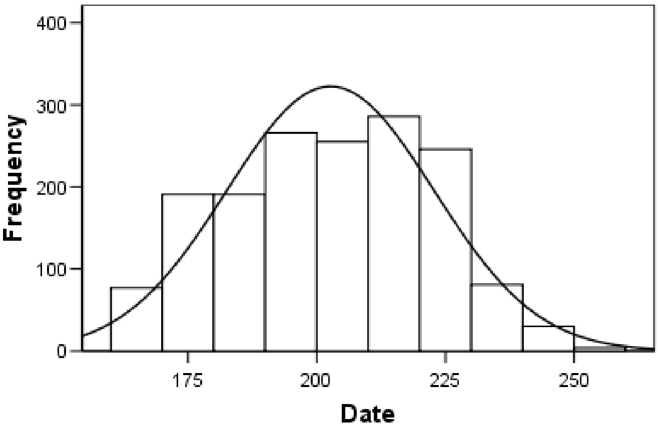
Site: 1



Site: 2

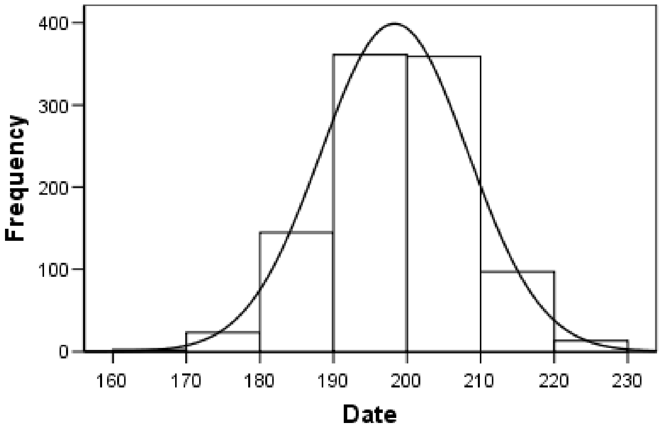


Site: 3

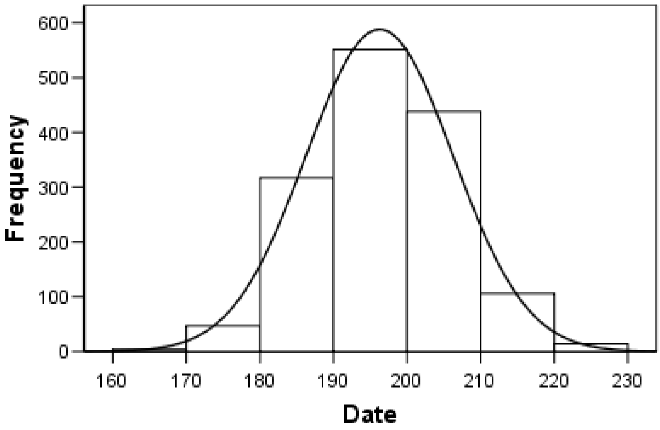


Centaurea nigra – Urophora quadrifasciata

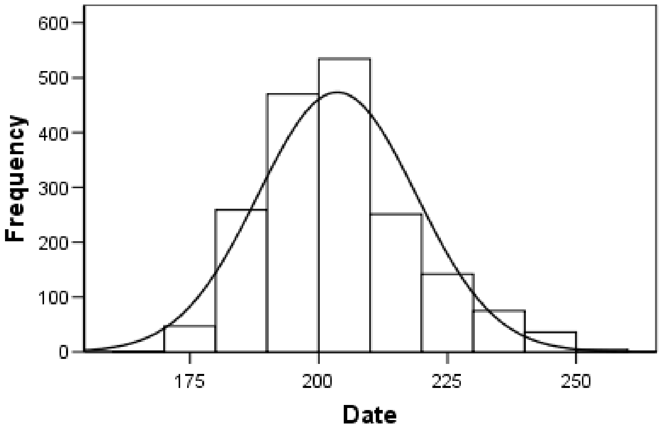
Site: 1



Site: 2



Site: 3



Applying a one-sample Kolmogorov-Smirnov test for normality on the above data results in the following levels of significance for normal distributions.

Table 20 Significance of normality test for daily variation of predation

Site	***C. palustre***	***T. inodorum***	***L. vulgare***	***C. nigra- C. cyl.***	***C. nigra – Urophora***
1	p<0.099	p<1.000	p<0.001	p<1.000	p<0.001
2	p<0.821	p<0.001	p<0.002	p<1.000	p<1.000
3	p<0.325	p<0.001	p<0.004	p<1.000	p<1.000

The patterns of attack within each season also follow a relatively consistent, if not generally normally distributed pattern, based upon the particular plant/predator interaction. For both *Tephritis neesii* and *Urophora* spp the relative frequency of infestation follow a path closely linked with the phenology of *Leucanthemum vulgare* and *Centaurea nigra*. This would indicate a close relationship between predator and host.

The pattern for *Chaetostomella cylindrica*, however, is less closely linked, as Figure 14 shows, as the relative frequency of attack is higher at the beginning and end of the season. White (1988) describes *Chaetostomella* as oligophagous, feeding on a range of Asteraceae seed heads, whilst both *Tephritis neesii* and *Urophora* spp. are monophagous. The specificity of these interactions is likely to lead to a greater likelihood of their forming selective pressures on their hosts, and this should manifest itself in a greater correlation of predator choice with flower trait.

It should be noted, however, that for *Chaetostomella* despite the relative frequency of predation being at its lowest in mid season, the absolute number of attacks still peaks in mid-season. This is not unexpected, as both *Leucanthemum* and *Centaurea* phenologies are typical of the Asteraceae and *Chaetostomella* can be seen as having a specific relationship with the family rather than the individual species.

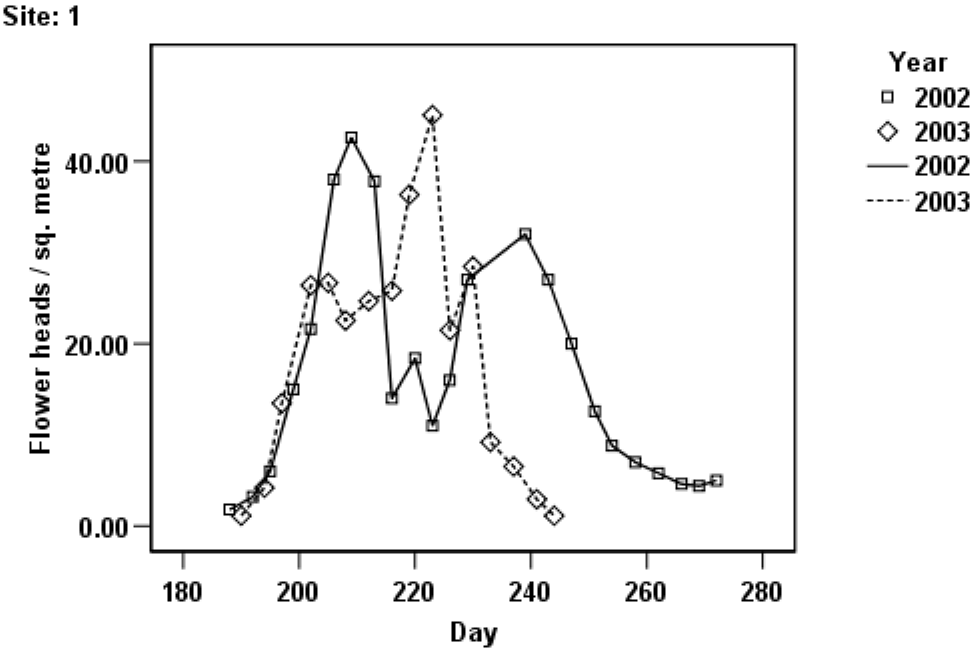
The overall hypothesis that the pattern of seed predation over the course of a season is repeatable over several seasons is not justified, although by definition it is a seasonal pattern. This may be because, in order to achieve a level of significance in the data, many more sets of data are required, however, as each set requires additional seasons to be sampled this is impractical. Therefore, any modelling of the interactions must include the high levels of variation found within these data as an important variable.

6.3 Phenology of the floral populations

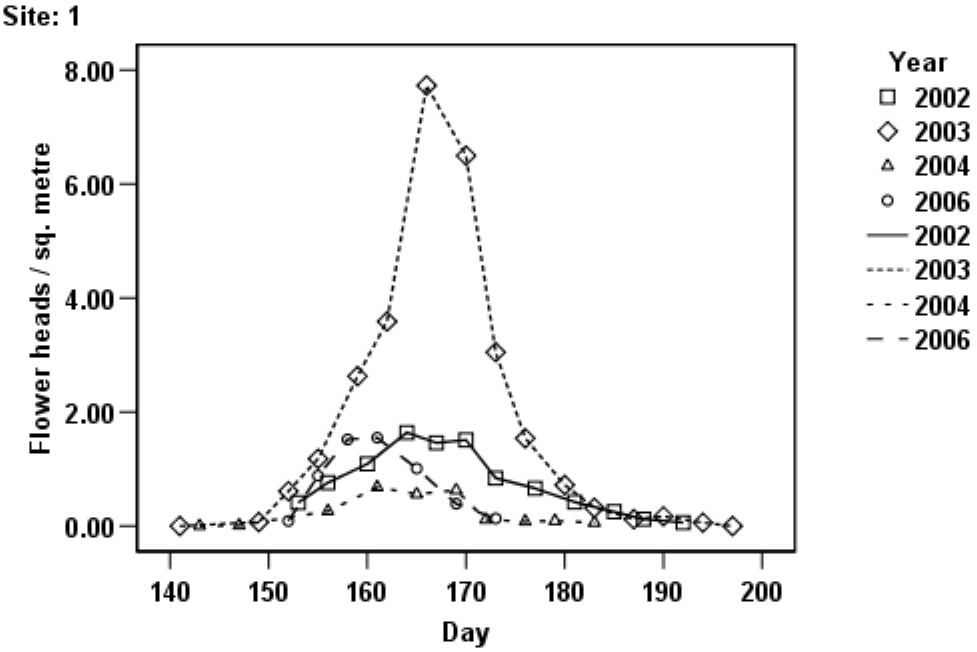
Overall population phenology is plotted in terms of available flower heads per square metre from the sites where continuous phenology data was recorded, in Figure 16. As expected the data tend to follow a peaked distribution.

Figure 16 Flowering phenology

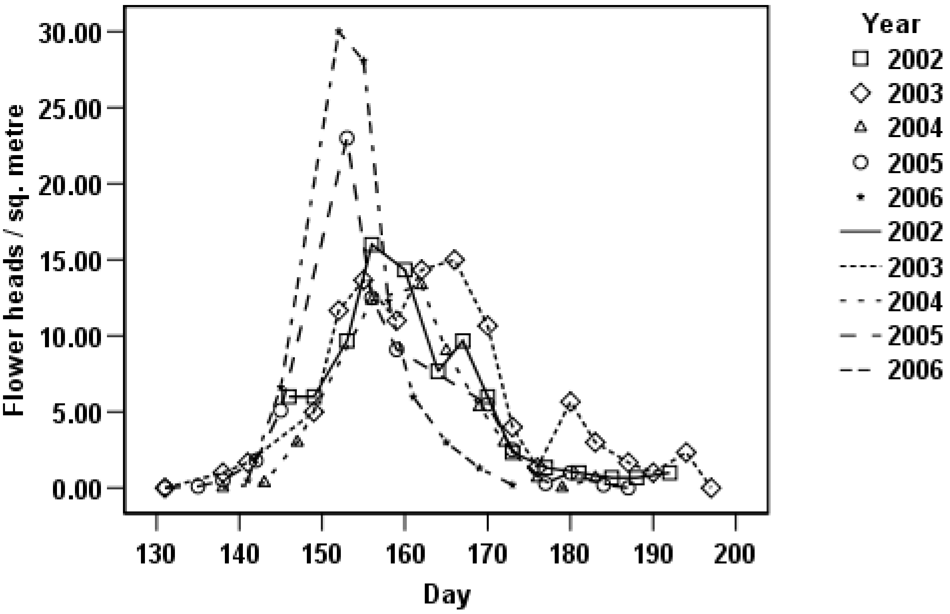
Tripleurospermum inodorum



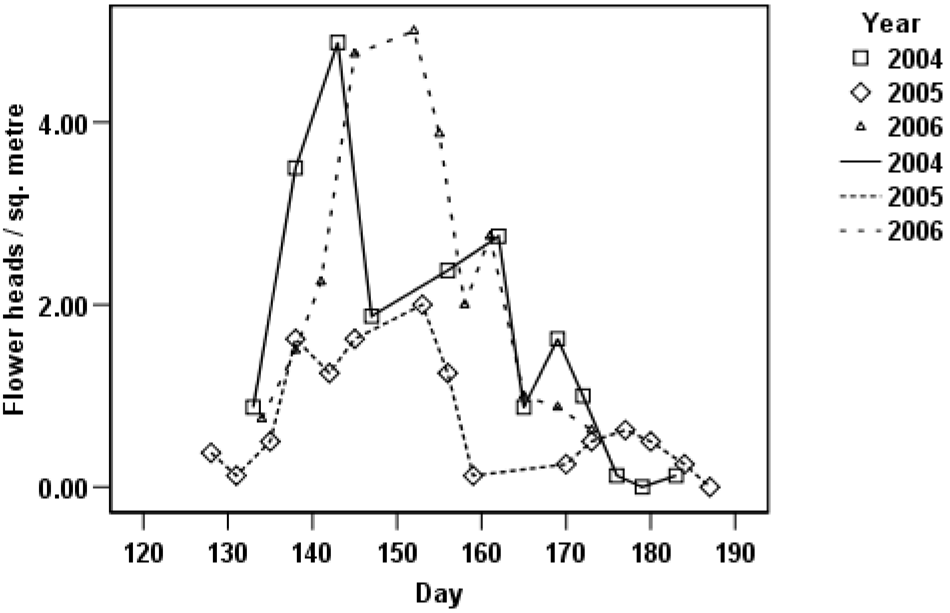
Leucanthemum vulgare



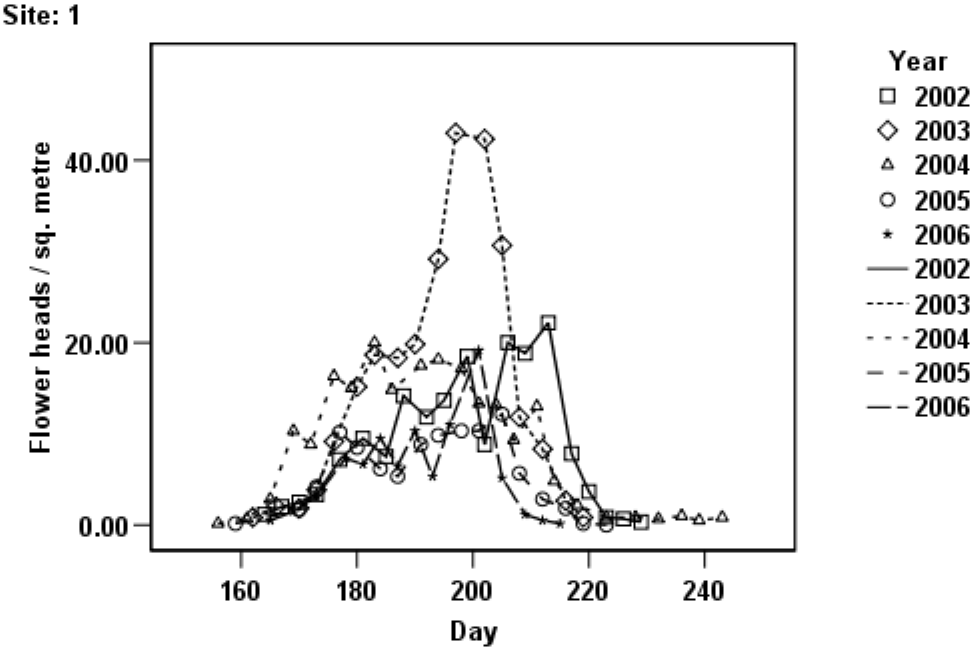
Site: 2

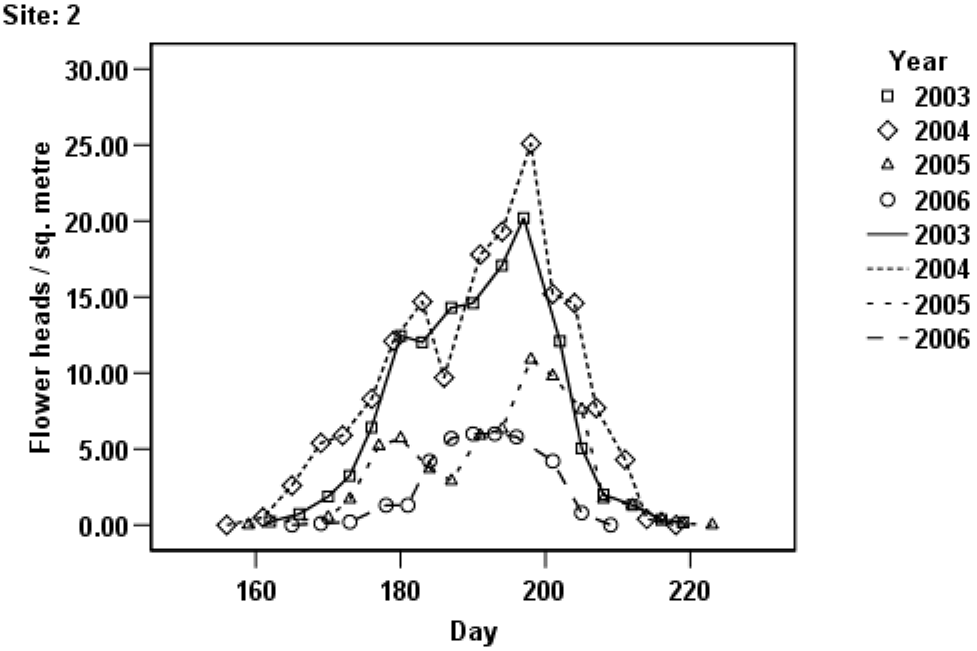

Flower heads / sq. metre
30.00
25.00
20.00
15.00
10.00
5.00
0.00
130
140
150
160
170
180
190
200
Day
Year
□ 2002
◇ 2003
△ 2004
○ 2005
* 2006
2002
2003
2004
2005
2006

Site: 3


Flower heads / sq. metre
4.00
2.00
0.00
120
130
140
150
160
170
180
190
Day
Year
□ 2004
◇ 2005
△ 2006
2004
2005
2006

Centaurea nigra





Applying a one-sample Kolmogorov-Smirnov test for normality on the above data results in the following levels of significance for normal distributions.

Table 21 Significance of normality test for plant phenology

Year	*T.inodorum* Site 1	*L.vulgare* Site 1	*L. vulgare* Site 2	*L.vulgare* Site 3	*C. nigra* Site 1	*C. nigra* Site 2
2002	p<1.000	p<0.001	p<0.001	-	p<1.000	-
2003	p<1.000	p<0.001	p<0.001	-	p<0.001	p<1.000
2004	-	p<0.001	p<0.001	p<0.001	p<0.001	p<1.000
2005	-	-	p<0.001	p<0.009	p<0.001	p<0.001
2006	-	p<0.001	p<0.001	p<0.002	p<0.001	P<0.006

As with the patterns of attack, the population phenologies of the sampled species cannot all be taken to be normally distributed, although by their nature they are seasonal, and peak in mid-season. *T.inodorum* does exhibit a normal distribution in all sampled periods, whilst *L.vulgare* and *C.nigra* do not.

6.4 Individual floral phenology

In order to create a mathematical model of the host-predator interactions that behaves like the sampled populations, it is necessary not just to model the population phenology, but also the phenologies of individual flower heads. This was undertaken in order to attribute realistic values to the availability of flower heads for both pollination and predation.

Investigation of individual flower head phenology showed that it takes on average ten days between the first opening of florets at the edge of the disc and the floret face becoming completely browned in *Tripleurospermum inodorum*. Similar data for *Leucanthemum vulgare* was between 14 and 21 days.

Centaurea nigra flowers show a different pattern, with the heads opening and displaying florets, and then closing again and beginning to dry after three or four days.

The phenological data gathered show a consistent flowering season for the individual populations, with small variations no doubt related to environmental differences year on year. All of the plant species investigated are perennial herbs. And there is evidence from Halley & Dempster (1996) that *Centaurea nigra* patches remain stable over several years.

On an individual basis, the major difference exhibited by the species is the length of individual flower head phenology, which is relatively short, three – four days, for all species except *Leucanthemum vulgare* where flowers remain open for ten to twenty days. Straw (1989b) shows that Tephritid attack on *Arctium minus* occurs at the flower bud stage, prior to opening, and suggests that this is a general characteristic of Tephritids in (1989a). So the individual phenology may not alter the infestation rate. However, several other orders and families of insect also predate these flowers, which may account for the loss of viable seed in heads not infested by the species under investigation.

Chapter 7. Mathematical modelling

The final hypotheses required to demonstrate an evolutionary effect of pre-dispersal seed predation on floral traits were set as follows:

> The combined effect of selective infestation of flower heads, and reduction in fecundity, and correlation of flower size and seed set, leads to an evolutionary pressure that selects for an optimum flower size in subsequent generations.
>
> The combined effect of the pattern of predation and reduction in fecundity leads to an evolutionary pressure that selects for phenological patterns in subsequent generations.

Both hypotheses require evidence of the effects of evolutionary pressures. Direct observation of such effects in organisms with life spans measured in years is clearly not a practical proposition, even in a research project of seven years duration, so some method other than direct observation is required in order to investigate the hypotheses.

To this end, a mathematical model of floral evolution has been developed that follows populations of plants over the course of a number of generations, monitoring both the mean flower size and flowering season when under pre-dispersal seed predation of adjustable strength. Despite the obvious objection that any mathematical model can only approximate some of the many different aspects of the organisms' life cycles, and also that it can only be as relevant as its underlying assumptions, it does at least, given sufficiently robust basic methodology, offer the possibility of investigating long term trends.

The basis of this model is to allow for various stochastic processes by generating populations with variance in several traits that comply with a normal distribution, and to allow subsequent generations a random variation within similar parameters.

7.1 Description of the model

The overall concept of the model is to create typical floral populations, with characteristics set by data obtained in the field, and to apply annual treatments of both pollination and predation, and monitor the effects of these on the traits of subsequent generations.

The model initially creates a population from the following set of constraints:

- Mean population size and variance
- Mean floral life span and variance
- Flowers per plant and variance
- Mean flower size and variance
- Mean floret diameter, determined from field data
- Viability of seed bank and variance
- Peak date of phenology and variance
- Length of individual plant fertility
- Annual variance of inherited flower size and phenology and flowers per plant

This population is allowed to follow a number of growing seasons during which both pollination success and predation probability are applied to it, on the basis of either random or variable probability based upon flower size. The population recruits from the resulting seed bank on a random basis. Both size and phenological traits are assumed to be heritable.

The model was written as a computer program in the Delphi programming environment, utilising the Pascal programming language and the Windows operating system.

7.2 Model assumptions

In order to model the evolutionary effects, a number of simplifying assumptions are made.

1. Where parameters are set with mean and variance the population assumes a normal distribution pattern.

2. There is a basic constraint on attainable flower size which, although arbitrarily set, models some aspects of resource cost due to larger flower heads.

3. Heritability of size or phenology is assumed to take the form either of a fully paternal, or fully maternal inheritance, or of a mean value of the parental plants. No attempt has been made to attribute Mendelian characteristics.

4. Pollination visits are assumed to occur to maternal plants, pollen is allotted randomly from the existing population.

5. All plants are pollinated by external insect visits, no self-pollination occurs.

6. Recruitment is a random event that maintains the population size within the mean and variance set for it initially.

7. There are no specific ecological pressures on the population other than those implied by the variance in allowable population size.

7.3 Model process

The model follows an iterative process, where each year receives a similar treatment.

1. From the initial set of parameters, set in the program, a normal distribution of flowers is generated.

2. Each plant is set to have a natural phenology (normally distributed within the population), a number of flower heads (normally distributed in number) and each flower head a number of potential seed (normally distributed in number). Each seed contains information regarding the plant's mean flower size and phenology, as well as their variance.

3. Predation is then applied to the population according to a set of parameters set within the model. Plants become available for predation throughout the full length of each plant's phonological season. Predation can occur either randomly with date, at a fixed level of the available population percentage, or follow a variable percentage of available population depending upon a correlation equation based upon the date.

4. Where an equation related to date is used, the peak predation date is set to equal the floral phenology maximum. In subsequent generations predation can be set either to keep this peak date, or automatically to track the phenological maximum of the population.

5. Within each flower head, predation can either occur randomly at a predetermined percentage of potential seeds per head, or at a frequency distribution that varies with flower head size.

6. Oviposition visits can also be set to be independent of flower head size or to vary in proportion to flower size, following a variable equation entered into the model.

7. Pollination is then applied to the population according to a set of parameters set within the model. Plants become available for pollination throughout the full length of each plant's phonological season. Pollination can occur either randomly with date, at a fixed level of the available population percentage, or follow a variable percentage of available population depending upon a correlation equation based upon the date.

8. Within each flower head, pollination can either occur randomly at a predetermined percentage of potential seeds per head, or at a frequency distribution that varies with flower head size.

9. Pollination visits can also be set to be independent of flower head size or to vary in proportion to flower size, following a variable equation entered into the model.

10. Seeds that are pollinated are designated with mean flower size (and variance) and mean phenological date (and variance) based upon the average of the parental traits.

11. Both pollination and predation can be set to have a probability dependant upon a linear equation that relates probability of visit or oviposition to relative flower size by means of an inbuilt equation parser, and a parametric decision maker.

12. Seeds that are pollinated enter the seed bank, and are designated a viability set in the program.

13. At the end of the season, plants increase their age, and are killed if their lifetime has been reached.

14. At the beginning of the next season a new population size is randomly allocated based upon the mean and variance set in the program. Recruitment is randomly selected from the available seed bank.

15. When generating a new generation, an additional set of variation on inherited flower size and phenological peak can be applied in order to model basic ecological variation.

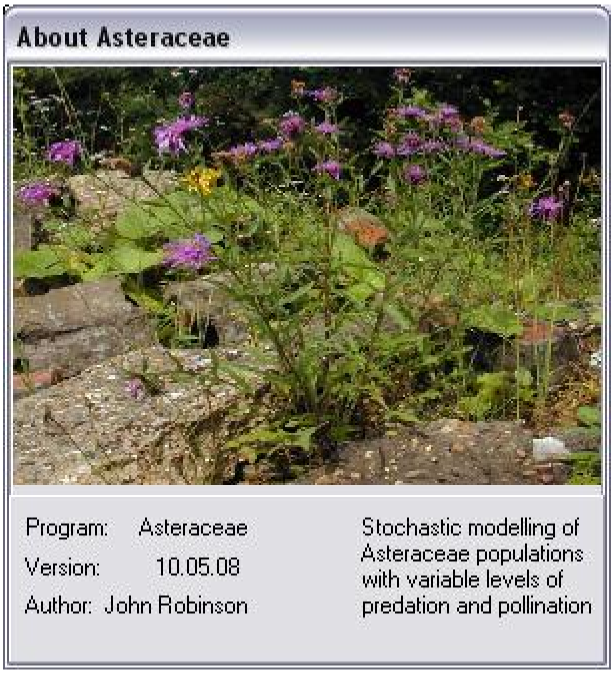
16. The seeds in the bank increase their age, and those at the end of their viable life removed.

17. The new plants are randomly allocated a number of flower heads and seeds as in step 2 but based upon inherited characteristics as appropriate.

18. The process repeats for a set number of generations.

7.4 Computer program

The model was coded into a program to run in the Windows operating system. The program consists in a set of pages that define the basic parameters described above, and allows the operator to alter them as required.

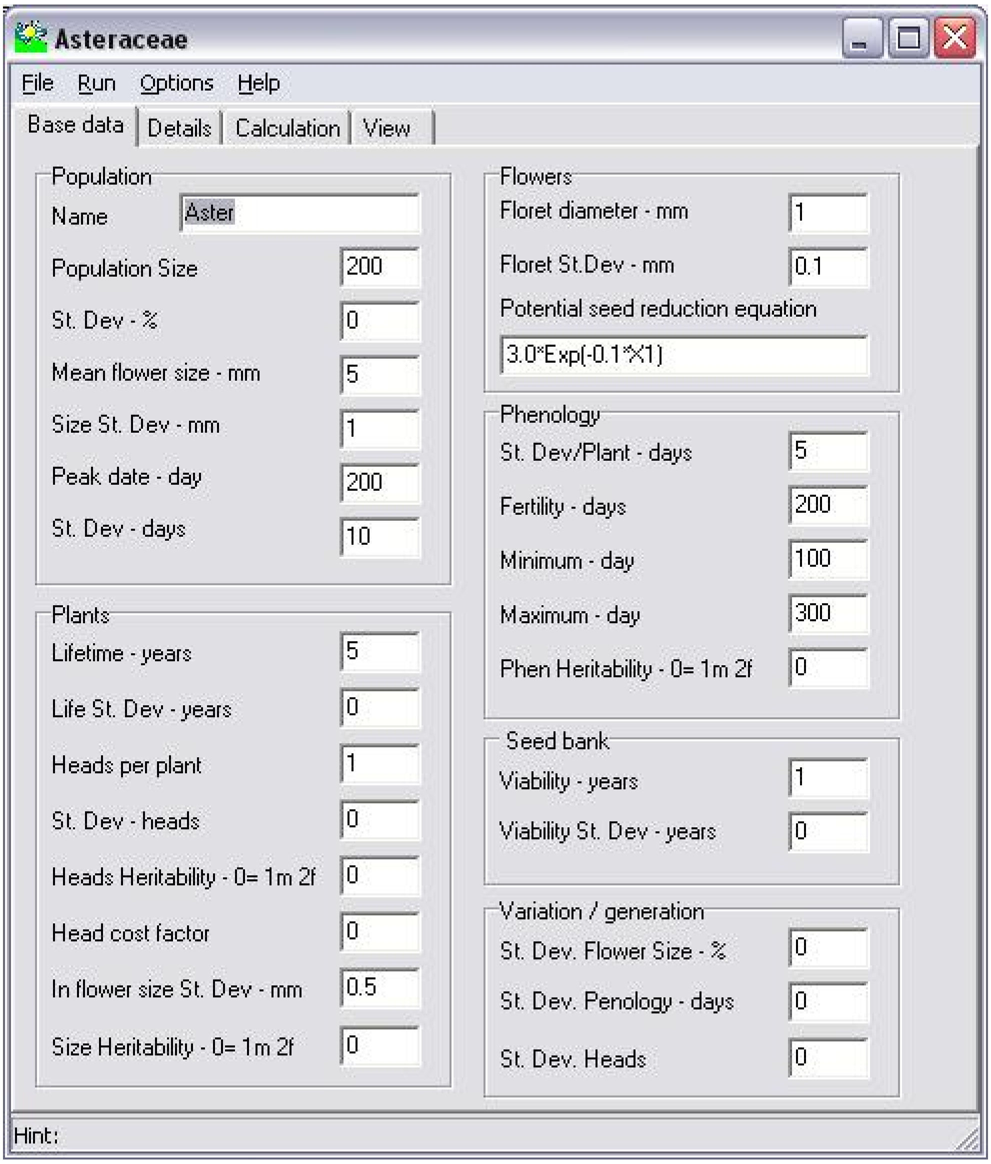


About Asteraceae

Program: Asteraceae
Version: 10.05.08
Author: John Robinson

Stochastic modelling of Asteraceae populations with variable levels of predation and pollination

The main page sets the basic parameters of the population, the individual plants, the individual flowers, phenology, seed bank and annual variations.



Population
Name: Aster
Population Size: 200
St. Dev - %: 0
Mean flower size - mm: 5
Size St. Dev - mm: 1
Peak date - day: 200
St. Dev - days: 10

Plants
Lifetime - years: 5
Life St. Dev - years: 0
Heads per plant: 1
St. Dev - heads: 0
Heads Heritability - 0= 1m 2f: 0
Head cost factor: 0
In flower size St. Dev - mm: 0.5
Size Heritability - 0= 1m 2f: 0

Flowers
Floret diameter - mm: 1
Floret St.Dev - mm: 0.1
Potential seed reduction equation: 3.0*Exp(-0.1*X1)

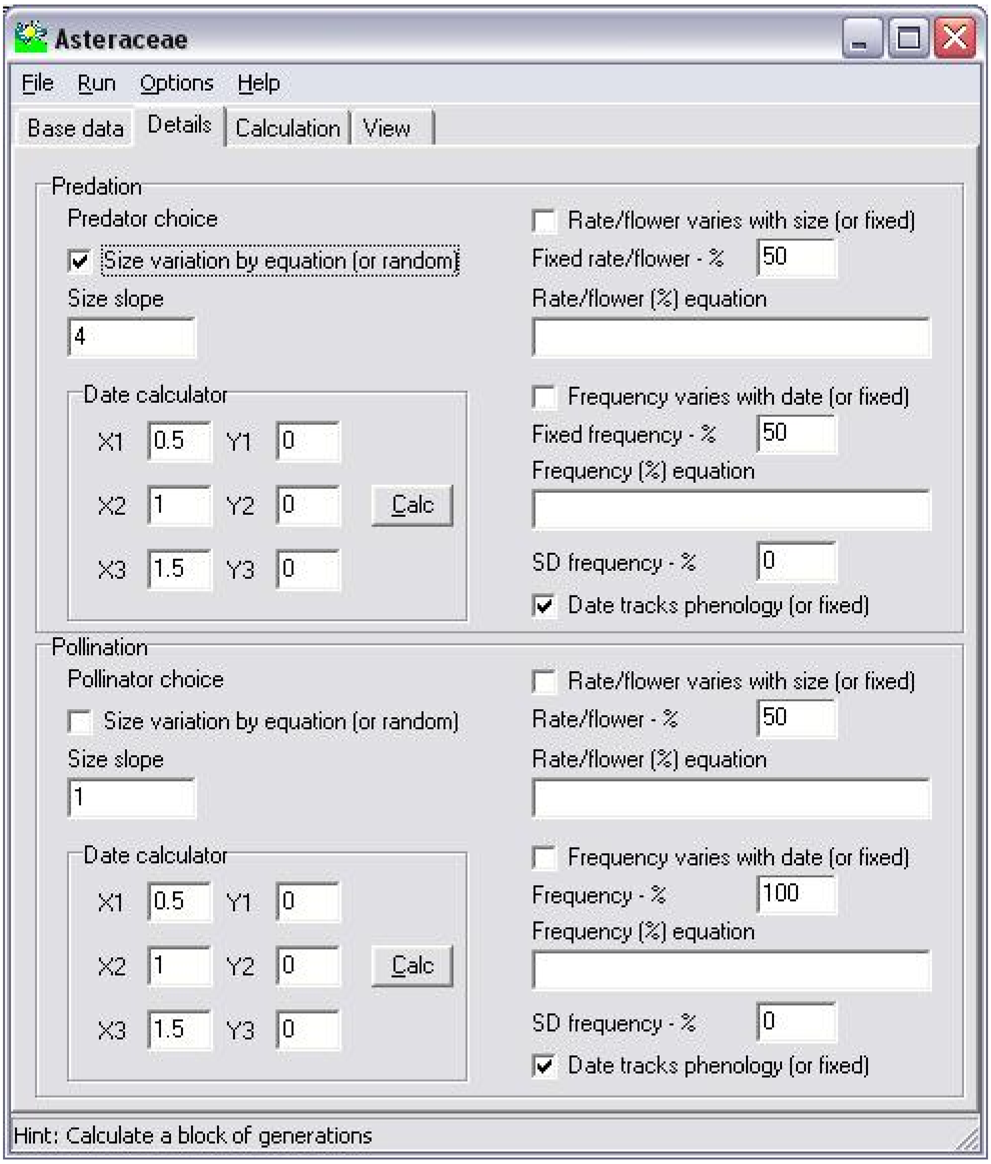
Phenology
St. Dev/Plant - days: 5
Fertility - days: 200
Minimum - day: 100
Maximum - day: 300
Phen Heritability - 0= 1m 2f: 0

Seed bank
Viability - years: 1
Viability St. Dev - years: 0

Variation / generation
St. Dev. Flower Size - %: 0
St. Dev. Penology - days: 0
St. Dev. Heads: 0

The second page allows details of the predation and pollination effects that will be applied to the population.

Each can be set to be applied either randomly, or with an association strength applied, in the same manner as was determined from the field studies as described in section 4.2. The rate of predation or pollination within the individual flower heads can be fixed to a set percentage, or varied with flower size. The overall frequency of predation or pollination within the season can be set at a fixed rate, or set to vary with date over the course of each season, and can also have its own annual variation set.



Predation

Predator choice

☑ Size variation by equation (or random)

Size slope: 4

Date calculator

X1: 0.5 Y1: 0

X2: 1 Y2: 0

X3: 1.5 Y3: 0

☐ Rate/flower varies with size (or fixed)

Fixed rate/flower - %: 50

Rate/flower (%) equation:

☐ Frequency varies with date (or fixed)

Fixed frequency - %: 50

Frequency (%) equation:

SD frequency - %: 0

☑ Date tracks phenology (or fixed)

Pollination

Pollinator choice

☐ Size variation by equation (or random)

Size slope: 1

Date calculator

X1: 0.5 Y1: 0

X2: 1 Y2: 0

X3: 1.5 Y3: 0

☐ Rate/flower varies with size (or fixed)

Rate/flower - %: 50

Rate/flower (%) equation:

☐ Frequency varies with date (or fixed)

Frequency - %: 100

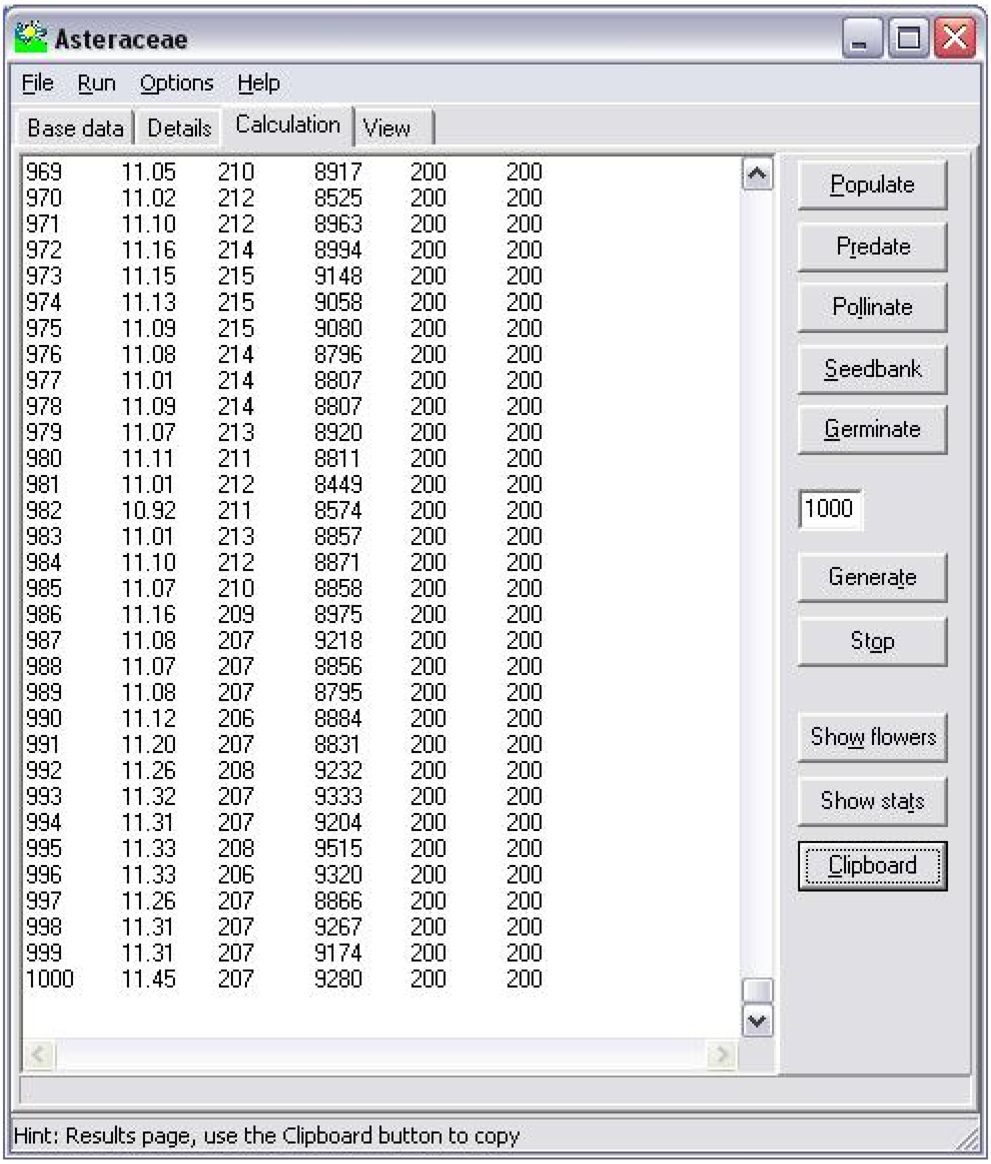
Frequency (%) equation:

SD frequency - %: 0

☑ Date tracks phenology (or fixed)

Hint: Calculate a block of generations

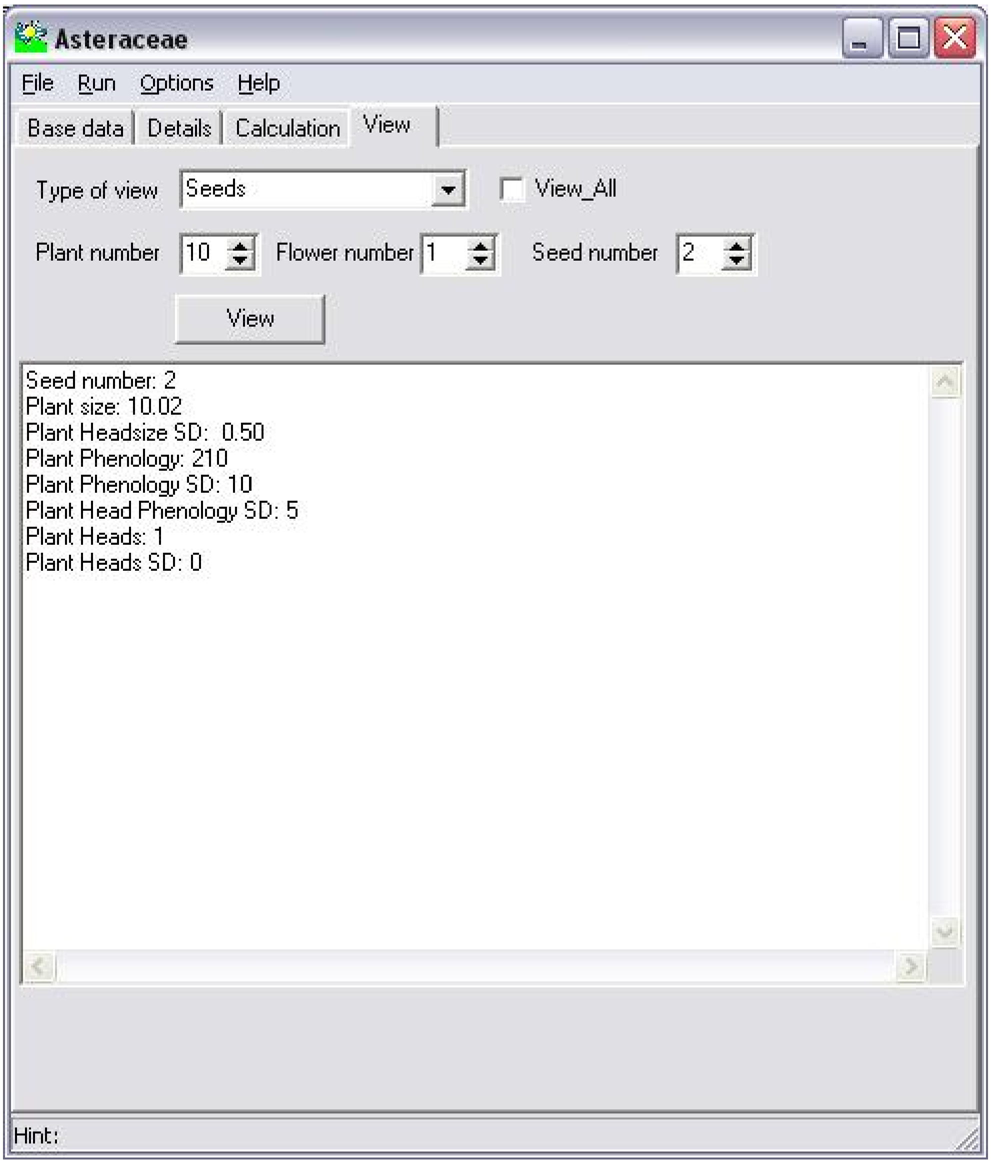
Calculations may then be run, and results are displayed on the calculation page. In order to test the program and the reliability of the algorithms, each action can be applied separately, such as creating a population, and applying predation, a full list of the population characteristics can then be displayed and checked for compliance with the program design. Once the model had been checked, it was possible to carry out the automated process of predation, pollination, seed setting, and germination over a number of cycles, using the generate option. The list of population characteristics thus generated was able to be copied into other programs for further analysis.



```
969     11.05   210     8917    200     200
970     11.02   212     8525    200     200
971     11.10   212     8963    200     200
972     11.16   214     8994    200     200
973     11.15   215     9148    200     200
974     11.13   215     9058    200     200
975     11.09   215     9080    200     200
976     11.08   214     8796    200     200
977     11.01   214     8807    200     200
978     11.09   214     8807    200     200
979     11.07   213     8920    200     200
980     11.11   211     8811    200     200
981     11.01   212     8449    200     200
982     10.92   211     8574    200     200
983     11.01   213     8857    200     200
984     11.10   212     8871    200     200
985     11.07   210     8858    200     200
986     11.16   209     8975    200     200
987     11.08   207     9218    200     200
988     11.07   207     8856    200     200
989     11.08   207     8795    200     200
990     11.12   206     8884    200     200
991     11.20   207     8831    200     200
992     11.26   208     9232    200     200
993     11.32   207     9333    200     200
994     11.31   207     9204    200     200
995     11.33   208     9515    200     200
996     11.33   206     9320    200     200
997     11.26   207     8866    200     200
998     11.31   207     9267    200     200
999     11.31   207     9174    200     200
1000    11.45   207     9280    200     200
```

Hint: Results page, use the Clipboard button to copy

The final page allows for checking program operation in detail by allowing for details of individual plats, flower heads or seeds to be viewed



Type of view: Seeds

☐ View_All

Plant number: 10

Flower number: 1

Seed number: 2

```
Seed number: 2
Plant size: 10.02
Plant Headsize SD:  0.50
Plant Phenology: 210
Plant Phenology SD: 10
Plant Head Phenology SD: 5
Plant Heads: 1
Plant Heads SD: 0
```

A full listing of the program code is supplied in the Appendix.

7.5 Program testing

Program testing initially required a detailed analysis of variables within the program itself, using the built in debugging facilities of the Delphi programming environment, which allow individual program variables to be tracked as the program steps through its individual lines of code.

Once the basic operating structure was debugged, the main task was to validate the predation and pollination selection procedure that allows selection of flower heads to be made on a statistical basis that allows for progressive probability of selection by means of an equation related to the flower size. Although the initial process worked well, in that it provided a progressive selection probability, it did need tuning to achieve the required rate of change of probability with relative flower size. This was achieved with a calibration function applied to the required slope of the curve, which proved robust under a range of rates, and allowed flower selection to mimic what had been recorded in the field.

Having verified the functions within the program, the next phase of testing was required to determine the stability of the process based upon the size of the population of flowers. Too small a population could lead to extinction, which, although a reality in the field, would not allow long term trends to be determined. A minimum population size of fifty individual plants appeared to be viable within the model, although the results published below use a population of 200.

Finally, repeatability of results was tested, by repeating runs with the same variable combinations several times. The stochastic nature of the program, where distributions, and levels of predation and pollination are subject to random variation during the course of each run meant that several runs in each condition were necessary.

7.6 Results from the model

7.6.1 General effects of the parameters

The model was run to investigate the general effects of several of its parameters on a non-specific Aster, with a characteristic mean flower size of 5mm. The basic parameter values set for each variable, other than that under investigation were as follows:

Population size	200	Lifetime: Annual	Seedbank: Annual
Flower size	Mean 5mm	St.Dev. 1mm	Floret size 1mm
Pollination	100% of flowers	50% of florets/flower	No association slope
Predation	50% of flowers	50% of florets/flower	4.0 association slope
Generations	1000		

The effect of association slopes of both predation and pollination with flower size were initially investigated and the results are shown in Figures 17 and 18. For the pollination case, the overall level of visitation was set at 50%.

Figure 17 Variation of flower size under different predation association slopes

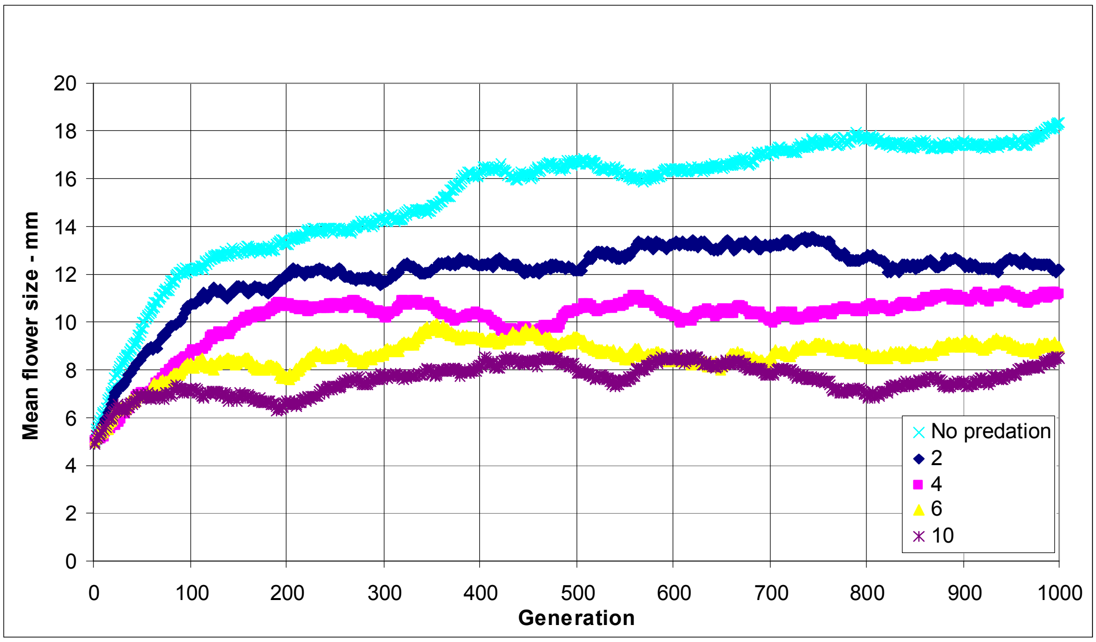
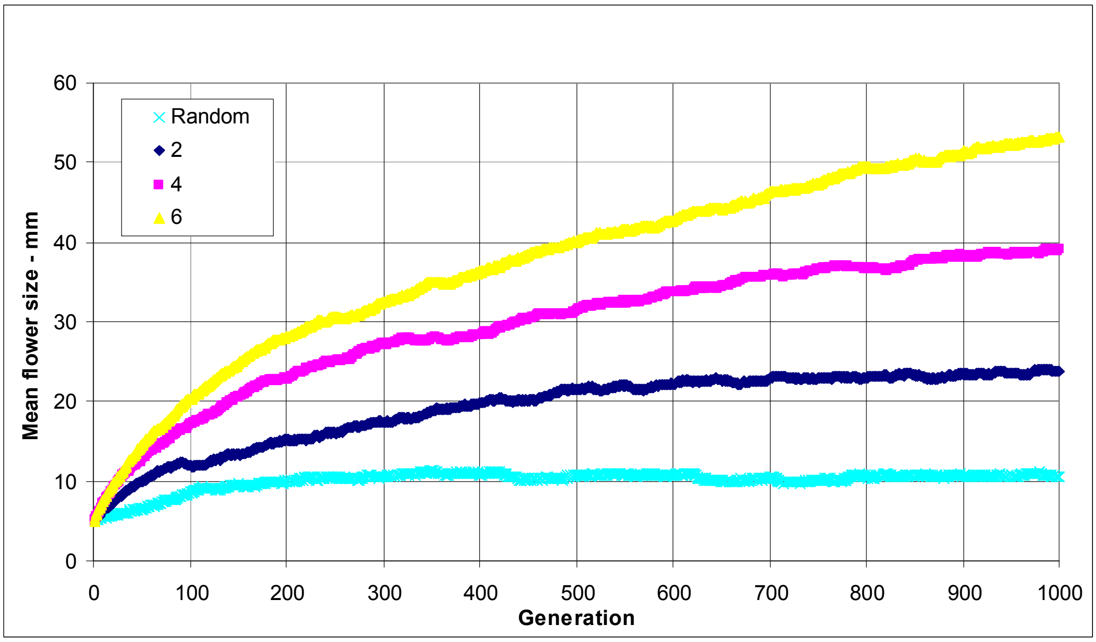


Figure 18 **Variation of flower size under different pollination association slopes**



Both factors have clear, and opposite selective effects on mean flower size within the population, although the effect of pollinator selection is both stronger, and increased in effectiveness as the strength of association increases, whilst progressive increases in the predation association strength has a diminishing return. However, it is clear that these two factors affect the evolution of flower size, within the assumptions of the model, although data from the field measurements indicates that pollination visits are close to 100% for both *L. vulgare* and *C. nigra*, which removes the possibility of selective association of pollination visiting with flower size.

Following the calculations on association slopes, the effect of predator damage and pollination levels within individual flower heads was investigated, and the results are shown in Figures 19 and 20.

Figure 19 Variation of flower size under different levels of overall predation

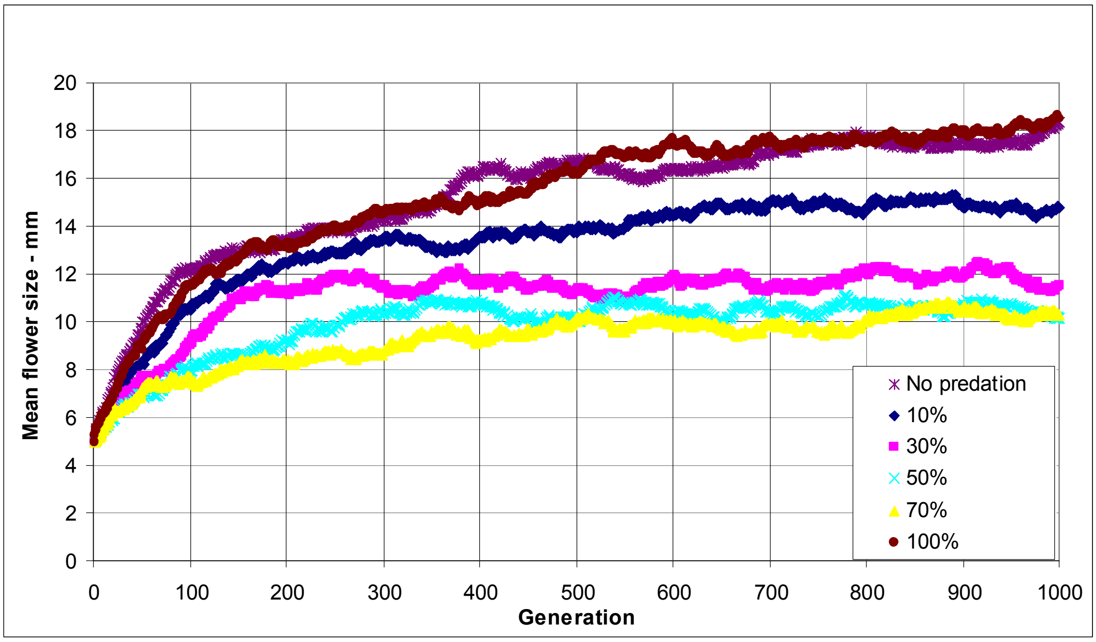
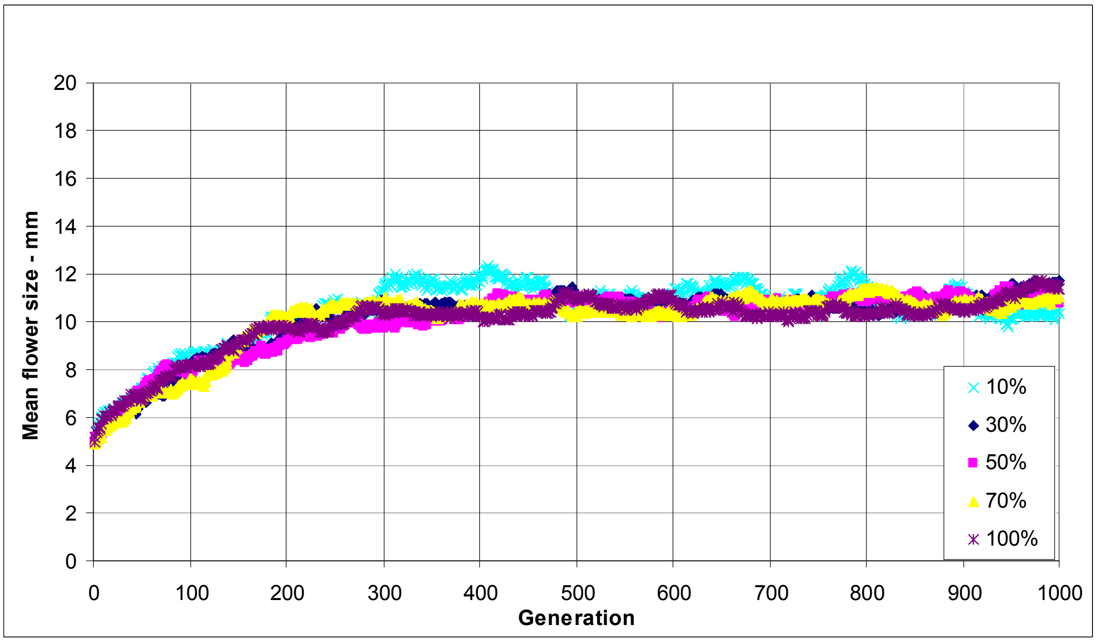


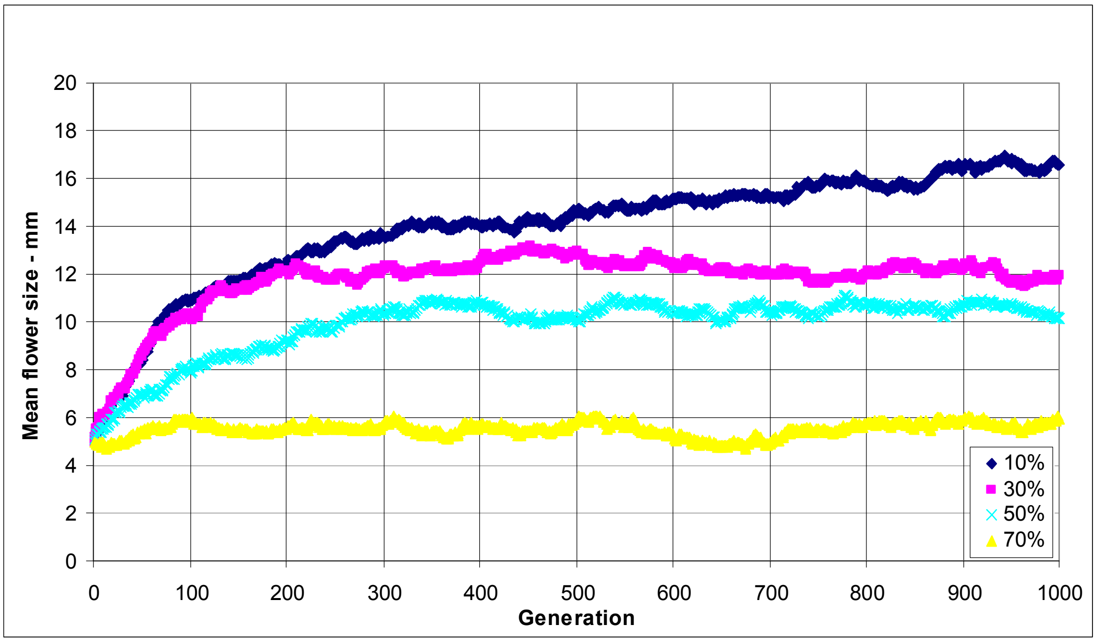
Figure 20 Variation of flower size under different levels of overall pollination



The effect of predation shows increasing effect of selective pressure from predation as the mean level increases up to 70% of flowers infested, but then reduces such that, if all are selected the evolutionary effect is cancelled as variation in outcome of predation reduces. For pollination, assuming no progressive effect of pollination visitation with size, the actual level has no evolutionary effect on flower size.

As expected, the individual damage level in each infestation has a strong effect on the evolutionary pressure caused by differential predation, as shown in Figure 21.

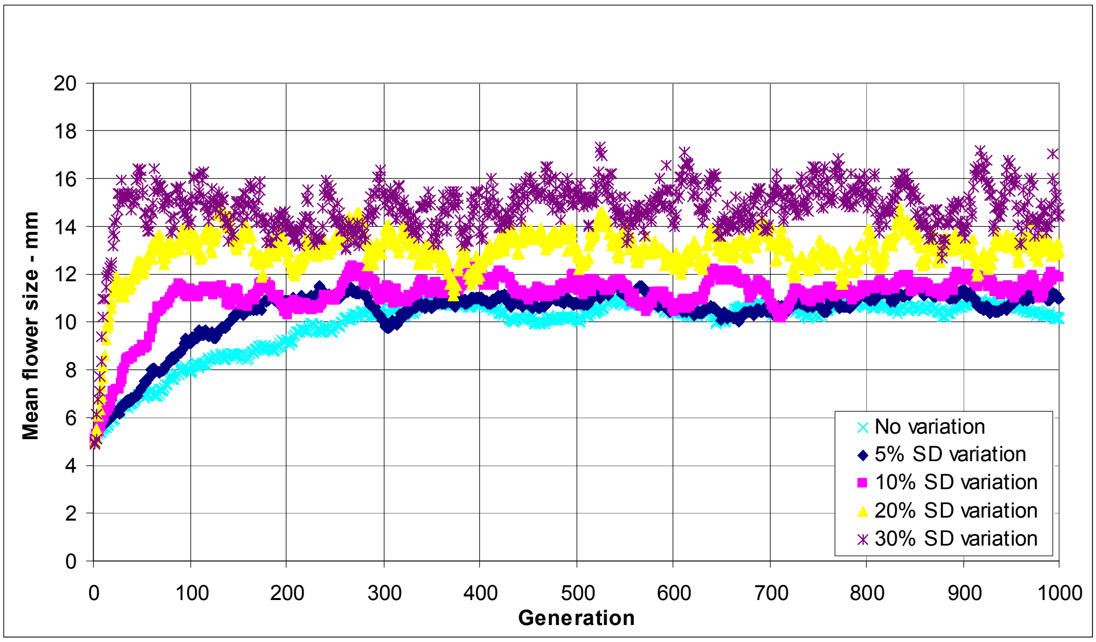
Figure 21 Variation of flower size under different predation damage per flower



Another feature expected to affect the outcome of selective pressure is the level of annual conditions for growth. Although the model does not contain any specific variables that count for annual resource availability, it can add a stochastic effect in terms of a variance in mean flower size, based upon the previous year's inheritance. This works by allocating seeds, and their seedlings a natural size, based on inheritance, but applying a randomly varying variance in final flower size on an annual basis.

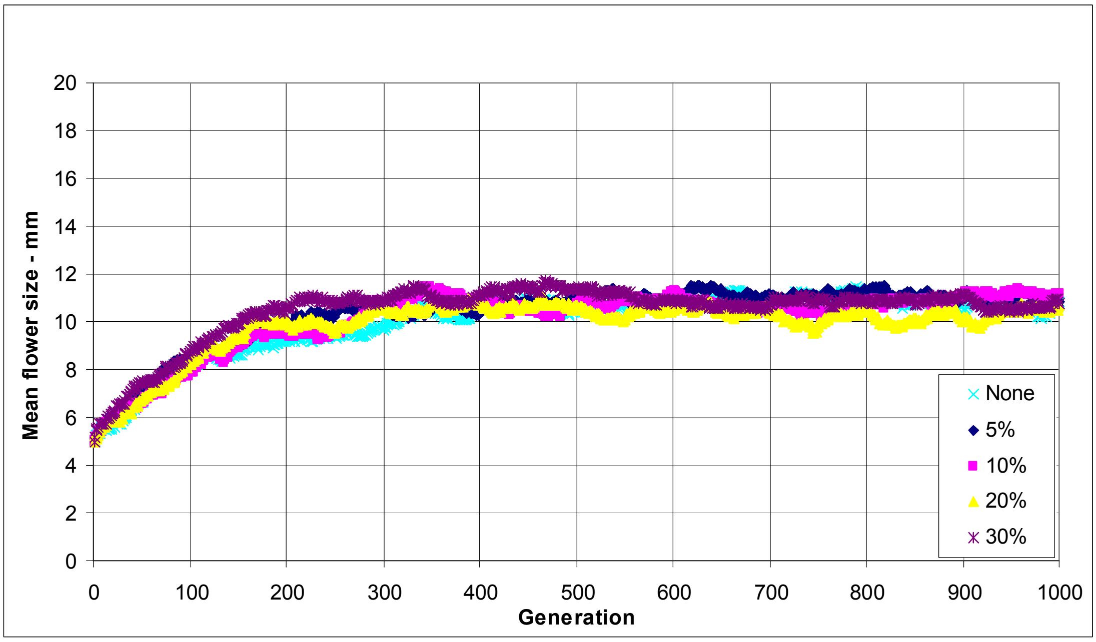
The resulting evolutionary effects are shown in Figure 22.

Figure 22 Variation of flower size with a stochastic annual variance of flower size



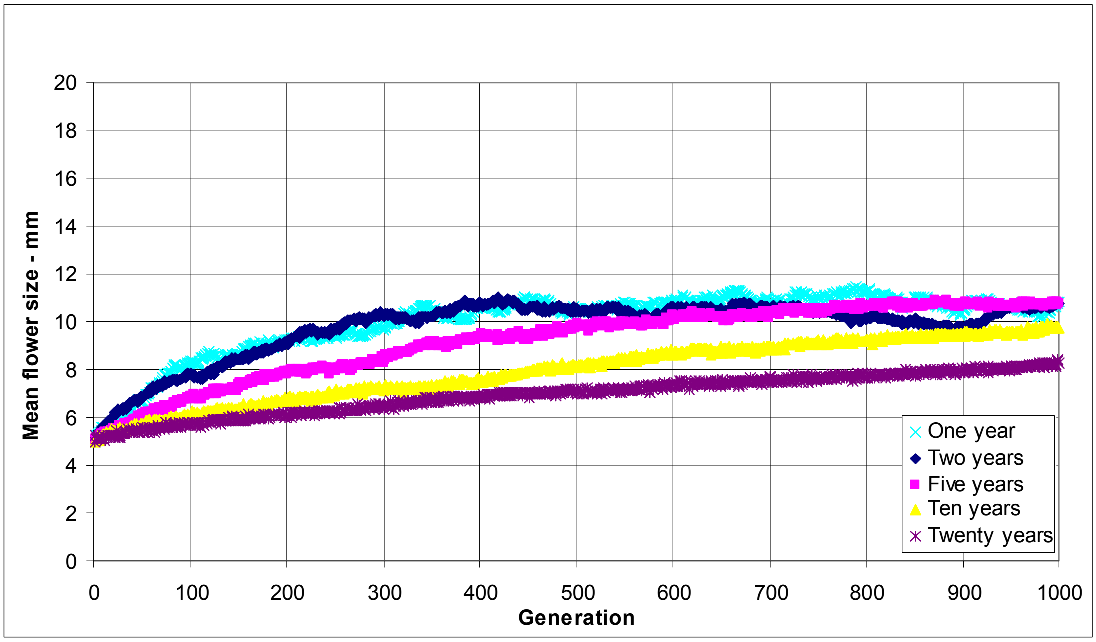
Here the effect of variation within populations acting as a catalyst for evolution is well shown, as increased levels of annual variation increase the speed with which the evolution of flower size occurs under the same base pressure. However, adding variance to the level of predation, separate from size variation does not seem to affect the results, as seen in Figure 23.

Figure 23 Variation of flower size with a stochastic annual variance of predation



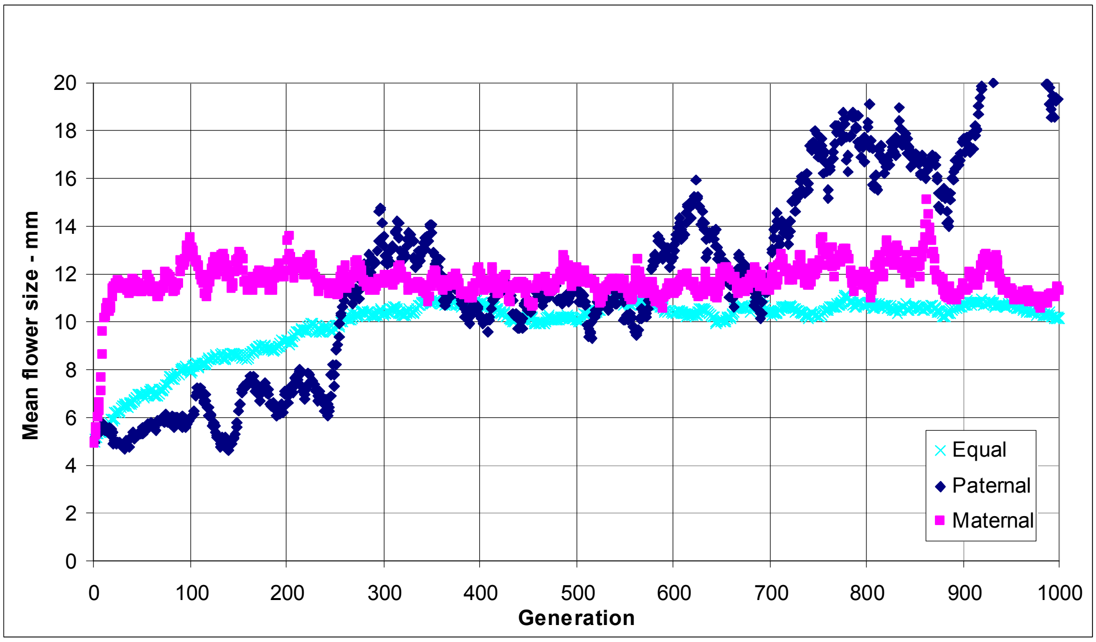
Neither the effects of multiple flower heads, varied up to 20 per plant, nor that of plant lifetime, investigated up to 10 years, made any difference to the effects of predation shown above. However, the addition of a long term seed viability in a seed bank did have a significant effect in slowing the course of evolution, as seen in Figure 24.

Figure 24 Variation of flower size with viability in the seed bank



Another factor that did have an effect was the mode of plant size heritability, as seen in Figure 25.

Figure 25 Variation of flower size with method of inheritance

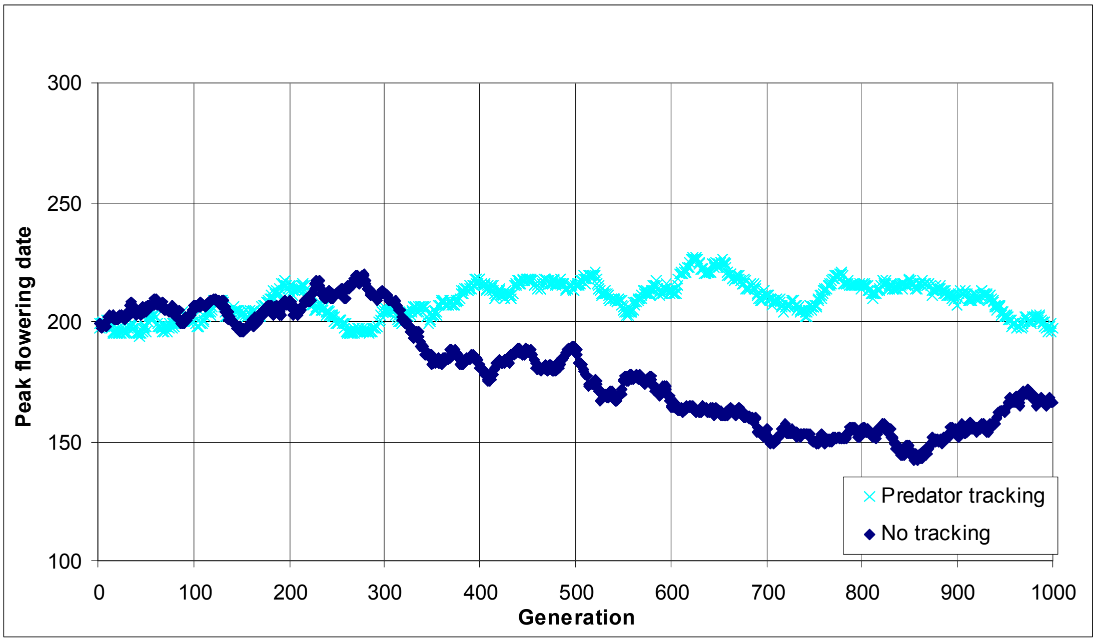


The model normally assumed a simple median inheritance of characteristics, averaging the components of both parents. The adoption of female only inheritance greatly increases the evolutionary effect, whilst that of purely paternal inheritance produces a much less predictable pattern, as the model assumes a purely random source of pollen, not making any assumption as to the pollinator's foraging habit.

Whilst the majority of the model predictions have covered the effects on flower size, some work was also undertaken on flowering phenology. The model has the ability to allow flowering phenology to alter without affecting the behaviour of the predators, or to link predator activity with the floral population.

When predation is unlinked in this manner, peak flowering can be altered by predation, as Figure 26 shows.

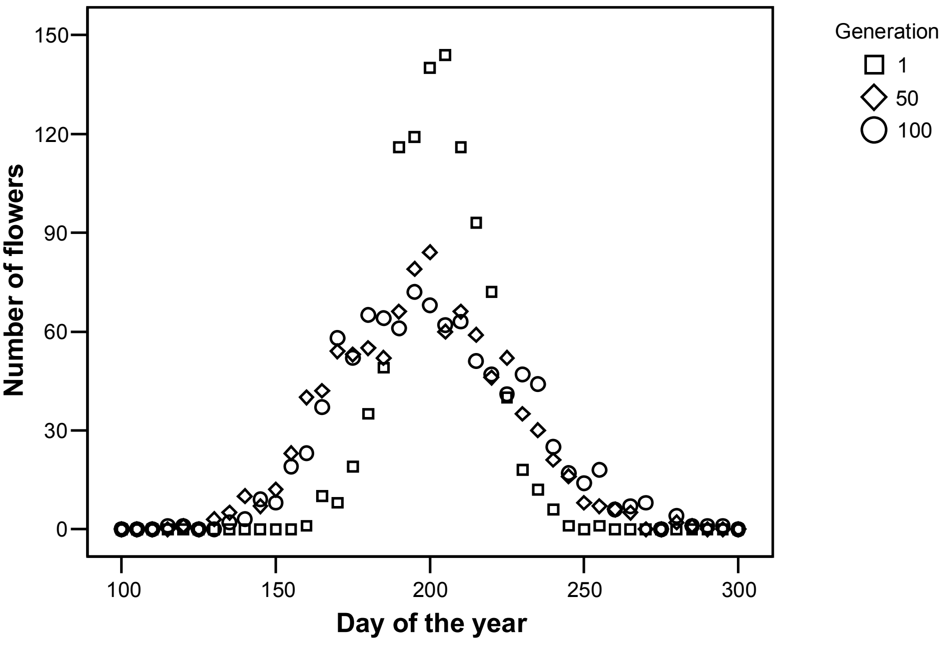
Figure 26 Variation of peak flowering date with predator behaviour



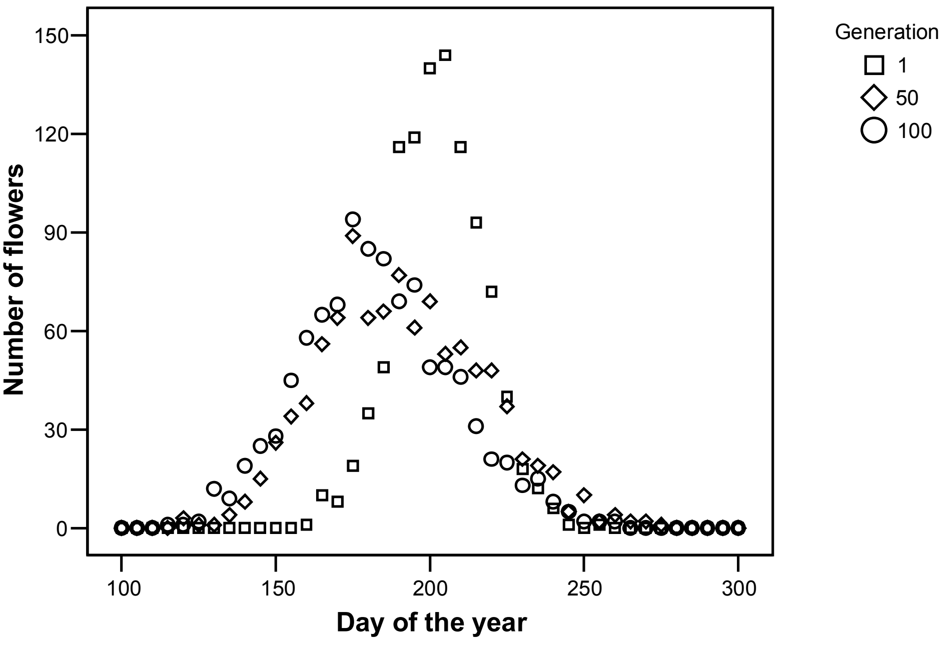
The effect can also be seen by examining the distribution of flowers in the model at the beginning and end of the test runs, which allows a secondary effect to be shown, that, when predation tracks phenology the model predicts an increased variance in the flower size of the population after 100 generations, shown in Figure 27.

Figure 27 Population phenology distributions

With predator tracking



Without predator tracking

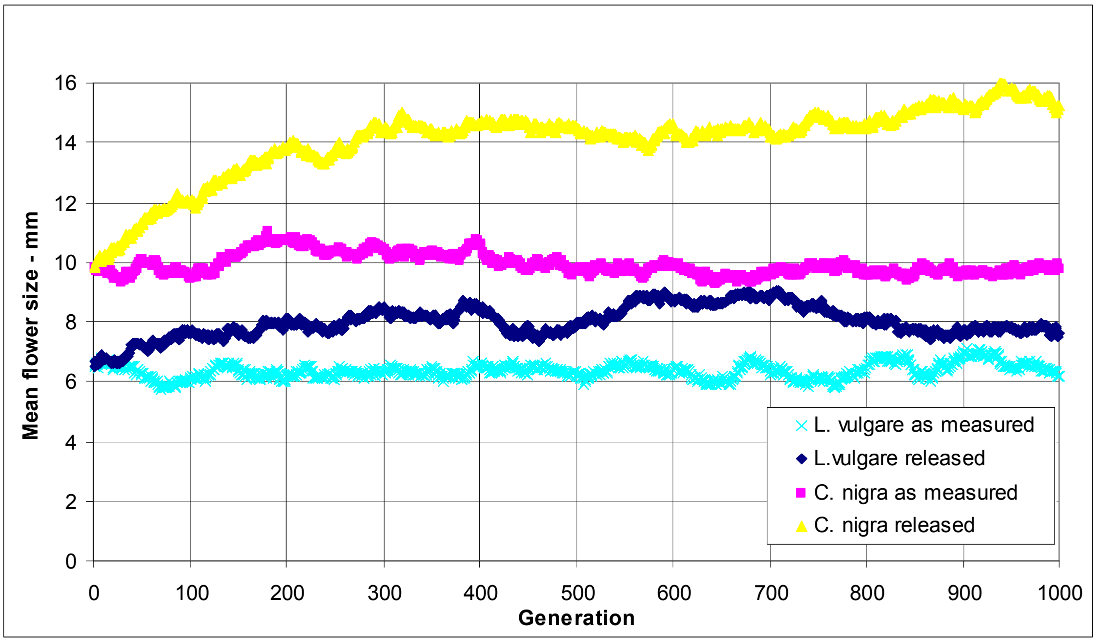


7.6.2 Release from predation

Having established the basic behaviour of the model, a final set of runs could be made to apply it to the species investigated in the field. The characteristic flower and floret sizes of both *Leucanthemum vulgare* and *Centaurea nigra*, together with measured patterns of seed set success from pollination and predation levels and damage, all presented above, were applied to the model. The equation used for the model's constraint on attainable flower size which, used to address resource cost, was, in each case tuned to maintain a constant flower size over 1000 generations, making the assumption that under the current levels of predation the populations are in equilibrium with evolutionary pressure for size increase.

The model was then re-run, for each flower species, with all predation removed, in order to assess, under the estimate of actual resource costs, whether predation has been reducing potential flower size. The results are presented in Figure 28.

Figure 28 **Release from predation of investigated species**



Populations from the end of each run were investigated for their statistical properties, as in Table 21.

Table 22 Statistics of pre and post predation release populations

Leucanthemum vulgare

Descriptive Statistics

Treatment		N	Minimum	Maximum	Mean	Std. Deviation
As measured	Diameter	200	2.84	11.07	6.5747	1.69580
	Valid N (listwise)	200				
Post release	Diameter	200	5.64	9.83	7.7009	.90740
	Valid N (listwise)	200				

ANOVA

Diameter

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	126.833	1	126.833	68.575	<0.001
Within Groups	736.123	398	1.850		
Total	862.956	399			

Centaurea nigra

Descriptive Statistics

Treatment		N	Minimum	Maximum	Mean	Std. Deviation
As measured	Diameter	200	5.37	14.78	9.9110	1.47770
	Valid N (listwise)	200				
Post release	Diameter	200	12.21	17.65	15.1616	.89320
	Valid N (listwise)	200				

ANOVA

Diameter

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2756.880	1	2756.880	1849.382	<0.001
Within Groups	593.300	398	1.491		
Total	3350.180	399			

In both cases, the model prediction, with its implicit assumptions, is that, at the measured levels of predation, the flower size is indeed constrained, and the difference is significant at $p<0.001$.

These data show that the computer model has shown itself to be capable of modelling trends in both flower size and phenology over many generations, at levels typical of those found in the field.

The most important aspect of its design has been the method of preferential choice of flowers within the population, both for pollination and predation, on a stochastic basis, modified by relative probability set by an equation entered as a variable factor. Once this was in place, and tuned to achieve results in line with those measured in the field, predictions became consistent.

The model shows that levels of predation actually measured for both *Leucanthemum vulgare* and *Centaurea nigra* are predicted to be capable of restricting or reducing floral size.
The model also predicts that predation may play a part in determining phenology, both in terms of the mean and variance of flowering seasons.

Thus, within the limits of the underlying assumptions, the results do confirm that, the original hypotheses, that the combined effect of selective infestation of flower heads, and reduction in fecundity, leads to an evolutionary pressure that selects for flower size in subsequent generations, and that the combined effect of the pattern of predation and reduction in fecundity leads to an evolutionary pressure that selects for phonological patterns in subsequent generations, do hold.

Chapter 8. General discussion

Over the course of six years, a combination of literature review, field work and mathematical modelling has been carried out in order to investigate the proposition that pollination and pre-dispersal seed predation can be selective force on the evolution of floral traits.

In order to investigate the proposition, a number of hypotheses were posed:

1 Pre-dispersal seed predators tend to select larger flower heads as oviposition sites in a repeatable pattern over time and population.

2 The presence of seed predators within flower heads reduces their fecundity.

3 The size of flower heads is correlated with successful seed set.

4 The pattern of seed predation over the course of a season is repeatable over several seasons and locations.

5 The combined effect of selective infestation of flower heads, and reduction in fecundity, and correlation of flower size and seed set, leads to an evolutionary pressure that selects for an optimum flower size in subsequent generations.

6 The combined effect of the pattern of predation and reduction in fecundity leads to an evolutionary pressure that selects for phenological patterns in subsequent generations.

Existing literature, discussed in Chapter 2, showed many examples of research to suggest that these hypotheses might be accepted, as well as suggesting mechanisms for them. However, the literature has little evidence of longer term repeatability, which might be seen as an important component of any selective pressure.

The field research examined in Chapter 3 presents a considerable body of data for four plant species in the Asteraceae, all of which are represented in Fenner's 2002 paper which presented data relating flower size with levels of infestation, collected at various sites throughout six growing seasons, and groups them by type of infestation found by dissection. As each species showed a trend to reduce the absolute flower size as the growing season progressed, size data were normalised by the mean of the measured size on each collection date. Descriptive statistics indicate differences in the group means. Each species showed patterns of infestation by several species of Tephritid fly within season, but relatively stable overall levels of infestation.

The different attack patterns of poly and mono phagous tephtitids on *Centaurea nigra* are in line with the work of several authors such as Clement and Sobhian (1991) who looked at specificity of predation, concluding that *Urophora* species demonstrated high levels of host specificity, whilst Kinkorova and Mickova (2006) show the polyphagous nature of *Chaetostomella cylindrica* infesting the same species.

Examination of the data collected by means of the t test, in Chapter 4, using oviposition choice of the predating species as a grouping criterion showed high levels of significance in the relative size of selected flower heads for some combinations of host and predator, especially for *Leucanthemum vulgare*/*Tephritis neesii* and *Centaurea nigra*/*Urophora spp.* These values confirm data from Fenner (2002), Campbell et al. (2002), Hainsworth et al. (1984) and Petersen (2000), all of whom noted a correlation of floral size with pre-dispersal seed predation, and postulated the latter as a possible evolutionary pressure.

A further analysis demonstrated a characteristic and progressive association of relative probability of predation with relative flower size, each interaction having its own characteristic slope that was maintained over a wide range of overall infestation levels. Further work in species of other plant families would be of great interest to see if such association was also present. However, the nature of floral morphology in the Asteraceae, and the oviposition techniques of small dipterae might mean that such an association is more common with this specific predation. In his detailed work on oviposition, Straw (1989b) demonstrated an ability for tephritid measurement of capitulum size in Asteraceae, thus providing a plausible mechanism for such floral selection to occur. Ehrlen (1996), Zimmerman and Brody (1998) and Brody and Morita (2000) also present good evidence on oviposition site selection by floral size in other plant families.

These results, however, show significant evidence for an acceptance of the hypothesis that, within the chosen interactions, predators do choose flowers for oviposition on the basis of size, with increased probability as flower size increases, in a pattern repeated over both time and space. Coupled with the heritability of flower size, specifically demonstrated by Bogle (1983) in *Leucanthemum vulgare,* and more generally discussed in Geber and Griffen (2003) and Mazer and Delsalle (1996), one of the prime requirements of selective pressure on a specific floral trait, which could thefere lead to evolutionary change, is well demonstrated. The extended time period covered in this study, which is relatively novel, should add particularly to the data on temporal variation of predation.

All field studies of infestation may suffer from the criticism that what is measured may be an artefact of the infestation itself. Here some evidence has been presented to show that these associations are indeed a genuine attribute of the host predator relationship, although this aspect cannot be ruled out entirely. The existence of a mechanism for flower measurement by the predators, Straw (1989b), the work of Hainsworth et al. (1984), Campbell et al. (2002), Petersen (2000) and Fenner et al. (2002), and the statistical differences between the measured data and models of random selection coupled with capitulum swelling, demonstrated in this work, adds significantly to the case of a genuine effect.

Chapter 5 presents evidence to support the second hypothesis that predation reduces fecundity. Analysis of seed set success shows high levels of significance for reduced fecundity under predation in two of the studied species, sufficient to accept the premise. However, for *T. inodorum* the relationship appeared to be commensal rather than predatory, illustrating the specificity of such interactions, and showing further the importance of studying individual instances as recommended by Janzen (1975).

These results confirm data presented by Louda et al. (1990) and Louda and Potvin (1995), as well as Maron (2002) where exclusion experiments on members of the Asteraceae demonstrated significant loss of fecundity under predation.

As with the heritability of flower size in the Asteraceae mentioned above, evidence from Young et al. (1994), Kleunen and Ritland (2004) and Worley and Barrett (2001) shows seed number to be a heritable trait within the family. All three species investigated for seed set showed a significant correlation of seed set success with flower size. The basic physiology of the Asteraceae is one of increasing seed potential as flower size increases, as the data show little difference in floret size with capitulum width. This is good evidence to accept the third hypothesis.

Taking the selective predation on flower size together with the effective reduction in fecundity of infested flowers, and the heritability suggested in the literature, a strong case can be made for the effects of pre-dispersal seed predation in the plant species investigated acting as an evolutionary pressure on flower size.

The evidence of this study showed an almost universal level of pollination visitation to flower heads, although this need not be the case in other populations of the same species, depending upon the local insect fauna. In this case therefore, the opposing selective pressures of predation and pollination on floral evolution noted by Brody (1997), Brody and Mitchell (1997), Kudoh and Whigham (1998) Herrera (2000), Galen and Cuba (2001) and Ehrlen et al. (2002) are not likely to be operating. However, as the natural morphology of the Asteraceae coupling seed production to flower size, may itself act as a selective force towards larger flowers, as demonstrated in the mathematical modelling of Chapter 7, predation pressure may well operate to maintain a relatively stable flower size within the population.

Finally, from the field data, Chapter 6 examined the patterns of predation, which are characteristic of each host-predator interaction. By nature, there is a certain level of repeatability in these, simply by their seasonal nature, and a statistical analysis failed to find evidence of a repeatable level of infestation across time or space. This may not be surprising bearing in mind that six seasons, whilst being a relatively long period for a single study, is very short in terms of the expected environmental variation within a few years, in effect being only six sample points within an analysis.

The resulting patterns of infestation might be seen as evidence supporting the suggestions of Albrechtsen (2000), Wright and Meagher (2003), Freeman et al. (2003), Desteven (1981) and Herrera (1991) that predation can affect flowering phenology by acting as a selective force. Clearly such an evolutionary implication could be created simply from these patterns, but the field data collected here does not add any hard evidence. However, values of overall mean infestation rate and their variance were measured, and these could be used in the subsequent theoretical investigation.

In order to examine the cumulative effects of the acceptance of the first three hypotheses, and the data gathered on seasonal repeatability, a novel mathematical model of floral populations was created that includes both stochastic effects of size and phenological traits within each generation, but allows selective pollination success and predation based upon floral size and oviposition date, as described in Chapter 7.

The model identified general trends in typical Asteraceae that showed that both pre-dispersal seed predation and pollination visitations, when applied with a progressive probability with increasing flower size, have contrasting effects on the evolution of flower size. Furthermore, increased variation in the distribution of flower sizes due to annual environmental variation tends to speed the evolutionary process.

The model showed no tendency to be affected by the lifespan of individual plants, nor whether they were single or multi flowered, but the persistence of seeds within a seed bank did slow evolutionary change.

Examination of predation patterns within the model showed the possible effects on plant phenology of the populations within the model, indicating the ability of predation to alter the plant's peak flowering date, and a possible escape route from predation as postulated by by Desteven (1981), and opening a possible path to speciation, as suggested by Hendry and Day (2005).

The model results support the hypothesis that pre-dispersal seed predation can indeed affect floral traits, both in terms of flower size and flowering pattern. When the parameters measured from the field were applied to the two species shown to be affected by predation in terms of fecundity, it predicted that predation does affect their characteristic flower size, with high levels of significance between the initial and later populations.

Of course, such a model cannot establish evolutionary change in the real populations, it can only suggest that, within the assumptions inherent in its design, such change might be occurring. Demonstrating evolution in real time is very difficult, even for a prolonged investigation. But within the terms of this study, the model confirms its basic hypotheses that pollination and predation can affect the evolutionary course of Asteraceae.

Taken together with the existing literature, this study adds further evidence to a view of evolution that is both universal in its effects and particular in its means. The role of insect behaviour in the evolutionary history of plants is primarily considered in terms of the development of the angiosperms and their pollinators. Here we see a counteracting force, perhaps indeed a stabilising force, caused by predation, which, in two particular cases, shows good evidence for its potency.

Chapter 9. References

W. G. Abrahamson, M. D. Eubanks, C. P. Blair, and A. V. Whipple. Gall flies, inquilines, and goldenrods: A model for host-race formation and sympatric speciation. *American Zoologist* 41 (4):928-938, 2001.

W. G. Abrahamson, C. P. Blair, M. D. Eubanks, and S. A. Morehead. Sequential radiation of unrelated organisms: the gall fly Eurosta solidaginis and the tumbling flower beetle Mordellistena convicta. *Journal of Evolutionary Biology* 16 (5):781-789, 2003.

F. J. Acosta, J. A. Delgado, E. Lopez, and J. M. Serrano. Functional features and ontogenetic changes in reproductive allocation and partitioning strategies of plant modules. *Plant Ecology* 132 (1):71-76, 1997.

J. R. Akeroyd and D. Briggs. Genecological Studies of Rumex-Crispus l .1. Garden Experiments Using Transplanted Material. *New Phytologist* 94 (2):309-323, 1983.

J. R. Akeroyd and D. Briggs. Genecological Studies of Rumex-Crispus l .2. Variation in Plants Grown from Wild-Collected Seed. *New Phytologist* 94 (2):325-343, 1983.

B. R. Albrectsen. Flowering phenology and seed predation by a tephritid fly: Escape of seeds in time and space. *Ecoscience* 7 (4):433-438, 2000.

L. K. Amsberry and J. L. Maron. Effects of herbivore identity on plant fecundity. *Plant Ecology* 187 (1):39-48, 2006.

W. S. Armbruster. Evolution of floral form: electrostatic forces, pollination and adaptive compromise. *New Phytologist* 152:181-183, 2001.

W. S. Armbruster. Exaptions link evolution of plant-herbivore and plant-pollinator interactions: a phylogenetic. *Ecology* 78 (6):1661-1672, 1997.

E. Asikainen and P. Mutikainen. Preferences of pollinators and herbivores in gynodioecious Geranium sylvaticum. *Annals of Botany* 95 (5):879-886, 2005.

D. E. Berube. Interspecific Competition Between Urophora-Affinis and Urophora-Quadrifasciata (Diptera, Tephritidae) for Ovipositional Sites on Diffuse Knapweed (Centaurea-Diffusa, Compositae). *Zeitschrift fur Angewandte Entomologie-Journal of Applied Entomology* 90 (3):299-306, 1980.

D. E. Berube and J. H. Myers. Reproductive Isolation Between Urophora-Affinis and Urophora-Quadrifasciata (Diptera, Tephritidae) in British-Columbia. *Canadian Journal of Zoology-Revue Canadienne de Zoologie* 61 (4):787-791, 1983.

D. S. Bigger. Consequences of patch size and isolation for a rare plant: Pollen limitation and seed predation. *Natural Areas Journal* 19 (3):239-244, 1999.

A. L. Bogle. Discoid Heads - An Inheritable Trait in Chrysanthemum-Leucanthemum l. *Rhodora* 85 (844):461-464, 1983.

M. Brandle, S. Knoll, S. Eber, J. Stadler, and R. Brandl. Flies on thistles: support for synchronous speciation? *Biological Journal of the Linnean Society* 84 (4):775-783, 2005.

A. Brewis. *The Flora of Hampshire*, Harley Books, 1996.

D. T. Briese. Impact of the Onopordum capitulum weevil Larinus latus on seed production by its host-plant. *Journal of Applied Ecology* 37 (2):238-246, 2000.

A. K. Brody and S. I. Morita. A positive association between oviposition and fruit set: female choice or manipulation? *Oecologia* 124 (3):418-425, 2000.

A. K. Brody and R. J. Mitchell. Effects of experimental manipulation of inflorescence size on pollination and pre-dispersal seed predation in the hummingbird-pollinated plant Ipomopsis aggregata. *Oecologia* 110 (1):86-93, 1997.

A. K. Brody. Oviposition Choices by A Predispersal Seed Predator (Hylemya Sp) .1. Correspondence with Hummingbird Pollinators, and the Role of Plant Size, Density and Floral Morphology. *Oecologia* 91 (1):56-62, 1992.

Alison K. Brody. Effects of Pollinators, Herbivores, and Seed Predators on Flowering Phenology. *Ecology* 78 (6):1624-1631, 1997.

R. Brys, H. Jacquemyn, P. Endels, F. van Rossum, M. Hermy, L. Triest, L. De Bruyn, and G. D. E. Blust. Reduced reproductive success in small populations of the self-incompatible Primula vulgaris. *Journal of Ecology* 92 (1):5-14, 2004.

B. Burkhardt and H. Zwolfer. Macro-evolutionary trade-offs in the tephritid genus Urophora: benefits and costs of an improved plant gall. *Evolutionary Ecology Research* 4 (1):61-77, 2002.

R. O. Bustamante and J. A. Simonetti. Seed predation and seedling recruitment in plants: the effect of the distance between parents. *Plant Ecology* 147 (2):173-183, 2000.

L. M. Calvo-Irabien and A. Islas-Luna. Predispersal predation of an understory rainforest herb Aphelandra aurantiaca (Acanthaceae) in gaps and mature forest. *American Journal of Botany* 86 (8):1108-1113, 1999.

D. R. Campbell. Evolution of floral traits in a hermaphroditic plant: Field measurements of heritabilitles and genetic correlations. *Evolution* 50 (4):1442-1453, 1996.

D. R. Campbell, N. Crawford, A. K. Brody, and T. A. Forbis. Resistance to pre-dispersal seed predators in a natural hybrid zone. *Oecologia* 131 (3):436-443, 2002.

D. Cariveau, R. E. Irwin, A. K. Brody, L. S. Garcia-Mayeya, and A. von der Ohe. Direct and indirect effects of pollinators and seed predators to selection on plant and floral traits. *Oikos* 104 (1):15-26, 2004.

A. L. Case, E. P. Lacey, and R. G. Hopkins. Parental effects in plantago lanceolata L .2. Manipulation of grandparental temperature and parental flowering time. *Heredity* 76:287-295, 1996.

N. P. Chacoff, J. M. Morales, and M. D. Vaquera. Effects of fragmentation on amount of aborted and predated seed in the Chaco Serrano Argentina. *Biotropica* 36 (1):109-117, 2004.

M. L. Cipollini and E. W. Stiles. Seed Predation by the Bean Weevil Acanthoscelides-Obtectus on Phaseolus Species - Consequences for Seed Size, Early Growth and Reproduction. *Oikos* 60 (2):205-214, 1991.

S. L. Clement and R. Sobhian. Host-Use Patterns of Capitulum-Feeding Insects of Yellow Starthistle - Results from A Garden Plot in Greece. *Environmental Entomology* 20 (2):724-730, 1991.

S. L. Clement. Resistance Among Populations of Yellow Starthistle to Thistle-Head Insects - Results from Garden Plots in Italy. *Biological Control* 4 (2):149-156, 1994.

D. R. Clements, D. E. Cole, S. Darbyshire, J. King, and A. McClay. The biology of Canadian weeds. 128. Leucanthemum vulgare lam. *Canadian Journal of Plant Science* 84 (1):343-363, 2004.

M. J. Crawley. *Seed predators and plant population dynamics*, 1992.

M. J. Crawley. *Seed predators and plant population dynamics*, 2000.

M. L. Crowe and R. S. Bourchier. Interspecific interactions between the gall-fly Urophora affinis Frfld. (Diptera : Tephritidae) and the weevil Larinus minutus Gyll. (Coleoptera : Curculionidae), two biological control agents released against spotted knapweed, Centaurea stobe L. ssp micranthos. *Biocontrol Science and Technology* 16 (4):417-430, 2006.

C. L. Cummings, H. M. Alexander, and A. A. Snow. Increased pre-dispersal seed predation in sunflower crop-wild hybrids. *Oecologia* 121 (3):330-338, 1999.

J. P. Dempster, D. A. Atkinson, and O. D. Cheesman. The Spatial Population-Dynamics of Insects Exploiting A Patchy Food Resource .1. Population Extinctions and Regulation. *Oecologia* 104 (3):340-353, 1995.

J. P. Dempster, D. A. Atkinson, and M. C. French. The Spatial Population-Dynamics of Insects Exploiting A Patchy Food Resource .2. Movements Between Patches. *Oecologia* 104 (3):354-362, 1995.

D. Desteven. Pre-Dispersal Seed Predation in A Tropical Shrub (Mabea-Occidentalis, Euphorbiaceae). *Biotropica* 13 (2):146-150, 1981.

G. Dieringer. Variation in Individual Flowering Time and Reproductive Success of Agalinis-Strictifolia (Scrophulariaceae). *American Journal of Botany* 78 (4):497-503, 1991.

G. M. Dlussky, K. P. Glazunova, and N. V. Lavrova. The flower aind blossom morphology of Asteraceae correlates with composition of their pollinators. *Zhurnal Obshchei Biologii* 65 (6):490-499, 2004.

A. E. Duggan. Pre-Dispersal Seed Predation by Anthocharis-Cardamines (Pieridae) in the Population-Dynamics of the Perennial Cardamine-Pratensis (Brassicaceae). *Oikos* 44 (1):99-106, 1985.

J. Ehrlen, S. Kack, and J. Agren. Pollen limitation, seed predation and scape length in Primula farinosa. *Oikos* 97 (1):45-51, 2002.

J. Ehrlen. Spatiotemporal variation in predispersal seed predation intensity. *Oecologia* 108 (4):708-713, 1996.

M. Fenner and W. G. Lee. Lack of pre-dispersal seed predators in introduced Asteraceae in New Zealand. *New Zealand Journal of Ecology* 25 (1):95-99, 2001.

M. Fenner, J. E. Cresswell, R. A. Hurley, and T. Baldwin. Relationship between capitulum size and pre-dispersal seed predation by insect larvae in common Asteraceae. *Oecologia* 130 (1):72-77, 2002.

M. Fenner. *Seed Ecology*, 1985.

J. A. Figueroa, A. A. Munoz, J. E. Mella, and M. T. K. Arroyo. Pre- and post-dispersal seed predation in a mediterranean-type climate montane sclerophyllous forest in central Chile. *Australian Journal of Botany* 50 (2):183-195, 2002.

P. M. Forget, K. Kitajima, and R. B. Foster. Pre- and post-dispersal seed predation in Tachigali versicolor (Caesalpiniaceae): effects of timing of fruiting and variation among trees. *Journal of Tropical Ecology* 15:61-81, 1999.

R. S. Freeman, A. K. Brody, and C. D. Neefus. Flowering phenology and compensation for herbivory in Ipomopsis aggregata. *Oecologia* 136 (3):394-401, 2003.

G. Freese. The Insect Complexes Associated with the Stems of 7 Thistle Species. *Entomologia Generalis* 19 (3):191-207, 1994.

M. Frenzel, K. Dettner, W. Boland, and P. Erbes. Identification and Biological Significance of 4-Methyl-3Z,5-Hexadienoic Acid Produced by Males of the Gall-Forming Tephritids Urophora-Cardui (L) and Urophora-Stylata (Fab) (Diptera, Tephritidae). *Experientia* 46 (5):542-547, 1990.

C. Galen and J. Cuba. Down the tube: Pollinators, predators, and the evolution of flower shape in the alpine skypilot, Polemonium viscosum. *Evolution* 55 (10):1963-1971, 2001.

C. Galen. Rates of floral evolution: Adaptation to bumblebee pollination in an alpine wildflower, Polemonium viscosum. *Evolution* 50 (1):120-125, 1996.

A. C. Gange, V. K. Brown, and D. M. Aplin. Ecological specificity of arbuscular mycorrhizae: evidence from foliar- and seed-feeding insects. *Ecology* 86 (3):603-611, 2005.

C. Garcia-Robledo, G. Kattan, C. Murcia, and P. Quintero-Marin. Equal and opposite effects of floral offer and spatial distribution on fruit production and predispersal seed predation in Xanthosoma daguense (Araceae). *Biotropica* 37 (3):373-380, 2005.

M. A. Geber and L. R. Griffen. Inheritance and natural selection on functional traits. *International Journal of Plant Sciences* 164 (3):S21-S42, 2003.

S. A. H. Geritz. Co-evolution of seed size and seed predation. *Evolutionary Ecology* 12 (8):891-911, 1998.

S. A. Graham. Gall makers on flowers of Cuphea (Lythraceae). *Biotropica* 27 (4):461-467, 1995.

Thomas W. Green and Ivan G. Palmbald. Effects of Insect Seed Predators on Astragalus cibarius and Astragalus utahensis (Leguminosae). *Ecology* 56 (6):1435-1440, 1975.

N. Greig. Predispersal Seed Predation on 5 Piper Species in Tropical Rain-Forest. *Oecologia* 93 (3):412-420, 1993.

F. R. Hainsworth, L. L. Wolf, and T. Mercier. Pollination and Pre-Dispersal Seed Predation - Net Effects on Reproduction and Inflorescence Characteristics in Ipomopsis-Aggregata. *Oecologia* 63 (3):405-409, 1984.

John M. Halley and J. P. Dempster. The Spatial Population Dynamics of Insects Exploiting a Patchy Food Resource: A Model Study of Local Persistence. *The Journal of Applied Ecology* 33 (3):439-454, 1996.

T. F. Hansen, C. Pelabon, W. S. Armbruster, and M. L. Carlson. Evolvability and genetic constraint in Dalechampia blossoms: components of variance and measures of evolvability. *Journal of Evolutionary Biology* 16 (4):754-766, 2003.

P. Harris and J. D. Shorthouse. Effectiveness of gall inducers in weed biological control. *Canadian Entomologist* 128 (6):1021-1055, 1996.

P. Harris. Effects of Urophora-Affinis Frfld and Urophora-Quadrifasciata (Meig) (Diptera, Tephritidae) on Centaurea-Diffusa Lam and Centaurea-Maculosa Lam (Compositae). *Zeitschrift fur Angewandte Entomologie-Journal of Applied Entomology* 90 (2):190-201, 1980.

A. M. Hemborg and L. Despres. Oviposition by mutualistic seed-parasitic pollinators and its effects on annual fitness of single- and multi-flowered host plants. *Oecologia* 120 (3):427-436, 1999.

A. P. Hendry and T. Day. Population structure attributable to reproductive time: isolation by time and adaptation by time. *Molecular Ecology* 14 (4):901-916, 2005.

C. M. Herrera. Measuring the effects of pollinators and herbivores: Evidence for non-additivity in a perennial herb. *Ecology* 81 (8):2170-2176, 2000.

C. M. Herrera. Selective Pressures on Fruit Seediness - Differential Predation of Fly Larvae on the Fruits of Berberis-Hispanica. *Oikos* 42 (2):166-170, 1984.

J. Herrera. Allocation of Reproductive Resources Within and Among Inflorescences of Lavandula Stoechas (Lamiaceae). *American Journal of Botany* 78 (6):789-794, 1991.

A. Honek and Z. Martinkova. Pre-dispersal predation of Taraxacum officinale (dandelion) seed. *Journal of Ecology* 93 (2):335-344, 2005.

S. E. Howarth and J. T. Williams. Chrysanthemum Leucanthemum L. *The Journal of Ecology* 56 (2):585-595, 1968.

R. E. Irwin, S. Y. Strauss, S. Storz, A. Emerson, and G. Guibert. The role of herbivores in the maintenance of a flower color polymorphism in wild radish. *Ecology* 84 (7):1733-1743, 2003.

D. H. Janzen. Intrahabitat and Interhabitat Variations in Guazuma-Ulmifolia (Sterculiaceae) Seed Predation by Amblycerus-Cistelinus (Bruchidae) in Costa-Rica. *Ecology* 56 (4):1009-1013, 1975.

L. A. Janzon. Descriptions of Tephritis-Neesii Meigen (Diptera, Tephritidae) and Its Parasitoids (Hymenoptera, Ichneumonoidea, Chalcidoidea) in Flower-Heads of Leucanthemum-Vulgare Lamarck (Asteraceae) in Sweden. *Entomologica Scandinavica* 11 (1):113-126, 1980.

Q. O. N. Kay. Tripleurospermum-Inodorum (L) Schultz Bip (Matricaria-Inodora L, Matricaria-Maritima Auct P P Non L, Matricaria-Perforata Merat, Tripleurospermum-Perforatum (Merat) Wagenitz, Tripleurospermum-Maritimum (L) Schultz Bip P P Non L). *Journal of Ecology* 82 (3):681-697, 1994.

T. N. Kaye. From flowering to dispersal: Reproductive ecology of an endemic plant, Astragalus australis var. olympicus (Fabaceae). *American Journal of Botany* 86 (9):1248-1256, 1999.

C. A. Kelly and R. J. Dyer. Demographic consequences of inflorescence-feeding insects for Liatris cylindracea, an iteroparous perennial. *Oecologia* 132 (3):350-360, 2002.

C. A. Kelly. Quantitative Genetics of Size and Phenology of Life-History Traits in Chamaecrista-Fasciculata. *Evolution* 47 (1):88-97, 1993.

D Kelly, Deirdre E. Hart, and Robert B. Allen. Evaluating the Wind Pollination Benefits of Mast Seeding. *Ecology* 82 (1):117-126, 2001.

D. Kelly, M. J. Mckone, K. J. Batchelor, and J. R. Spence. Mast Seeding of Chionochloa (Poaceae) and Pre-Dispersal Seed Predation by A Specialist Fly (Diplotoxa, Diptera, Chloropidae). *New Zealand Journal of Botany* 30 (2):125-133, 1992.

D. Kelly and J. J. Sullivan. Quantifying the benefits of mast seeding on predator satiation and wind pollination in Chionochloa pallens (Poaceae). *Oikos* 78 (1):143-150, 1997.

J. Kinkorova. Life Histories of Terellia-Serratulae and Urophora-Stylata (Diptera, Tephritidae) and Their Cooccurrence on Cirsium-Vulgare. *Acta Entomologica Bohemoslovaca* 88 (5):293-298, 1991.

J. Kinkorova and M. Mickova. Life history of the European tephritid fly Chaetostomella cylindrica (Diptera : Tephriticlae) and its possible use as a biocontrol agent in the USA. *Proceedings of the Entomological Society of Washington* 108 (1):51-61, 2006.

H. Kudoh and D. F. Whigham. The effect of petal size manipulation on pollinator/seed-predator mediated female reproductive success of Hibiscus moscheutosShow a selective pressure towards mast seeding in a grass species due to pre-dispersal seed predation. *Oecologia* 117 (1-2):70-79, 1998.

A. J. Lack. Competition for Pollinators in the Ecology of Centaurea-Scabiosa l and Centaurea-Nigra l .1. Variation in Flowering Time. *New Phytologist* 91 (2):297-308, 1982.

A. J. Lack. Competition for Pollinators in the Ecology of Centaurea-Scabiosa l and Centaurea-Nigra l .2. Observations on Nectar Production. *New Phytologist* 91 (2):309-320, 1982.

Andrew J. Lack. Competition for Pollinators in the Ecology of Centaurea scabiosa L. and Centaurea nigra L. III. Insect Visits and the Number of Successful Pollinations. *New Phytologist* 91 (2):321-339, 1982.

Andrew J. Lack. The Ecology of Flowers of Chalk Grassland and their Insect Pollinators. *The Journal of Ecology* 70 (3):773-790, 1982.

M. Lacko-Bartosova, M. Minar, Z. Vranovska, and D. Strasser. Weed seed bank in ecological and integrated farming system. *Rostlinna Vyroba* 46 (7):319-324, 2000.

R. G. Lalonde and J. D. Shorthouse. Developmental Morphology of the Gall of Urophora-Cardui (Diptera, Tephritidae) in the Stems of Canada Thistle (Cirsium-Arvense). *Canadian Journal of Botany-Revue Canadienne de Botanique* 62 (7):1372-1384, 1984.

R. G. Lalonde and J. D. Shorthouse. Exit Strategy of Urophora-Cardui (Diptera, Tephritidae) from Its Gall on Canada Thistle. *Canadian Entomologist* 114 (9):873-878, 1982.

R. G. Lalonde and J. D. Shorthouse. Growth and Development of Larvae and Galls of Urophora-Cardui (Diptera, Tephritidae) on Cirsium-Arvense (Compositae). *Oecologia* 65 (2):161-165, 1985.

R. G. Lalonde and B. D. Roitberg. Pollen Availability, Seed Production and Seed Predator Clutch Size in A Tephritid Thistle System. *Evolutionary Ecology* 8 (2):188-195, 1994.

S. Lavergne, M. Debussche, and J. D. Thompson. Limitations on reproductive success in endemic Aquilegia viscosa (Ranunculaceae) relative to its widespread congener Aquilegia vulgaris: the interplay of herbivory and pollination. *Oecologia* 142 (2):212-220, 2005.

R. Leimu and K. Syrjanen. Effects of population size, seed predation and plant size on male and female reproductive success in Vincetoxicum hirundinaria (Asclepiadaceae). *Oikos* 98 (2):229-238, 2002.

R. Leimu and K. Lehtila. Effects of two types of herbivores on the population dynamics of a perennial herb. *Basic and Applied Ecology* 7 (3):224-235, 2006.

R. Leimu, K. Syrjanen, J. Ehrlen, and K. Lehtila. Pre-dispersal seed predation in Primula veris: among-population variation in damage intensity and selection on flower number. *Oecologia* 133 (4):510-516, 2002.

S. M. Louda and M. A. Potvin. Effect of inflorescence-feeding insects on the demegraphy and lifetime fitness of a native plant. *Ecology* 76 (1):229-245, 1995.

Svata M. Louda, Martha A. Potvin, and Sharon K. Collinge. Predispersal Seed Predation, Postdispersal Seed Predation and Competition in the Recruitment of Seedlings of a Native Thistle in Sandhills Prairie. *American Midland Naturalist* 124 (1):105-113, 1990.

P. J. W. Lutman. Estimation of seed production by Stellaria media, Sinapis arvensis and Tripleurospermum inodorum in arable crops. *Weed Research* 42 (5):359-369, 2002.

A. L. Mack. Did fleshy fruit pulp evolve as a defence against seed loss rather than as a dispersal mechanism? *Journal of Biosciences* 25 (1):93-97, 2000.

S. Mahoro. Effects of flower and seed predators and pollinators on fruit production in two sequentially flowering congeners. *Plant Ecology* 166 (1):37-48, 2003.

S. Mahoro. Individual flowering schedule, fruit set, and flower and seed predation in Vaccinium hirtum Thunb. (Ericaceae). *Canadian Journal of Botany-Revue Canadienne de Botanique* 80 (1):82-92, 2002.

J. L. Maron, J. K. Combs, and S. M. Louda. Convergent demographic effects of insect attack on related thistles in coastal vs. continental dunes. *Ecology* 83 (12):3382-3392, 2002.

J. M. Marshall and A. J. Storer. Influence of Centaurea biebersteinii patch size on Urophora quadrifasciata (Dipt. Tephritidae) in Michigan, USA. *Journal of Applied Entomology* 130 (2):91-95, 2006.

M. Marshall and F. R. Ganders. Sex-biased seed predation and the maintenance of females in a gynodioecious plant. *American Journal of Botany* 88 (8):1437-1443, 2001.

G. J. Masters, T. H. Jones, and M. Rogers. Host-plant mediated effects of root herbivory on insect seed predators and their parasitoids. *Oecologia* 127 (2):246-250, 2001.

S. J. Mazer and V. A. Delesalle. Floral trait variation in Spergularia marina (Caryophyllaceae): Ontogenetic, maternal family, and population effects. *Heredity* 77:269-281, 1996.

T. R. Meagher, A. C. M. Gillies, and D. E. Costich. Genome size, quantitative genetics and the genomic basis for flower size evolution in Silene latifolia. *Annals of Botany* 95 (1):247-254, 2005.

L. W. Mitich. Oxeye daisy (Chrysanthemum leucanthemum L.), the white-flowered gold flower. *Weed Technology* 14 (3):659-662, 2000.

J. Morrow, L. Scott, B. Congdon, D. Yeates, M. Frommer, and J. Sved. Close genetic similarity between two sympatric species of tephritid fruit fly reproductively isolated by mating time. *Evolution* 54 (3):899-910, 2000.

J. H. Myers and P. Harris. Distribution of Urophora Galls in Flower Heads of Diffuse and Spotted Knapweed in British-Columbia. *Journal of Applied Ecology* 17 (2):359-367, 1980.

J. R. Obeso. The costs of reproduction in plants. *New Phytologist* 155 (3):321-348, 2002.

K. Ohashi and T. Yahara. Effects of flower production and predispersal seed predation on reproduction in Cirsium purpuratum. *Canadian Journal of Botany-Revue Canadienne de Botanique* 78 (2):230-236, 2000.

J. Ollerton and A. Lack. Relationships between flowering phenology, plant size and reproductive success in Lotus corniculatus (Fabaceae). *Plant Ecology* 139 (1):35-47, 1998.

H. Ostergard and J. Ehrlen. Among population variation in specialist and generalist seed predation - the importance of host plant distribution, alternative hosts and environmental variation. *Oikos* 111 (1):39-46, 2005.

C. Petersen. The relationship of inflorescence size to reproductive output and pre-dispersal seed predation in a population of Baptisia leucophaea (Fabaceae). *Article.in Transactions.of Illinois.State Academy.of*, 2000.

D. Pilson. Herbivory and natural selection on flowering phenology in wild sunflower, Helianthus annuus. *Oecologia* 122 (1):72-82, 2000.

B. Pors and P. A. Werner. Individual Flowering Time in A Goldenrod (Solidago-Canadensis) - Field Experiment Shows Genotype More Important Than Environment. *American Journal of Botany* 76 (11):1681-1688, 1989.

S. Raghu, C. Wiltshire, and K. Dhileepan. Intensity of pre-dispersal seed predation in the invasive legume Leucaena leucocephala is limited by the duration of pod retention. *Austral Ecology* 30 (3):310-318, 2005.

T. A. Rand. Variation in insect herbivory across a salt marsh tidal gradient influences plant survival and distribution. *Oecologia* 132 (4):549-558, 2002.

R. J. Reader. Herbivory, Competition, Plant Mortality and Reproduction on A Topographic Gradient in An Abandoned Pasture. *Oikos* 65 (3):414-418, 1992.

T. Reader. Strong interactions between species of phytophagous fly: a case of intraguild kleptoparasitism. *Oikos* 103 (1):101-112, 2003.

J. P. Rieder, E. W. Evans, and S. L. Durham. Distribution of insect attacks in biological control of weeds: Infestation of Centaurea virgata flowerheads by a gall fly. *Biological Control* 20 (3):254-260, 2001.

A. P. Riverolynch and T. H. Jones. The Choice of Oviposition Site by Terellia-Ruficauda on Cirsium-Palustre. *Acta Oecologica-International Journal of Ecology* 14 (5):643-651, 1993.

F. Rose. *The wild flower key*, 1991.

H. Schlumprecht. Dispersal of the Thistle Gallfly Urophora-Cardui and Its Endoparasitoid Eurytoma-Serratulae (Hymenoptera, Eurytomidae). *Ecological Entomology* 14 (3):341-348, 1989.

I. Steffan-Dewenter, U. Munzenberg, and T. Tscharntke. Pollination, seed set and seed predation on a landscape scale. *Proceedings of the Royal Society of London Series B-Biological Sciences* 268 (1477):1685-1690, 2001.

J. M. Story, K. W. Boggs, and W. R. Good. Voltinism and Phenological Synchrony of Urophora-Affinis and U-Quadrifasciata (Diptera, Tephritidae), 2 Seed Head Flies Introduced Against Spotted Knapweed in Montana. *Environmental Entomology* 21 (5):1052-1059, 1992.

N. A. Straw. Taxonomy, Attack Strategies and Host Relations in Flowerhead Tephritidae - A Review. *Ecological Entomology* 14 (4):455-462, 1989a.

N. A. Straw. The Timing of Oviposition and Larval Growth by 2 Tephritid Fly Species in Relation to Host-Plant Development. *Ecological Entomology* 14 (4):443-454, 1989b.

J. J. Sullivan and D. Kelly. Why is mast seeding in Chionochloa rubra (Poaceae) most extreme where seed predation is lowest? *New Zealand Journal of Botany* 38 (2):221-233, 2000.

A. Szentesi and T. Jermy. Pre-dispersal seed predation and seed limitation in an annual legume. *Basic and Applied Ecology* 4 (3):207-218, 2003.

M. van Kleunen and K. Ritland. Predicting evolution of floral traits associated with mating system in a natural plant population. *Journal of Evolutionary Biology* 17 (6):1389-1399, 2004.

R. M. P. van Poecke and M. Dicke. Induced parasitoid attraction by Arabidopsis thaliana: involvement of the octadecanoid and the salicylic acid pathway. *Journal of Experimental Botany* 53 (375):1793-1799, 2002.

G. C. Varley. The Natural Control of Population Balance in the Knapweed Gall-Fly (Urophora jaceana). *The Journal of Animal Ecology* 16 (2):139-187, 1947.

resources Web. BioImages - Virtual Fieldguide (UK). Anonymous. Anonymous. *http.://www.bioimages.org.uk/* , 2003. http://www.bioimages.org.uk/

resources Web. The diptera site. Anonymous. Anonymous. *http.://www.sel.barc.usda.gov./diptera./tephriti./tephri.* 2003. http://www.sel.barc.usda.gov/diptera/diptera.htm

A. Westerbergh and J. Westerbergh. Interactions between seed predators/pollinators and their host plants: a first step towards mutualism? *Oikos* 95 (2):324-334, 2001.

I. M. White. *Tephritid flies*, Royal Entomological Society of London, 1988.

K. D. Whitney and M. L. Stanton. Insect seed predators as novel agents of selection on fruit color. *Ecology* 85 (8):2153-2160, 2004.

B. Widen. Environmental and Genetic Influences on Phenology and Plant Size in A Perennial Herb, Senecio-Integrifolius. *Canadian Journal of Botany-Revue Canadienne de Botanique* 69 (1):209-217, 1991.

I. S. Williams, T. H. Jones, and S. E. Hartley. The role of resources and natural enemies in determining the distribution of an insect herbivore population. *Ecological Entomology* 26 (2):204-211, 2001.

A. C. Worley and S. C. H. Barrett. Evolution of floral display in Eichhornia paniculata (Pontederiaceae): Direct and correlated responses to selection on flower size and number. *Evolution* 54 (5):1533-1545, 2000.

A. C. Worley and S. C. H. Barrett. Evolution of floral display in Eichhornia paniculata (Pontederiaceae): genetic correlations between flower size and number. *Journal of Evolutionary Biology* 14 (3):469-481, 2001.

J. W. Wright and T. R. Meagher. Pollination and seed predation drive flowering phenology in Silene latifolia (Caryophyllaceae). *Ecology* 84 (8):2062-2073, 2003.

H. J. Young. Diurnal and nocturnal pollination of Silene alba (Caryophyllaceae). *American Journal of Botany* 89 (3):433-440, 2002.

H. J. Young, M. L. Stanton, N. C. Ellstrand, and J. M. Clegg. Temporal and Spatial Variation in Heritability and Genetic Correlations Among Floral Traits in Raphanus-Sativus, Wild Radish. *Heredity* 73:298-308, 1994.

M. Zimmerman and A. K. Brody. Choices and consequences of oviposition by Hylemya (Delia) sp. (Diptera : Anthomyiidae). *Journal of Insect Behavior* 11 (3):371-381, 1998.

H. Zwolfer and B. Stadler. The organization of phytophagous guilds in Cardueae flower heads: conclusions from null models. *Evolutionary Ecology Research* 6 (8):1201-1218, 2004.

Chapter 10. Appendix. Mathematical model listing.

```
program Asteraceae;

uses
  Forms,
  Aster in 'Aster.pas' {Mainform},
  Population in 'Population.pas',
  Seedbank in 'Seedbank.pas',
  Functions in 'Functions.pas',
  Predchoice in 'Predchoice.pas',
  LeastSqr in 'Leastsqr.pas',
  About in 'About.pas' {AboutBox};

{$R *.RES}

begin
  Application.Initialize;
  Application.Title := 'Asteraceae';
  Application.CreateForm(TMainform, Mainform);
  Application.CreateForm(TAboutBox, AboutBox);
  Aboutbox.showmodal;
  Aboutbox.timer1.Interval:=10000;
  Application.Run;


  end.
```

```
unit Aster;

interface

uses
  Windows, Messages, SysUtils, Classes, Graphics, Controls, Forms, Dialogs,
  StdCtrls, RZNEdit, ComCtrls, Menus, jpeg, ExtCtrls, Spin;

type
  TMainform = class(TForm)

  MainMenu1: TMainMenu;
     File1: TMenuItem;
     Exit1: TMenuItem;
     Run1: TMenuItem;
     Populate1: TMenuItem;
     Predate1: TMenuItem;
     Pollinate1: TMenuItem;
     Seedbank1: TMenuItem;
     Germinate1: TMenuItem;
     Generate1: TMenuItem;
     Options1: TMenuItem;
     AutoShow: TMenuItem;
     Statistics: TMenuItem;
     Help1: TMenuItem;
     About1: TMenuItem;

  Pagecontrol: TPageControl;

  Base_Sheet: TTabSheet;
   Basebox: TGroupBox;
    Name_label: TLabel;
    Namebox: TEdit;
    PopSize_Label: TLabel;
    Numberofflowers: TIntegerEdit;
    Size_Dev_Label: TLabel;
    Number_Dev: TIntegerEdit;
    Mean_Label: TLabel;
    Mean_size: TRealEdit;
    MeanSDlabel: TLabel;
    MeanSD: TRealEdit;
    Date_Label: TLabel;
    Date: TIntegerEdit;
    Date_Dev_Label: TLabel;
    Devdate: TIntegerEdit;

   Sizebox: TGroupBox;
    Lifetime: TIntegerEdit;
    LifeSD: TIntegerEdit;
    Lifelabel: TLabel;
    LifeSDlabel: TLabel;
    Headlabel: TLabel;
    Headcount: TIntegerEdit;
    Headdev: TIntegerEdit;
    Headdevlabel: TLabel;
```

```
        Sizeheritlabel: TLabel;
        Sizeherit: TIntegerEdit;
        Herit: TIntegerEdit;
        Heritlabel: TLabel;
        Headcost: TRealEdit;
        Headcostlabel: TLabel;
        StDev_Label: TLabel;
        StDev: TRealEdit;

      Flowerbox: TGroupBox;
        Floret_Label: TLabel;
        Floretsize: TRealEdit;
        Maxseedlabel: TLabel;
        MaxSeedEqn: TEdit;
        FloretSD: TRealEdit;
        FloretSDlabel: TLabel;

      Datebox: TGroupBox;
        Inflphensdlabel: TLabel;
        InflphenSD: TIntegerEdit;
        Fertile: TIntegerEdit;
        Phendayslabel: TLabel;
        MinPhen: TIntegerEdit;
        MaxPhen: TIntegerEdit;
        Minphenlabel: TLabel;
        Maxphenlabel: TLabel;
        Phenherit: TIntegerEdit;
        Phenheritlabel: TLabel;

      Seedbox: TGroupBox;
        viability: TIntegerEdit;
        ViabilitySD: TIntegerEdit;
        Viabilitylabel: TLabel;
        ViabilitySDlabel: TLabel;

      SeedSetVar_Box: TGroupBox;
        DayDev_Label: TLabel;
        SetSize_Dev: TIntegerEdit;
        Sizedev_Label: TLabel;
        SetDay_Dev: TIntegerEdit;
        SetHead_DevLabel: TLabel;
        SetHead_Dev: TIntegerEdit;

    Detail_sheet: TTabSheet;
      Predationbox: TGroupBox;
        Predchoicelabel: TLabel;
        Predate_Label: TLabel;
        Prerate_label: TLabel;
        Predation_level: TIntegerEdit;
        Predation_Rate: TIntegerEdit;
        Predlevelbyeqn: TCheckBox;
        Predleveleqn: TEdit;
        Predleveleqnlabel: TLabel;
        Predsizebyeqn: TCheckBox;
        Predsizeeqn: TEdit;
```

```
  Predsizeeqnlabel: TLabel;
  Predratebyeqn: TCheckBox;
  Predrateeqnlabel: TLabel;
  Predrateeqn: TEdit;
  Predation_level_SD: TIntegerEdit;
  Predate_SD_Label: TLabel;
  Predatecalbutton: TButton;
  PredTrackbox: TCheckBox;
Predatebox: TGroupBox;
  X1label: TLabel;
  X1: TRealEdit;
  Y3: TRealEdit;
  Y2: TRealEdit;
  X2: TRealEdit;
  X2Label: TLabel;
  X3Label: TLabel;
  X3: TRealEdit;
  Y1: TRealEdit;
  Y1Label: TLabel;
  Y2Label: TLabel;
  Y3Label: TLabel;

Pollinationbox: TGroupBox;
  Pollchoicelabel: TLabel;
  Pollrate_Label: TLabel;
  Poll_Label: TLabel;
  Pollination_rate: TIntegerEdit;
  Pollination_level: TIntegerEdit;
  Pollsizebyeqn: TCheckBox;
  PollsizeEqn: TEdit;
  Pollsizeeqnlabel: TLabel;
  polllevelbyeqn: TCheckBox;
  pollleveleqnlabel: TLabel;
  pollleveleqn: TEdit;
  pollratebyeqn: TCheckBox;
  pollrateeqnlabel: TLabel;
  pollrateeqn: TEdit;
  Pollination_level_SD: TIntegerEdit;
  Pollinate_SD_Label: TLabel;

Polldatebox: TGroupBox;
  pX1Label: TLabel;
  pX2Label: TLabel;
  pX3Label: TLabel;
  pY1Label: TLabel;
  pY2Label: TLabel;
  pY3Label: TLabel;
  pX1: TRealEdit;
  pY3: TRealEdit;
  pY2: TRealEdit;
  pX2: TRealEdit;
  pX3: TRealEdit;
  pY1: TRealEdit;
  Polldatecalbutton: TButton;
  PollTrackbox: TCheckBox;
```

```
Calculation_Sheet: TTabSheet;
   Data_memo: TMemo;
   Genprogress: TProgressBar;
   Button_Panel: TPanel;
   Populate_Button: TButton;
   Predate_Button: TButton;
   Pollinate_button: TButton;
   Seedbank_button: TButton;
   Germinate_Button: TButton;
   Numberofgenerations: TIntegerEdit;
   Generate_button: TButton;
   Stop_button: TButton;
   Show_Button: TButton;
   Stat_button: TButton;
   Clip_button: TButton;

Viewsheet: TTabSheet;
   View_memo: TMemo;
   Viewtype: TComboBox;
   ViewtypeLabel: TLabel;
   View_Plant: TSpinEdit;
   Viewplantlabel: TLabel;
   View_Button: TButton;
   Viewflowerlabel: TLabel;
   View_Flower: TSpinEdit;
   Viewseedlabel: TLabel;
   View_Seed: TSpinEdit;
   View_All: TCheckBox;

StatusLine: TStatusBar;
  Predslopelabel: TLabel;
  Predsizeslope: TRealEdit;
  Pollslopelabel: TLabel;
  Pollsizeslope: TRealEdit;

  procedure FormCreate(Sender: TObject);
  procedure FormClose(Sender: TObject; var Action: TCloseAction);

  procedure Populate_ButtonClick(Sender: TObject);
  procedure Predate_ButtonClick(Sender: TObject);
  procedure Pollinate_buttonClick(Sender: TObject);
  procedure Seedbank_buttonClick(Sender: TObject);
  procedure Germinate_ButtonClick(Sender: TObject);
  procedure Generate_buttonClick(Sender: TObject);
  procedure Stop_buttonClick(Sender: TObject);
  procedure Show_ButtonClick(Sender: TObject);
  procedure Clip_buttonClick(Sender: TObject);

  procedure Exit1Click(Sender: TObject);
  procedure AutoShowClick(Sender: TObject);
  procedure Stat_buttonClick(Sender: TObject);
  procedure StatisticsClick(Sender: TObject);
  procedure About1Click(Sender: TObject);
  procedure NumberofflowersChange(Sender: TObject);
```

```
    procedure ViewtypeChange(Sender: TObject);
    procedure View_ButtonClick(Sender: TObject);
    procedure ShowHint(Sender: TObject);
    procedure PredatecalbuttonClick(Sender: TObject);
    procedure PolldatecalbuttonClick(Sender: TObject);
    procedure PredsizeslopeChange(Sender: TObject);
    procedure PollsizeslopeChange(Sender: TObject);

  private
    { Private declarations }
  public
    { Public declarations }
  end;

var
  Mainform: TMainform;
  DataChanged,Generating:boolean;

implementation
Uses About,Functions,Parser,Population,Seedbank,Predchoice;
{$R *.DFM}

procedure TMainform.FormCreate(Sender: TObject);
begin
  Application.OnHint := ShowHint;
end;

procedure TMainform.FormClose(Sender: TObject; var Action: TCloseAction);
begin
if Mypopulation<>nil then Mypopulation.kill;
if Thebank<>nil then Thebank.die;
end;

procedure TMainform.Populate_ButtonClick(Sender: TObject);
begin
initialpopulation;
if AutoShow.checked then Show_ButtonClick(Sender);
if Statistics.checked then dostats(0);
end;

procedure TMainform.Predate_ButtonClick(Sender: TObject);
begin
predate;
if AutoShow.checked then Show_ButtonClick(Sender);
if Statistics.checked then dostats(1);
end;

procedure TMainform.Pollinate_buttonClick(Sender: TObject);
begin
pollinate;
if AutoShow.checked then Show_ButtonClick(Sender);
if Statistics.checked then dostats(2);
end;

procedure TMainform.Seedbank_buttonClick(Sender: TObject);
```

```
begin
sow;
if AutoShow.checked then Show_ButtonClick(Sender);
if Statistics.checked then dostats(3);
end;

procedure TMainform.Germinate_ButtonClick(Sender: TObject);
begin
germinate;
if AutoShow.checked then Show_ButtonClick(Sender);
if Statistics.checked then dostats(0);
end;

procedure TMainform.Generate_buttonClick(Sender: TObject);
begin
generate;
end;

procedure TMainform.Stop_buttonClick(Sender: TObject);
begin
Stop_Button.tag:=1;
end;

procedure TMainform.Show_ButtonClick(Sender: TObject);
var i,j,k,noseeds,noset:integer;
begin
  if Mypopulation=nil then begin
   Messagedlg('No population to show',mtError,[mbOk],0);
   exit;
   end;
Data_memo.clear;
Data_memo.lines.add(Mypopulation.name);
Data_memo.lines.add('Pl'+chr(9)+'Fl'+ chr(9) + 'Size' + chr(9) + 'Date' + chr(9) + 'Seeds' + chr(9) +
'Set'+chr(9)+ 'Pred');
for k:=0 to Mypopulation.Plants.Count-1 do
 For i:=0 to aPlant(Mypopulation.Plants.Items[k]).flowers.Count-1 do begin
    Noset:=0;
    Noseeds:=0;
    for j:=0 to aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).seeds.count-1 do
        if
aseed(aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).seeds.items[j]).eaten=false
then inc(Noseeds);
    for j:=0 to aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).seeds.count-1 do
        if aseed(aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).seeds.items[j]).isset
then inc(Noset);
    Data_memo.lines.add(format('%1d',[k+1])+ chr(9)+format('%1d',[i+1])+ chr(9)
            +
format('%5.2f',[aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).actualsize]) +
chr(9)
            +
format('%1.0d',[aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).Actualphenology])
+ chr(9)
            + format('%1.0d',[Noseeds]) + chr(9)
```

```
          +
format('%1.0d',[Noset])+chr(9)+inttostr(byte(aflower(aplant(Mypopulation.Plants.items[k]).Flower
s.items[i]).predated)));
    end;
if Thebank<>nil then begin
  Data_memo.lines.add(format('Seedbank size: ' + '%1.0d',[Thebank.bank.count]));
  Data_memo.lines.add(format('Mean seed size: ' + '%1.2f',[thebank.meansize]));
  end;
end;

procedure TMainform.Stat_buttonClick(Sender: TObject);
begin
  if Mypopulation=nil then begin
   Messagedlg('No population to show',mtError,[mbOk],0);
   exit;
   end;
doStats(0);
end;


procedure TMainform.Clip_buttonClick(Sender: TObject);
begin
Data_memo.Selectall;
Data_memo.copytoclipboard;
end;

procedure TMainform.Exit1Click(Sender: TObject);
begin
close;
end;

procedure TMainform.AutoShowClick(Sender: TObject);
begin
AutoShow.Checked:=AutoShow.Checked=false;
end;


procedure TMainform.StatisticsClick(Sender: TObject);
begin
Statistics.Checked:=Statistics.Checked=false;
end;

procedure TMainform.About1Click(Sender: TObject);
begin
Aboutbox.showmodal;
end;

procedure TMainform.NumberofflowersChange(Sender: TObject);
begin
DataChanged:=true;
end;

procedure TMainform.ViewtypeChange(Sender: TObject);
begin
With Mainform do begin
```

```
  Case Viewtype.ItemIndex of
  0:begin
     View_Plant.MinValue:=0;
     View_Plant.MaxValue:=0;
     View_Plant.Value:=0;
     View_Flower.MinValue:=0;
     View_Flower.MaxValue:=0;
     View_Flower.Value:=0;
     View_Seed.MinValue:=0;
     View_Seed.MaxValue:=0;
     View_Seed.Value:=0;
    end;
  1:begin
     View_Plant.MinValue:=1;
     View_Plant.MaxValue:=Mypopulation.Number;
     View_Plant.Value:=1;
    end;
  2:begin
     View_Flower.MinValue:=1;
     View_Flower.MaxValue:=aPlant(aPopulation(Mypopulation).Plants[View_Plant.Value-
1]).Heads;
     View_Flower.Value:=1;
    end;
  3:begin
     View_Seed.MinValue:=1;
     View_Seed.MaxValue:=aflower(aPlant(aPopulation(Mypopulation).Plants[View_Plant.Value-
1]).Flowers.Items[View_Flower.Value-1]).Seeds.count;
     View_Seed.Value:=1;
    end;
  4:begin
     View_Seed.MinValue:=1;
     View_Seed.MaxValue:=thebank.Bank.Count;
     View_Seed.Value:=1;
    end;
  end;
 end;
View_buttonclick(sender);
end;

procedure TMainform.View_ButtonClick(Sender: TObject);
var i,first,last:integer;
begin
View_memo.clear;
  if Mypopulation=nil then begin
   Messagedlg('No population to show',mtError,[mbOk],0);
   exit;
   end;

Case Viewtype.ItemIndex of
0:begin
  View_memo.lines.add('Population name :  '+Mypopulation.name);
  view_memo.lines.add('Number of plants: '+format('%1d',[Mypopulation.number]));
  view_memo.lines.add('Base size       : '+format('%5.2f',[Mypopulation.Meansize]));
  end;
1:begin
```

```
  if View_All.Checked then begin
    first:=1;
    last:=Mypopulation.Plants.Count;
    View_memo.Lines.Add('Number'+chr(9)+'Size'+chr(9)+'Phen'+chr(9)+'Heads');
    for i:=first to last do
     with aPlant(Mypopulation.Plants.Items[i-1]) do

view_memo.lines.add(format('%1d',[i])+chr(9)+format('%5.2f',[Meansize])+chr(9)+format('%1d',[
Phenology])+chr(9)+format('%1d',[Heads]));
    end
    else
  with aPlant(Mypopulation.Plants.Items[View_Plant.Value-1]) do begin
   view_memo.lines.add('Plant number: '+format('%1d',[View_Plant.Value]));
   view_memo.lines.add('Base size: '+format('%5.2f',[Meansize]));
   view_memo.lines.add('Base Phenology: '+format('%1d',[Phenology]));
   view_memo.lines.add('Flower heads: '+format('%1d',[Heads]));
   end;
  end;
2:begin
  if View_All.Checked then begin
    first:=1;
    last:=aPlant(Mypopulation.Plants.Items[View_Plant.value-1]).Flowers.Count;
    View_memo.Lines.Add('Number'+chr(9)+'Size'+chr(9)+'Phen'+chr(9)+'Seeds');
    for i:=first to last do
     with aFlower(aPlant(Mypopulation.Plants.Items[View_Plant.value-1]).Flowers.Items[i-1]) do

view_memo.lines.add(format('%1d',[i])+chr(9)+format('%5.2f',[Actualsize])+chr(9)+format('%1d',[
ActualPhenology])+chr(9)+format('%1d',[Seeds.Count]));
    end
    else
  with aFlower(aPlant(Mypopulation.Plants.Items[View_Plant.value-
1]).Flowers.Items[View_Flower.value-1]) do begin
   view_memo.lines.add('Flower number: '+format('%1d',[View_Flower.value]));
   view_memo.lines.add('Flower size: '+format('%5.2f',[Actualsize]));
   view_memo.lines.add('Flower Phenology: '+format('%1d',[ActualPhenology]));
   view_memo.lines.add('Seeds: '+format('%1d',[Seeds.Count]));
   end;
  end;
3:begin
  if View_All.Checked then begin
    first:=1;
    last:=aflower(aPlant(aPopulation(Mypopulation).Plants[View_Plant.Value-
1]).Flowers.Items[View_Flower.Value-1]).Seeds.count;
    View_memo.Lines.Add('Number'+chr(9)+'Size'+chr(9)+'Phen');
    for i:=first to last do
     with aseed(aFlower(aPlant(Mypopulation.Plants.Items[View_Plant.value-
1]).Flowers.Items[View_Flower.Value-1]).Seeds.Items[i-1]) do

view_memo.lines.add(format('%1d',[i])+chr(9)+format('%5.2f',[Plantsize])+chr(9)+format('%1d',[P
lantPhen])
      +chr(9)+format('%1d',[Plantheads]));
    end
    else
     with aseed(aFlower(aPlant(Mypopulation.Plants.Items[View_Plant.value-
1]).Flowers.Items[View_Flower.Value-1]).Seeds.Items[View_Seed.Value-1]) do begin
```

```
   view_memo.lines.add('Seed number: '+format('%1d',[View_Seed.value]));
   view_memo.lines.add('Plant size: '+format('%5.2f',[Plantsize]));
   view_memo.lines.add('Plant Headsize SD: '+format('%5.2f',[Plantheadsize_sd]));
   view_memo.lines.add('Plant Phenology: '+format('%1d',[PlantPhen]));
   view_memo.lines.add('Plant Phenology SD: '+format('%1d',[PlantPhen_sd]));
   view_memo.lines.add('Plant Head Phenology SD: '+format('%1d',[PlantheadPhen_sd]));
   view_memo.lines.add('Plant Heads: '+format('%1d',[Plantheads]));
   view_memo.lines.add('Plant Heads SD: '+format('%1d',[Plantheads_sd]));
  end;
  end;
4:begin
  if View_All.Checked then begin
     first:=1;
     last:=thebank.Bank.Count;
     View_memo.Lines.Add('Number'+chr(9)+'Size'+chr(9)+'Phen');
     for i:=first to last do
      with aseed(thebank.Bank.Items[i-1]) do

view_memo.lines.add(format('%1d',[i])+chr(9)+format('%5.2f',[Plantsize])+chr(9)+format('%1d',[P
lantPhen]));
     end
     else
      with aseed(thebank.Bank.Items[View_Seed.Value-1]) do begin
   view_memo.lines.add('Seed number: '+format('%1d',[View_Seed.value]));
   view_memo.lines.add('Plant size: '+format('%5.2f',[Plantsize]));
   view_memo.lines.add('Plant Headsize SD: '+format('%5.2f',[Plantheadsize_sd]));
   view_memo.lines.add('Plant Phenology: '+format('%1d',[PlantPhen]));
   view_memo.lines.add('Plant Phenology SD: '+format('%1d',[PlantPhen_sd]));
   view_memo.lines.add('Plant Head Phenology SD: '+format('%1d',[PlantheadPhen_sd]));
   view_memo.lines.add('Plant Heads: '+format('%1d',[Plantheads]));
   view_memo.lines.add('Plant Heads SD: '+format('%1d',[Plantheads_sd]));


  end;
  end;
 end;
end;

procedure TMainForm.ShowHint(Sender: TObject);

begin
  StatusLine.Panels[0].Text := 'Hint: '+Application.Hint;
end;

procedure TMainform.PredatecalbuttonClick(Sender: TObject);
var x1,x2,x3,y1,y2,y3:real;
    x12,x22,x32:real;
    x2_x1,x3_x1,y2_y1,y3_y1:real;
    x22_x12,x32_x12:real;
    a,b,c:real;
    s:string;
begin
 Predlevelbyeqn.Checked:=false;
 Predleveleqn.Text:='';
 s:='';
```

```
 x1:=Mainform.X1.Value;
 x2:=Mainform.X2.Value;
 x3:=Mainform.X3.Value;
 y1:=Mainform.Y1.Value;
 y2:=Mainform.Y2.Value;
 y3:=Mainform.Y3.Value;
 x12:=x1*x1;
 x22:=x2*x2;
 x32:=x3*x3;
 x2_x1:=x2-x1;
 x3_x1:=x3-x1;
 y2_y1:=y2-y1;
 y3_y1:=y3-y1;
 x22_x12:=x22-x12;
 x32_x12:=x32-x12;
 if (X2>X1) and (X3>X2) then
   begin
    a:=(y3_y1/x3_x1-y2_y1/x2_x1)/(x32_x12/x3_x1-x22_x12/x2_x1);
    b:=y2_y1/x2_x1-a*x22_x12/x2_x1;
    c:=y1-a*x12-b*x1;
    s:=floattostr(a)+'*X1*X1';
    if b>0 then s:=s+'+';
    s:=s+floattostr(b)+'*X1';
    if c>0 then s:=s+'+';
    s:=s+floattostr(c);
    Predlevelbyeqn.Checked:=true;
    Predleveleqn.Text:=s;
   end;
end;

procedure TMainform.PolldatecalbuttonClick(Sender: TObject);
var x1,x2,x3,y1,y2,y3:real;
   x12,x22,x32:real;
   x2_x1,x3_x1,y2_y1,y3_y1:real;
   x22_x12,x32_x12:real;
   a,b,c:real;
   s:string;
begin
 polllevelbyeqn.Checked:=false;
 pollleveleqn.Text:='';
 s:='';
 x1:=Mainform.pX1.Value;
 x2:=Mainform.pX2.Value;
 x3:=Mainform.pX3.Value;
 y1:=Mainform.pY1.Value;
 y2:=Mainform.pY2.Value;
 y3:=Mainform.pY3.Value;
 x12:=x1*x1;
 x22:=x2*x2;
 x32:=x3*x3;
 x2_x1:=x2-x1;
 x3_x1:=x3-x1;
 y2_y1:=y2-y1;
 y3_y1:=y3-y1;
 x22_x12:=x22-x12;
```

```
 x32_x12:=x32-x12;
 if (X2>X1) and (X3>X2) then
   begin
    a:=(y3_y1/x3_x1-y2_y1/x2_x1)/(x32_x12/x3_x1-x22_x12/x2_x1);
    b:=y2_y1/x2_x1-a*x22_x12/x2_x1;
    c:=y1-a*x12-b*x1;
    s:=floattostr(a)+'*X1*X1';
    if b>0 then s:=s+'+';
    s:=s+floattostr(b)+'*X1';
    if c>0 then s:=s+'+';
    s:=s+floattostr(c);
    Polllevelbyeqn.Checked:=true;
    Pollleveleqn.Text:=s;
   end;
end;

procedure TMainform.PredsizeslopeChange(Sender: TObject);
var a,b:double;
   sl,inter:string;
begin
a:=(Predsizeslope.Value-0.29)/0.54;
b:=a-1;
sl:=format('%5.1f',[a]);
inter:=format('%5.1f',[b]);
Predsizeeqn.text:=sl+'*X1 -'+inter;
end;

procedure TMainform.PollsizeslopeChange(Sender: TObject);
var a,b:double;
   sl,inter:string;
begin
a:=(Pollsizeslope.Value-0.29)/0.54;
b:=a-1;
sl:=format('%5.1f',[a]);
inter:=format('%5.1f',[b]);
Pollsizeeqn.text:=sl+'*X1 -'+inter;
end;


begin
DataChanged:=false;
Generating:=false;
end.
```

```
unit Population;

interface

Uses Classes;

type aPlant=Class(Tobject)
       age,diewhen:integer;
       Meansize,Headsize_sd,Florsize:real;
       Phenology,Phenology_sd,Headphenology_sd:integer;
       Heads,heads_sd:integer;
       Flowers:Tlist;
       constructor Create(Size,Head_sd,fl:real;
Phen,Phen_sd,Headphen_sd,Nofl,Nofl_sd,old,agelimit:integer);
       destructor die;
     end;

type aFlower=Class(Tobject)
       Actualsize:real;
       ActualPhenology:integer;
       Seeds:Tlist;
       Predated:boolean;
       Pollinated:boolean;
       Plantsize,Plantheadsize_sd:real;
       Plantphen,Plantphen_sd,Plantheadphen_sd:integer;
       Plantheads,Plantheads_sd:integer;
       constructor  Create(Size,PSize,Pheadsizesd,flsize:real;
Phen,Pphen,Pphensd,Pheadphensd,PHeads,PHeadssd:integer);
       destructor die;
     end;

type aSeed=Class(Tobject)
       Plantsize,Plantheadsize_sd:real;
       Plantphen,Plantphen_sd,Plantheadphen_sd:integer;
       Plantheads,Plantheads_sd:integer;
       IsSet:boolean;
       Eaten:boolean;
       age:integer;
       constructor create(size,headsize_sd:real; phen,phen_sd,headphen_sd,head,head_sd,ag:integer);
       destructor perish;
     end;

type aPopulation=Class(TObject)
      public
       Name:string;
       Number:integer;
       Noflowers:integer;

       Meansize:real;  {data about flower size}
       SizeStDev:real;
       InflowerheadStDev:real;
       Sherit:integer;

       NoHeads:integer; {data about head numbers/plant}
       NoDev:integer;
```

```
      Herit:integer;

      Florsize:real; {floretsize}

      Meandate:integer; {phenology data}
      Devdate:integer;
      Headdatesd:integer;
      Fertility:integer;
      Firstdate:integer;
      Lastdate:integer;
      Pherit:integer;

      Plants:Tlist;
      Meanlife,mlvar:integer;

      constructor create (st:string; m,s,fl,infs:real;
num,d,dd,infdd,NoH,NoD,life,lifevar,sh,ph,h,fert,first,last:integer);
      destructor kill;
      procedure die;
      procedure repopulate(No,Dev:integer);
     end;

Var Mypopulation:aPopulation;

implementation

Uses Math,aster,seedbank,parser;

constructor aSeed.create(size,headsize_sd:real;
                         phen,phen_sd,headphen_sd,
                         head,head_sd,
                         ag:integer);
begin
 Plantsize:={size;}RandG(Size,headsize_sd);
 Plantheadsize_sd:=headsize_sd;
 Plantphen:={Phen}round(RandG(Phen,Phen_sd));
 Plantphen_sd:=Phen_sd;
 Plantheadphen_sd:=headphen_sd;
 Plantheads:={head}round(RandG(head,head_sd));
 Plantheads_sd:=head_sd;
 IsSet:=false;
 Eaten:=false;
 age:=ag;
end;

destructor aSeed.perish;
begin
 inherited destroy;
end;

constructor aFlower.Create(Size,PSize,Pheadsizesd,flsize:real;
Phen,Pphen,Pphensd,Pheadphensd,PHeads,PHeadssd:integer);
var i,no:integer;
    x:real;
begin
```

```
 Actualsize:=size;
 ActualPhenology:=phen;
 Plantsize:=PSize;
 Plantheadsize_sd:=Pheadsizesd;
 Plantphen:=Pphen;
 Plantphen_sd:=Pphensd;
 Plantheadphen_sd:=Pheadphensd;
 Plantheads:=PHeads;
 Plantheads_sd:=PHeadssd;
 Predated:=false;
 Pollinated:=false;
 Seeds:=TList.create;
 ParseX:=actualsize;
 getexp(Mainform.Maxseedeqn.text,x);
 if Mainform.Headcost.Value>0 then x:=x/Plantheads/Mainform.Headcost.Value;
 if flsize=0 then no:=0 else no:=trunc(Power(size/flsize,2)*Min(1.0,X));
 for i:=1 to no do
   Seeds.Add(aSeed.create(Psize,Pheadsizesd,Pphen,Pphensd,Pheadphensd,Pheads,Pheadssd,0));
end;

destructor aFlower.die;
var i:integer;
begin
 for i:=0 to Seeds.count-1 do
   aseed(Seeds.Items[i]).perish;
 Seeds.free;
 inherited destroy;
end;

constructor aPlant.Create(Size,Head_sd,fl:real;
Phen,Phen_sd,Headphen_sd,Nofl,Nofl_sd,old,agelimit:integer);
var i:integer;
begin
 age:=old;
 Meansize:=size;
 Headsize_sd:=Head_sd;
 Phenology:=Phen;
 Phenology_sd:=Phen_sd;
 Headphenology_sd:=Headphen_sd;
 Florsize:=fl;
 if Nofl<1 then Nofl:=1;
 Heads:=Nofl;
 Heads_sd:=Nofl_sd;
 Flowers:=Tlist.create;
 for i:=1 to Nofl do

Flowers.add(aflower.create(RandG(Size,Head_sd),Size,Head_sd,RandG(fl,Mainform.FloretSD.Val
ue),Trunc(RandG(Phen,Headphen_sd)),Phen,Phen_sd,Headphen_sd,Nofl,Nofl_sd));
end;

destructor aplant.die;
var i:integer;
begin
 for i:=0 to Flowers.count-1 do
   aflower(Flowers.items[i]).die;
```

```
 flowers.free;
 inherited destroy;
end;

constructor aPopulation.create (st:string; m,s,fl,infs:real;
num,d,dd,infdd,NoH,NoD,life,lifevar,sh,ph,h,fert,first,last:integer);
var i,j:integer;
begin
 Name:=st;
 Meansize:=m;
 SizeStDev:=s;
 Florsize:=fl;
 InflowerheadStDev:=infs;
 Number:=num;
 Meandate:=d;
 Devdate:=dd;
 Headdatesd:=infdd;
 NoHeads:=NoH;
 NoDev:=NoD;

 Meanlife:=life;
 Mlvar:=lifevar;
 Sherit:=sh;
 Pherit:=ph;
 Herit:=h;
 Plants:=Tlist.create;
 NoFlowers:=0;
 Firstdate:=365;
 Lastdate:=0;

 for i:=1 to Number do begin

Plants.Add(aplant.Create(RandG(m,s),infs,Florsize,trunc(RandG(d,dd)),dd,infdd,trunc(RandG(No
H,NoD)),NoD,Random(life),trunc(RandG(Meanlife,mlvar))));
     with aplant(plants.items[i-1]) do begin
          Noflowers:=Noflowers+Flowers.count;
          for j:=0 to Flowers.count-1 do begin
            if aflower(Flowers.items[j]).ActualPhenology<Firstdate then
firstdate:=aflower(Flowers.items[j]).ActualPhenology;
            if aflower(Flowers.items[j]).ActualPhenology>Lastdate then
Lastdate:=aflower(Flowers.items[j]).ActualPhenology;
            end;
          end;
     end;
end;

destructor aPopulation.kill;
var i:integer;
begin
 for i:=0 to Plants.count-1 do
  aPlant(Plants.Items[i]).die;
 Plants.free;
 Noflowers:=0;
 inherited destroy;
end;
```

```
procedure aPopulation.die;
var i,j:integer;
begin
 i:=Plants.count-1;
 while i>=0 do begin
  aPlant(Plants.Items[i]).age:=aPlant(Plants.Items[i]).age+1;
  if aPlant(Plants.Items[i]).age>=aPlant(Plants.Items[i]).diewhen then begin
      aPlant(Plants.Items[i]).die;
      Plants.delete(i);
      Plants.Capacity:=Plants.Count;
      end;
  dec(i);
  end;
 for i:=1 to Plants.Count do begin
      with aplant(plants.items[i-1]) do begin
          Noflowers:=Noflowers+Flowers.count;
          for j:=0 to Flowers.count-1 do begin
            if aflower(Flowers.items[j]).ActualPhenology<Firstdate then
firstdate:=aflower(Flowers.items[j]).ActualPhenology;
            if aflower(Flowers.items[j]).ActualPhenology>Lastdate then
Lastdate:=aflower(Flowers.items[j]).ActualPhenology;
            end;
          end;
      end;
end;

procedure aPopulation.repopulate(No,Dev:integer);
var i,j,available:integer;
    thisseed:aseed;
begin
 Thisseed:=aseed.create(0,0,0,0,0,0,0,0);
 Number:=Trunc(RandG(No,No*Dev/100));
 available:=0;
 for i:=0 to thebank.bank.count-1 do
    if (aseed(thebank.Bank.items[i]).isset=true) then
     if (aseed(thebank.Bank.items[i]).PlantPhen>=Mainform.Minphen.value) and
        (aseed(thebank.Bank.items[i]).Plantphen<=Mainform.Maxphen.value) then
        inc(available);
 i:=Plants.Count;
 while (i<number) and (available>0) do begin
 j:=random(thebank.bank.count);

thisseed.Plantsize:=RandG(aseed(thebank.bank.items[j]).Plantsize,aseed(thebank.bank.items[j]).Pla
ntsize*Mainform.SetSize_Dev.Value/100);
 thisseed.Plantheadsize_sd:=aseed(thebank.bank.items[j]).Plantheadsize_sd;

thisseed.PlantPhen:=Round(RandG(aseed(thebank.bank.items[j]).Plantphen,Mainform.SetDay_Dev
.Value)+Random(1)/10);
 thisseed.PlantPhen_sd:=aseed(thebank.bank.items[j]).Plantphen_sd;
 thisseed.PlantheadPhen_sd:=aseed(thebank.bank.items[j]).Plantheadphen_sd;

thisseed.PlantHeads:=Round(RandG(aseed(thebank.Bank.items[j]).Plantheads,Mainform.SetHead_
Dev.Value)+Random(1)/10);
 thisseed.PlantHeads_sd:=aseed(thebank.Bank.items[j]).Plantheads_sd;
```

```
 thisseed.isset:=aseed(thebank.Bank.items[j]).isset;
  if (thisseed.PlantPhen>=Mainform.Minphen.value) and
    (thisseed.Plantphen<=Mainform.Maxphen.value) and
    (thisseed.isset) and
    (thisseed.Plantsize>Mypopulation.Florsize) then
  begin
   aseed(thebank.bank.items[j]).isset:=false;
   Plants.add(aplant.create(thisseed.Plantsize,thisseed.Plantheadsize_sd,Mypopulation.Florsize,
                        thisseed.Plantphen,thisseed.Plantphen_sd,thisseed.Plantheadphen_sd,
                        thisseed.Plantheads,thisseed.Plantheads_sd,0,trunc(RandG(Meanlife,mlvar))));
   inc(i);
   dec(available);
  end;
 end;
 Thisseed.perish;
 Noflowers:=0;
 Firstdate:=365;
 Lastdate:=0;
 Meansize:=0;
 Meandate:=0;
 for i:=0 to Plants.count-1 do
      with aplant(plants.items[i]) do begin
          Noflowers:=Noflowers+Flowers.count;
          for j:=0 to Flowers.count-1 do begin
            Mypopulation.Meansize:=Mypopulation.Meansize+aflower(Flowers.items[j]).actualsize;
            Meandate:=Meandate+aflower(Flowers.items[j]).Actualphenology;
            if aflower(Flowers.items[j]).ActualPhenology<Firstdate then
firstdate:=aflower(Flowers.items[j]).ActualPhenology;
            if aflower(Flowers.items[j]).ActualPhenology>Lastdate then
Lastdate:=aflower(Flowers.items[j]).ActualPhenology;
            end;
          end;
  if Noflowers<>0 then begin
     Meansize:=Meansize/Noflowers;
     Meandate:=trunc(Meandate/Noflowers);
     end;
end;

end.
```

```
unit Seedbank;

interface

uses Classes;

type aSeedbank=Class(Tobject)
       Bank:TList;
       Meansize:real;
       Meanphen:real;
       diewhen:integer;
       constructor create(s,sd,hdsvsd:real; p,pd,infphsd,h,hd,l,ld,No,life:integer);
       destructor die;
       procedure fill;
       procedure decimate;
     end;

var Thebank:aseedbank;

implementation

uses Math,Aster,Population;

constructor aSeedbank.create(s,sd,hdsvsd:real;  p,pd,infphsd,h,hd,l,ld,No,life:integer);
var i:integer;
begin
Bank:=Tlist.create;
For i:=1 to No do begin

Bank.Add(aseed.create(RandG(s,sd),sd,hdsvsd,trunc(RandG(p,pd)),pd,infphsd,trunc(RandG(h,hd)),
hd,round(RandG(l,ld))));
       aseed(bank.items[bank.count-1]).isset:=true;
      end;
Meansize:=0;
Meanphen:=0;
diewhen:=life; if diewhen<1 then diewhen:=1;
for i:=0 to Bank.count-1 do begin
  Meansize:=Meansize+aseed(bank.items[i]).Plantsize;
  Meanphen:=Meanphen+aseed(bank.items[i]).Plantphen;
  end;
if Bank.count<>0 then begin
  Meansize:=Meansize/Bank.count;
  Meanphen:=Meanphen/bank.count;
  end;
end;

destructor aSeedbank.die;
var i:integer;
begin
 for i:=0 to Bank.Count-1 do
   aseed(Bank.Items[i]).perish;
 Bank.free;
 inherited destroy;
end;
```

```
procedure aSeedbank.fill;
var i,j,k,Pl,Fl,ss:integer;
begin
Pl:=Mypopulation.Plants.Count;
for k:=0 to pl-1 do begin
 Fl:=aPlant(Mypopulation.Plants.Items[k]).Flowers.count;
 For i:=0 to Fl-1 do begin
  ss:=aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).seeds.count;
  For j:=0 to ss-1 do
    if
(aseed(aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).Seeds.items[j]).isset=true)
and

(aseed(aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).Seeds.items[j]).eaten=false)
       then with
aseed(aflower(aplant(Mypopulation.Plants.items[k]).Flowers.items[i]).Seeds.items[j]) do begin

Bank.Add(aseed.create(Plantsize,Plantsize_sd,Plantheadsize_sd,Plantphen,Plantphen_sd,Planthead
phen_sd,Plantheads,Plantheads_sd,0));
       aseed(bank.items[bank.count-1]).isset:=true;
      end;
  end;
 end;
Meansize:=0;
Meanphen:=0;
for i:=0 to Bank.count-1 do begin
  Meansize:=Meansize+aseed(bank.items[i]).Plantsize;
  Meanphen:=Meanphen+aseed(bank.items[i]).Plantphen;
  end;
if Bank.count<>0 then begin
  Meansize:=Meansize/Bank.count;
  Meanphen:=Meanphen/bank.count;
  end;
end;

procedure aseedbank.decimate;
var i:integer;
begin
 i:=Bank.count-1;
  while i>=0 do begin
  aSeed(Bank.Items[i]).age:=aseed(Bank.Items[i]).age+1;
  if aSeed(Bank.Items[i]).age>=diewhen then begin
      aSeed(Bank.Items[i]).perish;
      Bank.delete(i);
      Bank.Capacity:=Bank.Count;
      end;
  dec(i);
  end;
end;

end.
```

```
unit Predchoice;

interface

Uses Classes;

type pFlower=Class(Tobject)
       Size:real;
       fSize:real;
       pOrigin:integer;
       fOrigin:integer;
       selected:boolean;
       constructor  Create(whichp,whichf:integer);
       destructor die;
     end;

type pSet=Class(TObject)
       Nofertile:integer;
       Setmembers:Tlist;
       Meansize:real;
       Minimum,Maximum:real;
       pMin,pMax:Real;
       Pred:boolean;
       public
       constructor create(Predation:boolean);
       destructor kill;
       procedure transform;
       procedure setfertility(first,last:real);
       function choice(var Plant:integer):integer;
       private
       procedure DoSort;
     end;

var MySet:pSet;

implementation

uses SysUtils,Parser,Population,Aster,functions;

constructor pFlower.Create(whichp,whichf:integer);
begin
 Size:=aFlower(aplant(Mypopulation.Plants.items[whichp]).Flowers.Items[whichf]).Actualsize;
 fSize:=Size;
 pOrigin:=whichp;
 forigin:=whichf;
 Selected:=false;
end;

destructor pFlower.die;
begin
 inherited destroy;
end;


constructor pSet.create(Predation:boolean);
```

```
var i,j,Nop,No:integer;
begin
 Pred:=predation;
 Meansize:=0;
 Nop:=Mypopulation.plants.count;
 Setmembers:=Tlist.create;
 No:=0;
 Nofertile:=0;
 for i:=0 to Nop-1 do
    for j:=0 to aplant(Mypopulation.plants.items[i]).Flowers.count-1 do
      begin
      Setmembers.add(pflower.create(i,j));
      Meansize:=Meansize+pflower(Setmembers.Items[Setmembers.count-1]).Size;
      inc(No);
      end;
 if No=0 then Meansize:=0 else Meansize:=Meansize/No;
end;

destructor pSet.kill;
var i:integer;
begin
 for i:=0 to Setmembers.count-1 do
  pFlower(Setmembers.Items[i]).die;
 Setmembers.free;
 inherited destroy;
end;

procedure pSet.setfertility(first,last:real);
var i,Pl,Fl:integer;
    phen:real;
begin
 Nofertile:=0;
 for i:=0 to Setmembers.count-1 do begin
    Pl:=pFlower(Setmembers.Items[i]).pOrigin;
    Fl:=pFlower(Setmembers.Items[i]).fOrigin;
    phen:=aflower(aplant(Mypopulation.plants.items[Pl]).Flowers.Items[Fl]).Actualphenology;
    if (phen<=last) and (phen>=First) then begin
           pflower(Setmembers.items[i]).Selected:=false;
           inc(Nofertile);
           end
        else pflower(Setmembers.items[i]).Selected:=true;
    end;
end;

Procedure pSet.DoSort;
 procedure sort(number:integer);
  procedure quick(l,r:integer);
  var i,j:integer;
      x:real;
  begin
  i:=l; j:=r;
  x:=pFlower(Setmembers.items[(l+r) div 2]).Size;
  repeat
     while pFlower(Setmembers.items[i]).Size < x do i:=i+1;
     while x < pFlower(Setmembers.items[j]).Size do j:=j-1;
```

```
    if i<=j then
      begin
        Setmembers.Exchange(i,j);
        i:=i+1; j:=j-1;
      end;
  until i>j;
  if l<j then quick(l,j);
  if l<r then quick(i,r);
  end;{quick}
 begin
 quick(0,number-1);
 end;{sort}
begin
    Sort(Setmembers.count);
    Minimum:=pFlower(Setmembers.Items[0]).fsize;
    Maximum:=pFlower(Setmembers.Items[Setmembers.Count-1]).fsize;
    pMin:=pFlower(Setmembers.Items[0]).size;
    pMax:=pFlower(Setmembers.Items[Setmembers.Count-1]).size;
end;

procedure pSet.transform;
var i:integer;
   a:real;
begin
 For i:=0 to Setmembers.count-1 do begin
   if Meansize=0 then a:=1 else
      a:=pflower(Setmembers.Items[i]).Size/Meansize;
      pflower(Setmembers.Items[i]).Size:=a;
   end;
 DoSort;
end;

function pSet.choice(var Plant:integer):integer;
var i,j,Pl,Fl:integer;
   a,b,c,U:real;
   OK:boolean;
begin
 OK:=false; i:=0; j:=0;
 while OK=false do begin
 inc(j);
 i:=Random(Setmembers.Count);
 U:=Random;
 Parsex:=pflower(Setmembers.Items[i]).Size;
 if Pred then getexp(Mainform.Predsizeeqn.text,a)
          else getexp(Mainform.Pollsizeeqn.text,a);
 Parsex:=pMax;
 if Pred then getexp(Mainform.Predsizeeqn.text,b)
          else getexp(Mainform.Pollsizeeqn.text,b);
 Parsex:=pMin;
 if Pred then getexp(Mainform.Predsizeeqn.text,c)
          else getexp(Mainform.Pollsizeeqn.text,c);
 OK:=pflower(Setmembers.items[i]).selected=false;
 {U:=c+U*(b-c);}
 if OK then if j<10000 then OK:=U<a/b;
 end;
```

```
 Fl:=pflower(Setmembers.Items[i]).fOrigin;
 Pl:=pflower(Setmembers.Items[i]).pOrigin;
 if Pl<Mypopulation.Plants.count then begin
    pflower(setmembers.items[i]).selected:=true;
    dec(Nofertile);
    end;
 Plant:=Pl;
 Choice:=Fl;
end;

end.
```

```
unit Functions;

interface
procedure initialpopulation;
procedure predate;
procedure pollinate;
procedure sow;
procedure germinate;
procedure generate;
procedure doStats(sort:integer);

implementation

Uses Forms,Math,dialogs,Sysutils,aster,population,seedbank,predchoice,parser;

procedure doStats(sort:integer);
var i,j,no:integer;
    means,sd,skew,a,b,c,d,meanp,sdp:real;
begin
means:=0;
sd:=0;
skew:=0;
meanp:=0;
sdp:=0;
no:=0;
b:=0;
c:=0;
if sort=3 then
 with thebank do begin
   for i:=0 to bank.count-1 do begin
        inc(no);
        a:=aseed(bank.items[i]).Plantsize;
        d:=aseed(bank.items[i]).Plantphen;
        means:=means+a;
        meanp:=meanp+d;
        b:=b+a*a;
        c:=c+d*d;
        end;
  end
  else
 with Mypopulation do begin
   for i:=0 to Plants.Count-1 do
      for j:=0 to aplant(plants.items[i]).Flowers.Count-1 do begin
        a:=0; d:=0;
        case sort of
        0:begin
          inc(no);
          a:=aflower(aplant(plants.items[i]).flowers.items[j]).actualsize;
          d:=aflower(aplant(plants.items[i]).flowers.items[j]).actualphenology;
         end;
        1:if aflower(aplant(plants.items[i]).flowers.items[j]).Predated then
          begin
          inc(no);
          a:=aflower(aplant(plants.items[i]).flowers.items[j]).actualsize;
          d:=aflower(aplant(plants.items[i]).flowers.items[j]).actualphenology;
```

```
     end;
    2:if aflower(aplant(plants.items[i]).flowers.items[j]).Pollinated then
     begin
     inc(no);
     a:=aflower(aplant(plants.items[i]).flowers.items[j]).actualsize;
     d:=aflower(aplant(plants.items[i]).flowers.items[j]).actualphenology;
     end;
    end;
    means:=means+a;
    meanp:=meanp+d;
    b:=b+a*a;
    c:=c+d*d;
    end;
  end;
if No>1 then begin
 sd:=sqrt((no*b-means*means)/no/(no-1));
 sdp:=sqrt((no*c-meanp*meanp)/no/(no-1));
 means:=means/no;
 meanp:=meanp/no;
 end;
c:=0;
with Mypopulation do begin
  for i:=0 to Plants.Count-1 do
   for j:=0 to aplant(plants.items[i]).Flowers.Count-1 do begin
    a:=aflower(aplant(plants.items[i]).flowers.items[j]).actualsize;
    if sd<>0 then c:=c+Power((a-means)/sd,3);
    end;
  end;
if no>2 then skew:=c*no/(no-1)/(no-2);

With mainform do begin
 Data_memo.lines.add('');
 case sort of
 0:Data_memo.lines.add('Total population');
 1:Data_memo.lines.add('Predated population');
 2:Data_memo.lines.add('Pollinated population');
 3:Data_memo.lines.add('Seedbank');
 end;
 Data_memo.lines.add('Mean'+chr(9)+'St.Dev'+ chr(9) + 'Skew'+chr(9)+'Phen.'+chr(9)+'PhenD');
 Data_memo.lines.add(format('%5.2f',[means]) + chr(9)
         + format('%5.2f',[sd]) + chr(9)
         + format('%5.2f',[skew])+chr(9)
         + format('%5.2f',[meanp]) + chr(9)
         + format('%5.2f',[sdp]));

 end;
end;


procedure initialpopulation;
begin
Randomize;
if Mypopulation<>nil then begin
  Mypopulation.kill;
  Mypopulation:=nil;
```

```
   end;
if Thebank<>nil then begin
   Thebank.die;
   Thebank:=nil;
   end;
with mainform do

Mypopulation:=aPopulation.create(namebox.text,mean_size.value,MeanSD.value,Floretsize.value,
Stdev.value,

trunc(RandG(Numberofflowers.value,Numberofflowers.value*Number_Dev.value/100)),
                        Date.value,Devdate.value,inflphensd.value,Headcount.value,Headdev.value,
                        Lifetime.value,Lifesd.value,Sizeherit.value,Phenherit.value,Herit.Value,
                        Fertile.value,Minphen.value,Maxphen.value);
with mainform do Thebank:=aseedbank.create(mean_size.value,meanSd.value,StDev.value,

date.value,devdate.value,inflphensd.value,headcount.value,headdev.value,

lifetime.value,lifesd.value,Numberofflowers.value*10,round(RandG(viability.value,viabilitysd.valu
e)));
if generating=false then dostats(0);
end;

procedure predate;
var i,j,k:integer;
    Noflowers,NoSeeds,NoPredf,NoPreds,Pl,Fl:integer;
    a,first,last:real;
begin
  if (Mypopulation=nil) or (Mypopulation.NoFlowers=0) then begin
     Messagedlg('No population to predate',mtError,[mbOk],0);
     exit;
     end;
   Myset:=pSet.create(true);
   Myset.transform;
   First:=Mypopulation.Firstdate;
   while first<Mypopulation.Lastdate do begin
      Last:=First+Mainform.fertile.value-1;
      Myset.Setfertility(first,last);
      with Mainform do begin
      Noflowers:=Myset.Nofertile;
      if Predlevelbyeqn.checked then begin
        if PredTrackbox.Checked then ParseX:=(First+Last)/2/Mypopulation.Meandate
           else ParseX:=(First+Last)/2/Mainform.Date.Value;
        getexp(Predleveleqn.text,a);
        If a>100 then a:=100 else if a<0 then a:=0;
        NoPredf:=round(Noflowers*a/100);
        end
      else NoPredf:=Noflowers*Predation_Level.value div 100;
      i:=0;
      Nopredf:=Round(RandG(NoPredf,NoPredf*Predation_level_SD.value div 100));
      while (i<NoPredf) and (Myset.Nofertile>0)  do begin
        if Predsizebyeqn.checked=true then Fl:=Myset.choice(Pl) else
           begin
             Pl:=Random(Mypopulation.Plants.count);
             Fl:=Random(aplant(Mypopulation.Plants.Items[Pl]).flowers.count);
```

```
           end;
        if Pl<Mypopulation.Plants.Count then begin
        if aflower(aplant(Mypopulation.Plants.items[Pl]).Flowers.items[Fl]).Predated=false then
          with aflower(aplant(Mypopulation.Plants.items[Pl]).Flowers.items[Fl]) do
          begin
          Predated:=true;
          if Predsizebyeqn.checked=false then dec(Myset.Nofertile);
          NoSeeds:=Seeds.count;
                if Predratebyeqn.checked then begin
                ParseX:=actualsize/Mypopulation.Meansize;
                getexp(Predrateeqn.text,a);
                If a>100 then a:=100 else if a<0 then a:=0;
                NoPreds:=round(Noseeds*a/100);
                end
                else NoPreds:=Noseeds*Predation_Rate.value div 100;
          j:=0;
          while j<NoPreds do begin
            k:=random(Seeds.count);
            if aseed(Seeds.Items[k]).eaten=false then begin
              aseed(Seeds.Items[k]).eaten:=true;
              inc(j);
              end;
            end;
          inc(i);
          end;
        end;
       end;
      end;
      First:=First+Mainform.fertile.value;
   end;
   Myset.kill;
if generating=false then dostats(0);
end;

procedure pollinate;
var i,j,k,l:integer;
    NoFlowers,NoFlPoll,NoSeeds,NoPolls,sd,fatherp,fatherf,Pl,Fl:integer;
    fs,ms:real;
    fsd,msd:real;
    fhsd,mhsd:real;
    fp,mp:integer;
    fpd,mpd:integer;
    fhpd,mhpd:integer;
    fh,mh:integer;
    fhd,mhd:integer;
    a,first,last:real;
begin
if Mypopulation=nil then begin
   Messagedlg('No population to pollinate',mtError,[mbOk],0);
   exit;
   end;
  Myset:=pSet.create(false);
  Myset.transform;
with Mainform do begin
  First:=Mypopulation.Firstdate;
```

```
 while first<Mypopulation.Lastdate do begin
   Last:=First+fertile.value-1;
   Myset.Setfertility(first,last);
   Noflowers:=Myset.Nofertile;
   if Pollevelbyeqn.checked then begin
     if PollTrackbox.Checked then ParseX:=(First+Last)/2/Mypopulation.Meandate
       else ParseX:=(First+Last)/2;
     getexp(Pollleveleqn.text,a);
     If a>100 then a:=100 else if a<0 then a:=0;
     NoFlPoll:=round(Noflowers*a/100);
     end
   else NoFlPoll:=Noflowers*Pollination_Level.value div 100;
   NoFlPoll:=Round(RandG(NoFlPoll,NoFlPoll*Pollination_level_SD.value div 100));
   i:=0; l:=0;
   while (i<NoFlPoll) and (Myset.Nofertile>0) and (l<10000) do begin
     inc(l);
     if Pollsizebyeqn.checked=true then Fl:=Myset.choice(Pl) else
       begin
         Pl:=Random(Mypopulation.Plants.count);
         Fl:=Random(aplant(Mypopulation.Plants.Items[Pl]).flowers.count);
       end;
     if (pl<Mypopulation.Plants.Count) and
(aflower(aplant(Mypopulation.Plants.items[Pl]).Flowers.items[Fl]).Pollinated=false) then
     with aflower(aplant(Mypopulation.Plants.items[Pl]).Flowers.items[Fl]) do
       begin
       l:=0;
       if Pollsizebyeqn.checked=false then dec(Myset.Nofertile);
       Pollinated:=true;
       NoSeeds:=0;
       for j:=0 to Seeds.count-1 do
         if aseed(seeds.items[j]).eaten=false then inc(Noseeds);
       if Pollratebyeqn.checked then begin
       ParseX:=actualsize/Mypopulation.Meansize;
       getexp(Mainform.Pollrateeqn.text,a);
       If a>100 then a:=100 else if a<0 then a:=0;
       NoPolls:=round(Noseeds*a/100);
       end
       else Nopolls:=Noseeds*Pollination_Rate.value div 100;
       j:=0; k:=0;
       while (j<Nopolls) and (k<10000) do begin
         inc(k);
         sd:=Random(Seeds.count);
         fatherp:=Random(Mypopulation.Plants.Count);
         fatherf:=Random(aplant(Mypopulation.Plants.items[fatherp]).Flowers.Count);
         if (aSeed(Seeds.items[sd]).isset=false) and
           (aSeed(Seeds.items[sd]).eaten=false) and

((aflower(aplant(Mypopulation.Plants.items[fatherp]).Flowers.items[fatherf]).actualphenology>=First-0.5) and

(aflower(aplant(Mypopulation.Plants.items[fatherp]).Flowers.items[fatherf]).actualphenology<=Last+0.5)) then begin
           k:=0;
           fs:=aplant(Mypopulation.Plants.items[fatherp]).meansize;
           ms:=aSeed(Seeds.items[sd]).plantsize;
```

```
fhsd:=aplant(Mypopulation.Plants.items[fatherp]).headsize_sd;
mhsd:=aSeed(Seeds.items[sd]).plantheadsize_sd;
fp:=aplant(Mypopulation.Plants.items[fatherp]).phenology;
mp:=aSeed(Seeds.items[sd]).plantphen;
fpd:=aplant(Mypopulation.Plants.items[fatherp]).phenology_sd;
mpd:=aSeed(Seeds.items[sd]).plantphen_sd;
fhpd:=aplant(Mypopulation.Plants.items[fatherp]).headphenology_sd;
mhpd:=aSeed(Seeds.items[sd]).plantheadphen_sd;
fh:=aplant(Mypopulation.Plants.items[fatherp]).heads;
mh:=aSeed(Seeds.items[sd]).plantheads;
fhd:=aplant(Mypopulation.Plants.items[fatherp]).heads_sd;
mhd:=aSeed(Seeds.items[sd]).plantheads_sd;
case Mypopulation.Sherit of
0:begin
  aSeed(Seeds.Items[sd]).Plantsize:=(fs+ms)/2;
  aSeed(Seeds.Items[sd]).Plantheadsize_sd:=(fhsd+mhsd)/2;
  end;
1:begin
  aSeed(Seeds.Items[sd]).Plantsize:=fs;
  aSeed(Seeds.Items[sd]).Plantheadsize_sd:=fhsd;
  end;
2:begin
  aSeed(Seeds.Items[sd]).Plantsize:=ms;
  aSeed(Seeds.Items[sd]).Plantheadsize_sd:=mhsd;
  end;
end;
case Mypopulation.Pherit of
0:begin
  aSeed(Seeds.Items[sd]).Plantphen:=round(((fp+mp)+Random(1)/10)/2);
  aSeed(Seeds.Items[sd]).Plantphen_sd:=round(((fpd+mpd)+Random(1)/10)/2);
  aSeed(Seeds.Items[sd]).Plantheadphen_sd:=round(((fhpd+mhpd)+Random(1)/10)/2);
  end;
1:begin
  aSeed(Seeds.Items[sd]).Plantphen:=fp;
  aSeed(Seeds.Items[sd]).Plantphen_sd:=fpd;
  aSeed(Seeds.Items[sd]).Plantheadphen_sd:=fhpd;
  end;
2:begin
  aSeed(Seeds.Items[sd]).Plantphen:=mp;
  aSeed(Seeds.Items[sd]).Plantphen_sd:=mpd;
  aSeed(Seeds.Items[sd]).Plantheadphen_sd:=mhpd;
  end;
end;
case Mypopulation.herit of
0:begin
  aSeed(Seeds.Items[sd]).Plantheads:=round(((fh+mh)+Random(1)/10)/2);
  aSeed(Seeds.Items[sd]).Plantheads_sd:=round(((fhd+mhd)+Random(1)/10)/2);
  end;
1:begin
  aSeed(Seeds.Items[sd]).Plantheads:=fh;
  aSeed(Seeds.Items[sd]).Plantheads_sd:=fhd;
  end;
2:begin
  aSeed(Seeds.Items[sd]).Plantheads:=mh;
  aSeed(Seeds.Items[sd]).Plantheads_sd:=mhd;
```

```
        end;
       end;
       aSeed(Seeds.items[sd]).isset:=true;
       inc(j);
       end;
      end;
     inc(i);
     end;
    end;
   First:=First+Mainform.fertile.value;
  end;
 end;
Myset.kill;
if generating=false then dostats(0);
end;

procedure sow;
begin
  if Mypopulation=nil then begin
   Messagedlg('No population to create a seedbank',mtError,[mbOk],0);
   exit;
   end;
  Thebank.fill;
end;

procedure germinate;
var i,j,k:integer;
begin
if Thebank=nil then begin
   Messagedlg('No seedbank to create a population',mtError,[mbOk],0);
   exit;
   end;
Mypopulation.die;
with Mainform do
  Mypopulation.Repopulate(Numberofflowers.Value,Number_Dev.value);
  Thebank.decimate;
for i:=0 to Mypopulation.Plants.count-1 do
  for j:= 0 to aplant(Mypopulation.Plants.Items[i]).flowers.count-1 do
   with aflower(aplant(Mypopulation.Plants.items[i]).Flowers.items[j]) do
    begin
    Predated:=false;
    Pollinated:=false;
    for k:=0 to Seeds.Count-1 do
        begin
         aseed(Seeds.Items[k]).eaten:=false;
         aseed(Seeds.Items[k]).Isset:=false;
        end;
    end;
end;

procedure generate;
var i:integer;
    s:string;
begin
with Mainform do begin
```

```
  if Generating=false then DataChanged:=false;
  Generating:=true;
  Genprogress.Min:=0;
  Genprogress.Max:=Numberofgenerations.Value;
  Data_memo.clear;
  Data_Memo.lines.Add('Gen.'+chr(9)+'Size'+chr(9)+'Phen.'+chr(9)+'Bank'
                          +chr(9)+'Plants'+chr(9)+'Heads');
  if (Mypopulation=nil) or (DataChanged=true) then begin
     initialpopulation;
     DataChanged:=false;
     end;
  for i:=1 to Numberofgenerations.Value do begin
   Application.ProcessMessages;
   predate;
   pollinate;
   sow;
   s:=format('%3d',[i])+chr(9)+format('%1.2f',[Mypopulation.meansize])
                    +chr(9)+format('%1.0f',[thebank.meanphen])
                    +chr(9)+format('%1.0d',[Thebank.bank.count]);
   germinate;

s:=s+chr(9)+format('%3d',[Mypopulation.Plants.count])+chr(9)+format('%3d',[Mypopulation.NoFl
owers]);
   Data_Memo.lines.Add(s);
   Genprogress.position:=i;
   if (Mypopulation.NoFlowers=0) or (Stop_Button.tag=1) then
     begin
     Genprogress.position:=0;
     Stop_Button.tag:=0;
     exit;
     end;
   end;
  Genprogress.position:=0;
  end;
  Generating:=false;
end;

end.
```

```
unit LeastSqr;

{-------------------------------------------------------------------------}
{-                                                   -}
{-    Turbo Pascal Numerical Methods Toolbox                        -}
{-    Copyright (c) 1986, 87 by Borland International, Inc.              -}
{-                                                   -}
{-  This unit provides procedures for modelling data with a function of a   -}
{-  given type, given a set of data points.                          -}
{-                                                   -}
{-------------------------------------------------------------------------}

interface

{  $IFOPT N+
type
  Float = Double; { 8 byte real, requires 8087 math chip

const
  TNNearlyZero = 1E-015;
  $ELSE}
type
  Float = real;   { 6 byte real, no math chip required }

const
  TNNearlyZero = 1E-07;
{   $ENDIF}

  TNRowSize = 10;               { Maximum number of terms  }
                              { in Least Squares fit     }
  TNColumnSize =100000;           { Maximum number of data points  }

type
  TNColumnVector = array[1..TNColumnSize] of Float;
  TNRowVector = array[1..TNRowSize] of Float;
  TNmatrix = array[1..TNColumnSize] of TNRowVector;
  TNSquareMatrix =array[1..TNRowSize] of TNRowVector;
  TNString40 = string[40];
  FitType = (Expo, Fourier, Log, Poly, Power, User);

function ModuleName(Fit : FitType) : TNString40;

procedure LeastSquares(NumPoints         : integer;
                var XData            : TNColumnVector;
                var YData            : TNColumnVector;
                    NumTerms          : integer;
                var Solution          : TNRowVector;
                var YFit             : TNColumnVector;
                var Residuals         : TNColumnVector;
                var StandardDeviation : Float;
                var Variance          : Float;
                var Error            : byte;
                    Fit              : FitType);

{-------------------------------------------------------------------------}
```

```
{-                                                       -}
{-   Input: NumPoints, XData, YData, NumTerms                          -}
{-   Output: Solution, YFit, Residuals, StandardDeviation, Error          -}
{-                                                       -}
{-   Purpose:  Given NumPoints data points of the form (X, Y), this         -}
{-           procedure finds the least square solution of NumTerms terms  -}
{-           (NumTerms <= NumPoints) to then matrix equation AC = B       -}
{-           where A is a NumPoints by NumTerms matrix, B is a            -}
{-           NumPoints vector and C is the least squares solution.  The   -}
{-           elements of A are A[i, j] = Tj(X[i]) where Tj is the jth      -}
{-           basis vector and X[i] is the X-value of the ith data point.  -}
{-           The basis vectors are created by a separate include file,    -}
{-           or module.  The choice of module will determine whether      -}
{-           the least squares solution is a polynomial fit, trigono-     -}
{-           metric fit (Fourier series), power fit (e.g. Y=ax^b, where   -}
{-           b is fractional), exponential fit (e.g. Y=a-Exp(bx)), or     -}
{-           logarithmic fit (e.g. Y=a-Ln(bx)).  The user may also        -}
{-           create modules for other functional forms.  See the          -}
{-           documentation for details.                          -}
{-                                                       -}
{- User-Defined Types: TNColumnVector = array[1..TNColumnSize] of real;      -}
{-                   TNRowVector = array[1..TNRowSize] of real;           -}
{-                                                       -}
{-    Global Variables: NumPoints         : integer; Number of data points -}
{-                  XData            : TNColumnVector; X-value data   -}
{-                  YData            : TNColumnVector; Y-value data   -}
{-                  NumTerms          : integer; Number of terms in    -}
{_                                        least squares fit     -}
{-                  Solution          : TNRowVector; Least squares     -}
{_                                           solution in the   -}
{-                                           given basis       -}
{-                  YFit             : TNColumnVector; Y-values       -}
{-                                            predicted by   -}
{-                                            the LS solution -}
{-                  Residuals         :TNColumnVector Difference       -}
{-                                         between predicted   -}
{-                                         and actual Y values -}
{-                  StandardDeviation : real; Root of variance         -}
{-                  Error             : byte; Indicates an error      -}
{-                                                       -}
{-             Errors: 0: No errors                              -}
{-                  1: NumPoints < 2                               -}
{-                  2: NumTerms < 1                                -}
{-                  3: NumTerms > NumPoints                            -}
{-                  4: solution not possible (exact reason            -}
{-                    will depend upon the particular basis)          -}
{-                                                       -}
{-----------------------------------------------------------------------}

implementation

{    $I c:\tp6\numeric\Least1.inc}       { Include procedure code }
{-----------------------------------------------------------------------}
{-                                                       -}
{-    Turbo Pascal Numerical Methods Toolbox                           -}
```

```
{-    Copyright (c) 1986, 87 by Borland International, Inc.                -}
{-                                                             -}
{------------------------------------------------------------------------}

function ModuleName{(Fit : FitType) : TNString40};
begin
  case Fit of
    Poly    : ModuleName := '       Polynomial Least Squares Fit';
    Fourier : ModuleName := ' Finite Fourier Series Least Squares Fit';
    Power   : ModuleName := '       Power Law Least Squares Fit';
    Expo    : ModuleName := '      Exponential Least Squares Fit';
    Log     : ModuleName := '      Logarithmic Least Squares Fit';
    User    : ModuleName := '  User''s Fit - currently powers of X';
  end;
end; { function ModuleName }

{----------------------------------------------------------------------}
{-                                                            -}
{-            Chebyshev Polynomials                                  -}
{-                                                            -}
{- This module creates basis vectors to find a least squares solution   -}
{- of the form     f(X) = SUM from i=1 to n of (a[i] * Ti[X]), where Ti   -}
{- is the ith Chebyshev polynomial.  The coefficients of the Ti[X] are   -}
{- converted to coefficients of X^(i-1).                              -}
{-                                                            -}
{- The function  ModuleName identifies this as the polynomial fit.       -}
{- The procedure TransformPoly translates and scales the XData to the    -}
{-               interval [-1, 1]. The YData is unchanged.              -}
{- The procedure InverseTransform doesn't do anything in this module.    -}
{- The procedure CreateBasisFunctions creates above basis vectors.       -}
{- The procedure TransformSolution changes the solution vector from      -}
{- coefficients of the Chebyshev polynomials to coefficients of a power  -}
{- series, including adjusting for the shifted data done in TransformPoly-}
{-                                                            -}
{----------------------------------------------------------------------}
procedure PolyTransform(NumPoints  : integer;
                   var XData      : TNColumnVector;
                   var YData      : TNColumnVector;
                   var Multiplier : Float;
                   var Constant   : Float;
                   var WData      : TNColumnVector;
                   var ZData      : TNColumnVector;
                   var Error      : byte);

{------------------------------------------------------------}
{- Input : NumPoints, XData, YData,                          -}
{- Output: WData, ZData, Error                            -}
{-                                                 -}
{- This procedure maps the XData linearly into the interval    -}
{- [-1, 1] returning the transformed data in WData.  The YData -}
{- is passed to ZData unchanged.                          -}
{------------------------------------------------------------}

var
  XDataMin, XDataMax : Float;
```

```
  Row : integer;

begin
  XDataMin := XData[1];
  XDataMax := XData[1];
  for Row := 1 to NumPoints do
  begin
    if XDataMin > XData[Row] then
      XDataMin := XData[Row];
    if XDataMax < XData[Row] then
      XDataMax := XData[Row];
  end;
  Multiplier:=1;
  If XDataMax<>XDataMin then Multiplier := 2.0 / (XDataMax - XDataMin);
  Constant := - Multiplier * (XDataMax + XDataMin) / 2.0;
  for Row := 1 to NumPoints do
    WData[ Row ] := Multiplier * XData[ Row ] + Constant;
  ZDAta := YData;
end; { procedure PolyTransform }

procedure PolyInverseTransform(Multiplier : Float;
                               Constant   : Float;
                           var YFit       : Float);

{-------------------------------------------------}
{- Input: Multiplier, Constant, YFit             -}
{- Output: YFit                                  -}
{-                                               -}
{- This procedure undoes the transformation of   -}
{- the YFit values.  Here, no inverse transform  -}
{- is performed because there was no             -}
{- transformation of Y values in procedure       -}
{- PolyTransform.                                -}
{-------------------------------------------------}

begin
end; { procedure PolyInverseTransform }

procedure PolyCreateBasisFunctions(NumPoints : integer;
                                   NumTerms  : integer;
                               var WData     : TNColumnVector;
                               var Basis     : TNmatrix);

{-------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                   -}
{- Output: Basis                                       -}
{-                                                     -}
{- This procedure creates a matrix of basis vectors.   -}
{- The basis vectors are the CHEBYSHEV POLYNOMIALS.    -}
{-                                                     -}
{- The elements of the matrix are:                     -}
{-     Basis[i, j] = T[j](WData[i])                    -}
{- where T[j](WData[i]) is the jth basis vector        -}
{- evaluated at the value WData[i].                    -}
{-                                                     -}
```

```
{- The vectors are:                              -}
{-   T[1] = 1                                     -}
{-   T[2] = X                                     -}
{-   T[3] = 2x*X - 1                              -}
{-   T[4] = (4x*X - 3)*X                          -}
{-   T[5] = (8x*X - 8)*X*X + 1                    -}
{-   etc.                                         -}
{-                                                -}
{- The Chebyshev Polynomials can be defined recursively: -}
{-   T[1] = 1, T[2] = X                           -}
{-   T[j] = 2x * T[j - 1] - T[j - 2]              -}
{-                                                -}
{-------------------------------------------------------}

var
  Row, Column : integer;

begin
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := 1;
    Basis[Row, 2] := WData[Row];
    for Column := 3 to NumTerms do
      Basis[Row, Column] := 2 * WData[Row] * Basis[Row, Column - 1]
                            - Basis[Row, Column - 2];
  end;
end; { procedure PolyCreateBasisFunctions }

procedure PolyTransformSolution(NumTerms    : integer;
                            var OldSolution : TNRowVector;
                                Multiplier  : Float;
                                Constant    : Float;
                            var NewSolution : TNRowVector);

{-----------------------------------------------------------}
{- Input: NumTerms, OldSolution, Multiplier, Constant      -}
{- Output: NewSolution                                     -}
{-                                                         -}
{- The least squares solution will be more useful if it is -}
{- expressed as a linear expansion of powers of X, rather  -}
{- than as a linear expansion of Chebyshev polynomials.    -}
{-                                                         -}
{- This procedure converts the coefficients of the Chebyshev -}
{- polynomials to coefficients of powers of X.             -}
{- The vectors ConversionVec and OldConversionVec store    -}
{- information about the relationship between these two sets -}
{- of coefficients.  This relationship is defined recursively -}
{- above in the procedure PolyCreateBasisFunctions.        -}
{- The parameters Multiplier and Constant define the linear -}
{- transformation of the XData, so this is accounted for in -}
{- finding the polynomial coefficients.                    -}
{-----------------------------------------------------------}

var
  Index, Term : integer;
```

```
  Sum : Float;
  OldConversionVec, ConversionVec : TNRowVector;

begin
  FillChar(OldConversionVec, SizeOf(OldConversionVec), 0);
  for Index := 1 to NumTerms do
  begin
    Sum := 0;
    if Index > 1 then
      ConversionVec[Index - 1] := 0;
    for Term := Index to NumTerms do
    begin
      if Term = 1 then
        ConversionVec[Term] := 1.0
      else
        if Term = 2 then
          begin
            if Index = 1 then
              ConversionVec[Term] := Constant
            else
              ConversionVec[Term] := Multiplier
          end
        else
          ConversionVec[Term] := 2 * Multiplier * OldConversionVec[Term - 1]
                                 + 2 * Constant * ConversionVec[Term - 1]
                                 - ConversionVec[Term - 2];
      Sum := Sum + ConversionVec[Term] * OldSolution[Term];
    end;
    NewSolution[Index] := Sum;
    OldConversionVec := ConversionVec;
  end;
end; { procedure PolyTransformSolution }

{----------------------------------------------------------------------}
{-                                                                    -}
{-     Fourier series                                                 -}
{-                                                                    -}
{- This module creates basis vectors to find a least squares solution -}
{- of the form:  f(x) = a[0] + SUM from i=1 to n/2 of (a[i] - Cos(ix) + -}
{- a[i+1] - Sin(ix)).  A least squares fit with basis vectors 1, Cos(x), -}
{- Sin(x), Cos(2x), Sin(2x), etc. is made to the data (x, y).        -}
{-                                                                    -}
{- The function ModuleName identifies this as the finite Fourier      -}
{-                      series fit.                                   -}
{- The procedure Transform doesn't do anything in this module.        -}
{- The procedure InverseTransform doesn't do anything in this module. -}
{- The procedure CreateBasisFunctions creates the above basis vectors. -}
{- The procedure TransformSolution doesn't do anything in this module. -}
{- The first element of the solution vector will be the constant,     -}
{- the second element will be the coefficient of Cos(x), the third    -}
{- element will be the coefficient of Sin(x), the fourth element will -}
{- be the coefficient of Cos(2x), etc.                                -}
{-                                                                    -}
{----------------------------------------------------------------------}
```

```
procedure FourierTransform(NumPoints       : integer;
                   var XData          : TNColumnVector;
                   var YData          : TNColumnVector;
                      DummyMultiplier : Float;
                      DummyConstant   : Float;
                   var WData          : TNColumnVector;
                   var ZData          : TNColumnVector;
                   var Error          : byte);

{-------------------------------------------------------------}
{- Input : NumPoints, XData, YData, DummyMultiplier,          -}
{-         DummyConstant                                      -}
{- Output: WData, ZData, Error                                -}
{-                                                            -}
{- No transformations are needed for Fourier Series           -}
{-------------------------------------------------------------}

begin
  WData := XData;
  ZData := YData;
end; { procedure FourierTransform }

procedure FourierInverseTransform(DummyMultiplier : Float;
                              DummyConstant   : Float;
                          var YFit            : Float);

{-------------------------------------------------}
{- Input: DummyMultiplier, DummyConstant, YFit     -}
{- Output: YFit                                  -}
{-                                               -}
{- This procedure undoes the transformation of   -}
{- the YFit values.  Here, no inverse transform  -}
{- is performed because there was no             -}
{- transformation in procedure Transform.        -}
{-------------------------------------------------}

begin
end; { procedure FourierInverseTransform }

procedure FourierCreateBasisFunctions(NumPoints : integer;
                               NumTerms  : integer;
                           var WData     : TNColumnVector;
                           var Basis     : TNmatrix);

{-----------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                        -}
{- Output: Basis                                            -}
{-                                                          -}
{- This procedure creates a matrix of basis vectors.        -}
{- The basis vectors are the FOURIER SERIES.                -}
{-                                                          -}
{- The elements of the matrix are:                          -}
{-     Basis[i, j] = F[j](WData[i])                         -}
{- where F[j](WData[i]) is the jth basis vector             -}
{- evaluated at the value WData[i].                         -}
```

```
{-                                          -}
{- The vectors are:                         -}
{-     F[1] = 1                             -}
{-     F[2] = Cos(x);                       -}
{-     F[3] = Sin(x);                       -}
{-     F[4] = Cos(2x);                      -}
{-     F[5] = Sin(2x);                      -}
{-     F[6] = Cos(3x);                      -}
{-     F[7] = Sin(3x);                      -}
{-     etc.                                 -}
{-                                          -}
{- This series is defined recursively by:                -}
{-     F[1] = 1, F[2] = Cos(x),  F[3] = Sin(x)           -}
{-     F[j] = F[2] - F[j - 2] - F[3] - F[j - 1]  for even j -}
{-     F[j] = F[3] - F[j - 3] + F[2] - F[j - 2]  for odd j  -}
{----------------------------------------------------------}

var
  Row, Column : integer;

begin
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := 1;
    Basis[Row, 2] := Cos(WData[Row]);
    Basis[Row, 3] := Sin(WData[Row]);
    for Column := 4 to NumTerms do
      if Odd(Column) then
        Basis[Row, Column] := Basis[Row, 3] * Basis[Row, Column-3]
                      + Basis[Row, 2] * Basis[Row, Column-2]
      else
        Basis[Row, Column] := Basis[Row, 2] * Basis[Row, Column-2]
                      - Basis[Row, 3] * Basis[Row, Column-1];
  end;
end; { procedure FourierCreateBasisFunctions }


procedure FourierTransformSolution(NumTerms        : integer;
                               var OldSolution     : TNRowVector;
                                   DummyMultiplier : Float;
                                   DummyConstant   : Float;
                               var NewSolution     : TNRowVector);

{--------------------------------------------------}
{- Input: NumTerms, OldSolution, DummyMultiplier, -}
{-        DummyConstant                           -}
{- Output: NewSolution                            -}
{-                                                -}
{- No need to change the coefficients of the      -}
{- Fourier series.                                -}
{--------------------------------------------------}

begin
  NewSolution := OldSolution    { no transformation  }
end; { procedure FourierTransformSolution }
```

```
{------------------------------------------------------------------------}
{-                                                    -}
{-          Y = AX^B                                          -}
{-                                                    -}
{- This module creates basis vectors to find a least squares solution      -}
{- of the form    f(X) = A * X^B.  Taking the logarithm of both sides:     -}
{- Ln(f(X)) = Ln(A) + B * Ln(X).  A linear least squares fit              -}
{- (i.e. with basis vectors Ln(X) and 1) is then made to the data          -}
{- (Ln(X), Ln(Y)). The slope will be B, and the intercept will be Ln(A).   -}
{-                                                    -}
{- The function ModuleName identifies this as the power law fit.           -}
{- The procedure Transform converts the data from (X, Y) to (Ln(X), Ln(Y)). -}
{- The procedure InverseTransform converts from YFit to Exp(YFit)          -}
{- The procedure CreateBasisFunctions creates the vectors Ln(X) and 1.     -}
{- The procedure TransformSolution changes the solution vector from        -}
{- (Ln(A), B) to (A, B).  Therefore, the first coefficient is A,           -}
{- and the second coefficient is B.                             -}
{-                                                    -}
{------------------------------------------------------------------------}

procedure PowerTransform(NumPoints     : integer;
                  var XData        : TNColumnVector;
                  var YData        : TNColumnVector;
                  var Multiplier    : Float;
                     DummyConstant : Float;
                  var WData         : TNColumnVector;
                  var ZData        : TNColumnVector;
                  var Error        : byte);

{-------------------------------------------------------------}
{- Input : NumPoints, XData, YData, Multiplier, DummyConstant  -}
{- Output: WData, ZData, Error                          -}
{-                                           -}
{- This procedure transforms the X and Y values to their      -}
{- logarithms.  A linear least squares fit will then be made  -}
{- to to Ln(Y) = B * Ln(X) + Ln(A). If the Y values are of    -}
{- differing sign, Error = 3 is returned.                -}
{-------------------------------------------------------------}

var
  Index : integer;
  YPoint : Float;

begin
  Index := 0;
  if YData[1] < 0 then
    Multiplier := -1
  else
    Multiplier := 1;
  while (Index < NumPoints) and (Error = 0) do
  begin
    Index := Succ(Index);
    if XData[Index] <= 0 then
      Error := 3
```

```
    else
      begin
        YPoint := Multiplier * YData[Index];
        if YPoint <= 0 then { The data must all have the same sign  }
          Error := 3
        else
          begin
            WData[Index] := Ln(XData[Index]);
            ZData[Index] := Ln(YPoint);
          end;
      end;
  end; { while }
end; { procedure PowerTransform }

procedure PowerInverseTransform(Multiplier    : Float;
                                DummyConstant : Float;
                            var YFit          : Float);

{-----------------------------------------------}
{- Input: Multiplier, DummyConstant, YFit      -}
{- Output: YFit                                -}
{-                                             -}
{- This procedure undoes the transformation of -}
{- the YFit values.  Here, the function        -}
{- YFit := Exp(YFit) is performed to undo the  -}
{- Ln transformation in procedure Transform.   -}
{-----------------------------------------------}

begin
  YFit := Multiplier * Exp(YFit);
end; { procedure PowerInverseTransform }

procedure PowerCreateBasisFunctions(NumPoints : integer;
                                var NumTerms  : integer;
                                var WData     : TNColumnVector;
                                var Basis     : TNmatrix);

{-------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                   -}
{- Output: Basis                                       -}
{-                                                     -}
{- This procedure creates a matrix of basis vectors.   -}
{- The elements of the matrix are:                     -}
{-     Basis[i, j] = C[j](WData[i])                    -}
{- where C[j](WData[i]) is the jth basis vector        -}
{- evaluated at the value WData[i].                    -}
{-                                                     -}
{-                                                     -}
{- The vectors are:                                    -}
{-   C[1] = 1                                          -}
{-   C[2] = X                                          -}
{-------------------------------------------------------}

var
  Row : integer;
```

```
begin
  NumTerms := 2;  { This is only a straight line least squares  }
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := 1;
    Basis[Row, 2] := WData[Row];
  end;
end; { procedure PowerCreateBasisFunctions }

procedure PowerTransformSolution(NumTerms       : integer;
                              var OldSolution   : TNRowVector;
                                  Multiplier    : Float;
                                  DummyConstant : Float;
                              var NewSolution   : TNRowVector);

{------------------------------------------------------------}
{- Input: NumTerms, OldSolution, Multiplier, DummyConstant    -}
{- Output: NewSolution                                      -}
{-                                                          -}
{- The least squares solution will be more useful if it is    -}
{- expressed in terms of Y = AX^B, rather than in terms       -}
{- of Ln(Y) = B * Ln(X) + Ln(A).                            -}
{------------------------------------------------------------}

begin
  NewSolution[1] := Multiplier * Exp(OldSolution[1]);
end; { procedure PowerTransformSolution }

{-----------------------------------------------------------------------}
{-                                                          -}
{-          Y = A * Exp(bx)                                     -}
{-                                                          -}
{- This module creates basis vectors to find a least squares solution   -}
{- of the form    f(X) = A * Exp(bx).  Taking the logarithm of both      -}
{- sides:   Ln(f(X)) = Ln(A) + B * X. A linear least squares fit         -}
{- (i.e. with basis vectors X and 1) is then made to the data            -}
{- (X, Ln(Y)). The slope will be B, and the intercept will be Ln(A).     -}
{-                                                          -}
{- The function ModuleName identifies this as the exponential fit       -}
{- The procedure Transform converts the data from (X, Y) to (X, Ln(Y)).  -}
{- The procedure InverseTransform converts from YFit to Exp(YFit)        -}
{- The procedure CreateBasisFunctions creates the vectors 1 and X.       -}
{- The procedure TransformSolution changes the solution vector from      -}
{- (Ln(A), B) to (A, B).  Therefore, the first coefficient is a,         -}
{- and the second coefficient is B.                                 -}
{-                                                          -}
{-----------------------------------------------------------------------}

procedure ExpoTransform(NumPoints      : integer;
                    var XData          : TNColumnVector;
                    var YData          : TNColumnVector;
                    var Multiplier     : Float;
                        DummyConstant : Float;
                    var WData          : TNColumnVector;
```

```
                    var ZData          : TNColumnVector;
                    var Error         : byte);

{-------------------------------------------------------------}
{- Input : NumPoints, XData, YData, Multiplier, DummyConstant  -}
{- Output: WData, ZData, Error                                -}
{-                                                   -}
{- This procedure transforms the Y values to their            -}
{- logarithms.  A linear least squares fit will then be made   -}
{- to Ln(Y) = bx + Ln(A). If the Y values are of different     -}
{- sign, then Error = 3 is returned.                        -}
{-------------------------------------------------------------}

var
  Index : integer;
  YPoint : Float;

begin
  WData := XData;
  if  YData[1] < 0 then
    Multiplier := -1
  else
    Multiplier := 1;
  Index := 0;
  while (Index < NumPoints) and (Error = 0) do
  begin
    Index := Succ(Index);
    YPoint := Multiplier * YData[Index];
    if YPoint <= 0 then
      Error := 3   { The Y values must all have the same sign }
    else
      ZData[Index] := Ln(YPoint);
  end;
end; { procedure ExpoTransform }

procedure ExpoInverseTransform(Multiplier     : Float;
                               DummyConstant : Float;
                           var YFit          : Float);

{-----------------------------------------------}
{- Input: Multiplier, DummyConstant, YFit        -}
{- Output: YFit                               -}
{-                                       -}
{- This procedure undoes the transformation of   -}
{- the YFit values.  Here, the function          -}
{- YFit := Exp(YFit) is performed to undo the    -}
{- Ln transformation in procedure Transform.     -}
{-----------------------------------------------}

begin
  YFit := Multiplier * Exp(YFit);
end; { procedure ExpoInverseTransform }

procedure ExpoCreateBasisFunctions(NumPoints : integer;
                               var NumTerms  : integer;
```

```
                    var WData     : TNColumnVector;
                    var Basis     : TNmatrix);

{------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                  -}
{- Output: Basis                                      -}
{-                                                    -}
{- This procedure creates a matrix of basis vectors.  -}
{-                                                    -}
{- The elements of the matrix are:                    -}
{-    Basis[i, j] = C[j](WData[i])                    -}
{- where C[j](WData[i]) is the jth basis vector       -}
{- evaluated at the value WData[i].                   -}
{-                                                    -}
{- The vectors are:                                   -}
{-   C[1] = 1                                         -}
{-   C[2] = X                                         -}
{------------------------------------------------------}

var
  Row : integer;

begin
  NumTerms := 2;  { This is only a straight line least squares }
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := 1;
    Basis[Row, 2] := WData[Row];
  end;
end; { procedure ExpoCreateBasisFunctions }

procedure ExpoTransformSolution(NumTerms       : integer;
                      var OldSolution   : TNRowVector;
                          Multiplier    : Float;
                          DummyConstant : Float;
                      var NewSolution   : TNRowVector);

{------------------------------------------------------------}
{- Input: NumTerms, OldSolution, Multiplier, DummyConstant  -}
{- Output: NewSolution                                      -}
{-                                                          -}
{- The least squares solution will be more useful if it is  -}
{- expressed in terms of Y = A - Exp(bx), rather than in    -}
{- terms of Ln(Y) = B - X + Ln(A).                          -}
{------------------------------------------------------------}

begin
  NewSolution[1] := Multiplier * Exp(OldSolution[1]);
end; { procedure ExpoTransformSolution }

{----------------------------------------------------------------------}
{-                                                                    -}
{-           Y = A * Ln(bx)                                           -}
{-                                                                    -}
{- This module creates basis vectors to find a least squares solution -}
```

```
{- of the form    f(X) = A * Ln(bx).  Rewriting the right side of the    -}
{- equation:  f(X) = A * Ln(X) + A * Ln(B).  A linear least squares fit  -}
{- (i.e. with basis vectors Ln(X) and 1) is then made to the data        -}
{- (Ln(X), Y). The slope will be A, and the intercept will be A * Ln(B). -}
{-                                                                       -}
{- The function ModuleName identifies this as the logarithmic fit        -}
{- The procedure Transform converts the data from (X, Y) to (Ln(X), Y).  -}
{- The procedure InverseTransform doesn't do anything in this module.    -}
{- The procedure CreateBasisFunctions creates the vectors Ln(X) and 1.   -}
{- The procedure TransformSolution changes the solution vector from      -}
{- (A, A * Ln(B)) to (A, B).  Therefore, the first coefficient is A,     -}
{- and the second coefficient is B.                                      -}
{-                                                                       -}
{-------------------------------------------------------------------------}

procedure LogTransform(NumPoints      : integer;
                   var XData          : TNColumnVector;
                   var YData          : TNColumnVector;
                   var Multiplier     : Float;
                       DummyConstant  : Float;
                   var WData          : TNColumnVector;
                   var ZData          : TNColumnVector;
                   var Error          : byte);

{-------------------------------------------------------------}
{- Input : NumPoints, XData, YData, Multiplier, DummyConstant  -}
{- Output: WData, ZData, Error                                 -}
{-                                                             -}
{- This procedure transforms the X values to their             -}
{- logarithms.  A linear least squares fit will then be made   -}
{- to Y = ALn(X) + ALn(B).  If the X values are of different   -}
{- sign, then Error = 3 is returned.                           -}
{-------------------------------------------------------------}

var
  Index : integer;
  XPoint : Float;

begin
  ZData := YData;
  if  XData[1] < 0 then
    Multiplier := -1
  else
    Multiplier := 1;
  Index := 0;
  while (Index < NumPoints) and (Error = 0) do
  begin
    Index := Succ(Index);
    XPoint := Multiplier * XData[Index];
    if XPoint <= 0 then
      Error := 3   { The X values must all have the same sign  }
    else
      WData[Index] := Ln(XPoint);
  end;
end; { procedure LogTransform }
```

```
procedure LogInverseTransform(Multiplier    : Float;
                          DummyConstant : Float;
                      var YFit          : Float);

{-------------------------------------------------}
{- Input: Multiplier, DummyConstant, YFit         -}
{- Output: YFit                             -}
{-                                     -}
{- This procedure undoes the transformation of     -}
{- the YFit values.  Here, no inverse transform    -}
{- is performed because the was no transformation  -}
{- in procedure Transform.                   -}
{-------------------------------------------------}

begin
end;{ procedure LogInverseTransform }

procedure LogCreateBasisFunctions(NumPoints : integer;
                          var NumTerms  : integer;
                          var WData     : TNColumnVector;
                          var Basis     : TNmatrix);

{-------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                    -}
{- Output: Basis                               -}
{-                                          -}
{- This procedure creates a matrix of basis vectors.     -}
{-                                          -}
{- The elements of the matrix are:                   -}
{-    Basis[i, j] = C[j](WData[i])                  -}
{- where C[j](WData[i]) is the jth basis vector          -}
{- evaluated at the value WData[i].                 -}
{-                                          -}
{- The vectors are:                            -}
{-   C[1] = X                                -}
{-   C[2] = 1                                -}
{-------------------------------------------------------}

var
  Row : integer;

begin
  NumTerms := 2; { This is only a straight line least squares  }
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := WData[Row];
    Basis[Row, 2] := 1;
  end;
end; { procedure LogCreateBasisFunctions }

procedure LogTransformSolution(NumTerms       : integer;
                         var OldSolution   : TNRowVector;
                             Multiplier    : Float;
                             DummyConstant : Float;
```

```
                    var NewSolution   : TNRowVector);

{----------------------------------------------------------}
{- Input: NumTerms, OldSolution, Multiplier, DummyConstant    -}
{- Output: NewSolution                                   -}
{-                                                       -}
{- The least squares solution will be more useful if it is    -}
{- expressed in terms of Y = A - Ln(bx), rather than in       -}
{- terms of Y = ALn(X) + ALn(B).                          -}
{----------------------------------------------------------}

begin
  NewSolution[2] := Multiplier * Exp(OldSolution[2]/OldSolution[1]);
end; { procedure LogTransformSolution }

{----------------------------------------------------------------------}
{-                                                    -}
{-           User Defined function                            -}
{-                                                    -}
{- This module provides the format for the user to create her own basis  -}
{- vectors.                                              -}
{-                                                    -}
{- The function ModuleName identifies this as the user's Module.         -}
{-   This function should be changed to identify the user's basis.       -}
{- The procedure Transform converts the data from (X, Y) to an           -}
{-   appropriate format (e.g. (Ln(X), Ln(Y)) ). If no transformation     -}
{-   is needed, this procedure should not be changed.                 -}
{- The procedure InverseTransform undoes the transformation of the       -}
{-   Y-coordinate.  In the above example, the procedure would perform    -}
{-   the function YFit := Exp(YFit).  This allows comparison between     -}
{-   least squares approximation and the actual Y-values.              -}
{- The procedure CreateBasisFunctions creates the  basis vectors. The    -}
{-   least squares solution will be coefficients of these basis vectors. -}
{-   Currently the basis vectors are powers of X.                    -}
{- The procedure TransformSolution transforms the solution vector to     -}
{-   an appropriate format.  This usually undoes the transformation made -}
{-   in procedure Transform. If no transformation is needed, this        -}
{-   procedure should not be changed.                              -}
{-                                                    -}
{----------------------------------------------------------------------}

procedure UserTransform(NumPoints       : integer;
                  var XData           : TNColumnVector;
                  var YData           : TNColumnVector;
                  var DummyMultiplier : Float;
                  var DummyConstant   : Float;
                  var WData           : TNColumnVector;
                  var ZData           : TNColumnVector;
                  var Error           : byte);

{----------------------------------------------------------}
{- Input : NumPoints, XData, YData, DummyMultiplier,          -}
{-         DummyConstant                                  -}
{- Output: WData, ZData, Error                               -}
{-                                                       -}
```

```
{- This procedure transforms the input data to an appropriate -}
{- format.  The transformed (or possibly unchanged) data is    -}
{- returned in WData, ZData.                                   -}
{-------------------------------------------------------------}

{var
  Index : integer;}

begin
  WData := XData;     { No transformation  }
  ZData := YData;     { No transformation  }
end; { procedure UserTransform }

procedure UserInverseTransform(DummyMultiplier : Float;
                               DummyConstant   : Float;
                           var YFit            : Float);

{---------------------------------------------------}
{- Input: DummyMultiplier, DummyConstant, YFit     -}
{- Output: YFit                                    -}
{-                                                 -}
{- This procedure undoes the transformation of     -}
{- the YFit values.  No inverse transformation     -}
{- may be necessary.                               -}
{---------------------------------------------------}

begin
end; { procedure UserInverseTransform }

procedure UserCreateBasisFunctions(NumPoints : integer;
                               var NumTerms  : integer;
                               var WData     : TNColumnVector;
                               var Basis     : TNmatrix);

{-------------------------------------------------------}
{- Input: NumPoints, NumTerms, WData                   -}
{- Output: Basis                                       -}
{-                                                     -}
{- This procedure creates a matrix of basis vectors.   -}
{- The user must modify this procedure.                -}
{-                                                     -}
{- The elements of the matrix must be:                 -}
{-    Basis[i, j] = Bj(WData[i])                       -}
{- where Bj(WData[i]) is the jth basis vector evaluated -}
{- at the value WData[i].                              -}
{-                                                     -}
{- For example, the basis vector might be powers of X: -}
{-   B1 = 1                                            -}
{-   B2 = X                                            -}
{-   B3 = X^2                                          -}
{-   B4 = X^3                                          -}
{-   etc.                                              -}
{-                                                     -}
{- These vectors can be defined recursively:           -}
{-   B1 = 1                                            -}
```

```
{-   Bj = X * B[j - 1]                              -}
{------------------------------------------------------}

var
  Row, Column : integer;

begin
  for Row := 1 to NumPoints do
  begin
    Basis[Row, 1] := 1;
    for Column := 2 to NumTerms do
    Basis[Row, Column] := WData[Row] * Basis[Row, Column - 1];
  end;
end; { procedure UserCreateBasisFunctions }

procedure UserTransformSolution(NumTerms        : integer;
                            var OldSolution     : TNRowVector;
                                DummyMultiplier : Float;
                                DummyConstant   : Float;
                            var NewSolution     : TNRowVector);

{------------------------------------------------------------}
{- Input: NumTerms, OldSolution, DummyMultiplier,           -}
{-        DummyConstant                                     -}
{- Output: NewSolution                                      -}
{-                                                          -}
{- This procedure transforms the solution into an appropriate -}
{- form.  The transformed (or possibly unchanged) solution  -}
{- is returned in NewSoluition.                             -}
{------------------------------------------------------------}

begin
  NewSolution := OldSolution; { No transformation  }
end; { procedure UserTransformSolution }

{  $I c:\Least2.inc}
{--------------------------------------------------------------------------}
{-                                                                        -}
{-    Turbo Pascal Numerical Methods Toolbox                              -}
{-    Copyright (c) 1986, 87 by Borland International, Inc.               -}
{-                                                                        -}
{--------------------------------------------------------------------------}

procedure LeastSquares{(NumPoints        : integer;
                   var XData            : TNColumnVector;
                   var YData            : TNColumnVector;
                       NumTerms         : integer;
                   var Solution         : TNRowVector;
                   var YFit             : TNColumnVector;
                   var Residuals        : TNColumnVector;
                   var StandardDeviation : Float;
                   var Variance         : Float;
                   var Error            : byte;
                       Fit              : FitType)};
```

```
var
  WData : TNColumnVector;         { Transformed X-values  }
  ZData : TNColumnVector;         { Transformed Y-values  }
  Basis : TNmatrix;               { Matrix of basis functions  }

  Multiplier : Float;             { Multiplier and Constant are used in }
  Constant : Float;               { some modules to pass information    }
                                  { from Transform to InverseTransform  }
                                  { or TransformSolution.  These must   }
                                  { therefore have dummy parameters     }
                                  { when Multiplier and Constant are    }
                                  { not used.                           }

procedure InitializeAndFormBasisVectors(NumPoints  : integer;
                                    var XData      : TNColumnVector;
                                    var YData      : TNColumnVector;
                                    var Multiplier : Float;
                                    var Constant   : Float;
                                    var WData      : TNColumnVector;
                                    var ZData      : TNColumnVector;
                                    var NumTerms   : integer;
                                    var Solution   : TNRowVector;
                                    var Basis      : TNmatrix;
                                    var Error      : byte);

{----------------------------------------------------}
{- Input: NumPoints, XData, NumTerms                -}
{- Output: Solution, Error                          -}
{-                                                  -}
{- This procedure initializes Solution and Error    -}
{- to zero.  It also checks the data for errors.    -}
{----------------------------------------------------}
begin
  Error := 0;
  if NumPoints < 2 then
    Error := 1;  { Less than 2 data points  }
  if NumTerms < 1 then
    Error := 2;  { Less than 1 coefficient in the fit  }
  if NumTerms > NumPoints then
    Error := 3;  { Number of data points less than        }
                 { number of terms in Least Squares fit!  }
  FillChar(Solution, SizeOf(Solution), 0);
  if Error = 0 then
  begin
    { The next two procedures are particular to each        }
    { basis.  Consequently, they are included in each       }
    { module, not in this include file.                     }

    { The Transform procedure transforms the input data to  }
    { fit the particular basis.  This may mean taking the   }
    { logarithm, or linearly tranforming the data to a      }
    { particular interval. XData is transformed to WData    }
    { and YData is transformed to ZData.  For some of the   }
    { modules, Multiplier and Constant are used to pass     }
    { information, for others they are dummy variables.     }
```

```
    { See the code listing of the appropriate module for    }
    { more information.                                     }
    case Fit of
      Poly    : PolyTransform(NumPoints, XData, YData, Multiplier,
                              Constant, WData, ZData, Error);
      Fourier : FourierTransform(NumPoints, XData, YData, Multiplier,
                                 Constant, WData, ZData, Error);
      Power   : PowerTransform(NumPoints, XData, YData, Multiplier,
                               Constant, WData, ZData, Error);
      Expo    : ExpoTransform(NumPoints, XData, YData, Multiplier,
                              Constant, WData, ZData, Error);
      Log     : LogTransform(NumPoints, XData, YData, Multiplier,
                             Constant, WData, ZData, Error);
      User    : UserTransform(NumPoints, XData, YData, Multiplier,
                              Constant, WData, ZData, Error);
    end;

    if Error = 0 then

      { The CreateBasis procedure creates the matrix of       }
      { basis vectors, Basis.  The elements of Basis are:     }
      { Basis[i, j] = Tj(w[i]) where Tj is the jth basis      }
      { and w[i] is the ith data element of WData.            }
      case Fit of
        Poly    : PolyCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
        Fourier : FourierCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
        Power   : PowerCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
        Expo    : ExpoCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
        Log     : LogCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
        User    : UserCreateBasisFunctions(NumPoints, NumTerms, WData, Basis);
      end;
  end;
end; { procedure InitializeAndFormBasisVectors }

procedure CreateAndSolveEquations(NumPoints : integer;
                                  NumTerms  : integer;
                              var Basis     : TNmatrix;
                              var ZData     : TNColumnVector;
                              var Solution  : TNRowVector;
                              var Error     : byte);


var
  Coefficients : TNSquareMatrix;
  Constants : TNRowVector;

{---------------------------------------------------------}
{- Input: NumPoints, NumTerms, Basis, ZData              -}
{- Output: Solution, Error                               -}
{- This procedure computes and solves the normal equations. -}
{- The normal equations are represented in matrix notation  -}
{- as      Coefficients - Solution = Constants            -}
{- This matrix equation is solved by Gaussian Elimination   -}
{- with partial pivoting (TNToolbox routine: PARTPIVT.INC). -}
{- If no solution exists, Error 3 is returned.            -}
```

```
{----------------------------------------------------------}

procedure ComputeNormalEquations(NumPoints, NumTerms : integer;
                            var Basis            : TNmatrix;
                            var YData            : TNColumnVector;
                            var Coefficients     : TNSquareMatrix;
                            var Constants        : TNRowVector);

{-------------------------------------------------------}
{- Input: NumPoints, NumTerms, Basis, YData            -}
{- Output: Coefficients, Constants                     -}
{-                                                     -}
{- This procedure calculates the normal equations.  The -}
{- normal equations are of the form Ax=b, where A is the -}
{- Coefficients matrix, b is the Constants vector and X  -}
{- is the least squares solution to our problem in the  -}
{- given basis.                                        -}
{- The normal equations are derived from the basis     -}
{- functions and the condition of least squares.  The  -}
{- algorithm to create them is:                        -}
{-     Coefficients[i, j] = Sum from k=1 to NumPoints  -}
{-                          of Basis[k, i]-Basis[k, j] -}
{-                                                     -}
{-    Constants[i] = Sum from k=1 to NumPoints         -}
{-                  YData[k] - Basis[k, i]             -}
{-------------------------------------------------------}

var
  Row, Column, Index : integer;
  Sum : Float;

begin
  for Column := 1 to NumTerms do
  begin
    Sum := 0;
    for Index := 1 to NumPoints do
    Sum := Sum + YData[Index] * Basis[Index, Column];
    Constants[Column] := Sum;
    for Row := Column to NumTerms do
    begin
      Sum := 0;
      for Index := 1 to NumPoints do
        Sum := Sum + Basis[Index, Row] * Basis[Index, Column];
      Coefficients[Row, Column] := Sum;
      Coefficients[Column, Row] := Sum;
    end;
  end;
end; { procedure ComputeNormalEquations }

{-------------------------------------------------------------------}

procedure Partial_Pivoting(Dimen        : integer;
                          Coefficients : TNSquareMatrix;
                          Constants    : TNRowVector;
                      var Solution     : TNRowVector;
```

```
               var Error        : byte);

{-------------------------------------------------------------------------}
{-                                                                       -}
{-          Input: Dimen, Coefficients, Constants                       -}
{-         Output: Solution, Error                                      -}
{-                                                                       -}
{-        Purpose : calculate the solution of a linear set of           -}
{-                  equations using Gaussian elimination, maximal       -}
{-                  pivoting and backwards substitution.                -}
{-                                                                       -}
{-  User-defined Types : TNRowVector = array[1..TNArraySize] of real;   -}
{-               TNSquareMatrix = array[1..TNArraySize] of TNRowVector  -}
{-                                                                       -}
{-  Global Variables : Dimen        : integer;     Dimension of the     -}
{-                                  square matrix                       -}
{-                  Coefficients : TNSquareMatrix; Square matrix        -}
{-                  Constants    : TNRowVector; Constants of            -}
{-                                  each equation                       -}
{-                  Solution     : TNRowVector; Unique solution to      -}
{-                                  the set of equations                -}
{-                  Error        : integer;     Flags if something      -}
{-                                  goes wrong.                         -}
{-                                                                       -}
{-         Errors : 0: No errors;                                       -}
{-                  1: Dimen < 2                                        -}
{-                  2: no solution exists                               -}
{-                                                                       -}
{-------------------------------------------------------------------------}

procedure Initial(Dimen        : integer;
           var Coefficients : TNSquareMatrix;
           var Constants    : TNRowVector;
           var Solution     : TNRowVector;
           var Error        : byte);

{-------------------------------------------------------}
{- Input: Dimen, Coefficients, Constants               -}
{- Output: Solution, Error                             -}
{-                                                     -}
{- This procedure test for errors in the value of Dimen. -}
{- This procedure also finds the solution for the      -}
{- trivial case Dimen = 1.                             -}
{-------------------------------------------------------}

begin
  Error := 0;
  if Dimen < 1 then
    Error := 1
  else
    if Dimen = 1 then
      if ABS(Coefficients[1, 1]) < TNNearlyZero then
        Error := 2
      else
        Solution[1] := Constants[1] / Coefficients[1, 1];
```

```
end; { procedure Initial }

procedure EROswitch(var Row1 : TNRowVector;
                    var Row2 : TNRowVector);

{-----------------------------------------------}
{- Input: Row1, Row2                           -}
{- Output: Row1, Row2                          -}
{-                                             -}
{- Elementary row operation - switching two rows -}
{-----------------------------------------------}

var
  DummyRow : TNRowVector;

begin
  DummyRow := Row1;
  Row1 := Row2;
  Row2 := DummyRow;
end; { procedure EROswitch }

procedure EROmultAdd(Multiplier   : Float;
                         Dimen        : integer;
                     var ReferenceRow : TNRowVector;
                     var ChangingRow  : TNRowVector);

{---------------------------------------------------------}
{- Input: Multiplier, Dimen, ReferenceRow, ChangingRow     -}
{- Output: ChangingRow                                   -}
{-                                                       -}
{- row operation - adding a multiple of one row to another -}
{---------------------------------------------------------}

var
  Term : integer;

begin
  for Term := 1 to Dimen do
    ChangingRow[Term] := ChangingRow[Term] + Multiplier * ReferenceRow[Term];
end; { procedure EROmult&Add }

procedure UpperTriangular(Dimen        : integer;
                      var Coefficients : TNSquareMatrix;
                      var Constants    : TNRowVector;
                      var Error        : byte);

{----------------------------------------------------------------}
{- Input: Dimen, Coefficients, Constants                        -}
{- Output: Coefficients, Constants, Error                       -}
{-                                                              -}
{- This procedure makes the coefficient matrix upper triangular. -}
{- The operations which perform this are also performed on the   -}
{- Constants vector.                                            -}
{- If one of the main diagonal elements of the upper triangular  -}
{- matrix is zero, then the Coefficients matrix is singular and  -}
```

```
{- no solution exists (Error = 2 is returned).                -}
{-------------------------------------------------------------}

var
  Multiplier : Float;
  Row, ReferenceRow : integer;

procedure Pivot(Dimen         : integer;
                ReferenceRow : integer;
            var Coefficients : TNSquareMatrix;
            var Constants    : TNRowVector;
            var Error        : byte);

{-------------------------------------------------------------}
{- Input: Dimen, ReferenceRow, Coefficients                   -}
{- Output: Coefficients, Constants, Error                     -}
{-                                                             -}
{- This procedure searches the ReferenceRow column of the      -}
{- Coefficients matrix for the largest non-zero element below  -}
{- the diagonal. If it finds one, then the procedure switches  -}
{- rows so that the largest non-zero element is on the         -}
{- diagonal. It also switches the corresponding elements in    -}
{- the Constants vector. If it doesn't find a non-zero element, -}
{- the matrix is singular and no solution exists               -}
{- (Error = 2 is returned).                                    -}
{-------------------------------------------------------------}

var
  PivotRow, Row : integer;
  Dummy : Float;

begin
  { First, find the row with the largest element  }
  PivotRow := ReferenceRow;
  for Row := ReferenceRow + 1 to Dimen do
    if ABS(Coefficients[Row, ReferenceRow]) >
       ABS(Coefficients[PivotRow, ReferenceRow]) then
      PivotRow := Row;
  if PivotRow <> ReferenceRow then
    { Second, switch these two rows  }
    begin
      EROswitch(Coefficients[PivotRow], Coefficients[ReferenceRow]);
      Dummy := Constants[PivotRow];
      Constants[PivotRow] := Constants[ReferenceRow];
      Constants[ReferenceRow] := Dummy;
    end
  else
    { If the diagonal element is zero, no solution exists  }
    if ABS(Coefficients[ReferenceRow, ReferenceRow]) < TNNearlyZero then
      Error := 2;     { No solution  }
end; { procedure Pivot }

begin  { procedure UpperTriangular }
  { Make Coefficients matrix upper triangular  }
  ReferenceRow := 0;
```

```
  while (Error = 0) and (ReferenceRow < Dimen - 1) do
  begin
    ReferenceRow := Succ(ReferenceRow);
    { Find row with largest element in this column  }
    { and switch this row with the ReferenceRow     }
    Pivot(Dimen, ReferenceRow, Coefficients, Constants, Error);

    if Error = 0 then
      for Row := ReferenceRow + 1 to Dimen do
        { Make the ReferenceRow element of these rows zero  }
        if ABS(Coefficients[Row, ReferenceRow]) > TNNearlyZero then
        begin
          Multiplier := -Coefficients[Row, ReferenceRow] /
                        Coefficients[ReferenceRow,ReferenceRow];
          EROmultAdd(Multiplier, Dimen,
                     Coefficients[ReferenceRow], Coefficients[Row]);
          Constants[Row] := Constants[Row] +
                            Multiplier * Constants[ReferenceRow];
        end;
  end; { while }
  if ABS(Coefficients[Dimen, Dimen]) < TNNearlyZero then
    Error := 2;    { No solution  }
end; { procedure UpperTriangular }

procedure BackwardsSub(Dimen        : integer;
                   var Coefficients : TNSquareMatrix;
                   var Constants    : TNRowVector;
                   var Solution     : TNRowVector);

{--------------------------------------------------------------}
{- Input: Dimen, Coefficients, Constants                     -}
{- Output: Solution                                          -}
{-                                                           -}
{- This procedure applies backwards substitution to the upper   -}
{- triangular Coefficients matrix and Constants vector. The     -}
{- resulting vector is the solution to the set of equations and -}
{- is stored in the vector Solution.                         -}
{--------------------------------------------------------------}

var
  Term, Row : integer;
  Sum : Float;

begin
  Term := Dimen;
  while Term >= 1 do
  begin
    Sum := 0;
    for Row := Term + 1 to Dimen do
      Sum := Sum + Coefficients[Term, Row] * Solution[Row];
    Solution[Term] := (Constants[Term] - Sum) / Coefficients[Term, Term];
    Term := Pred(Term);
  end;
end; { procedure BackwardsSub }
```

```
begin { procedure Partial_Pivoting }
 Initial(Dimen, Coefficients, Constants, Solution, Error);
 if Dimen > 1 then
 begin
   UpperTriangular(Dimen, Coefficients, Constants, Error);
   if Error = 0 then
     BackwardsSub(Dimen, Coefficients, Constants, Solution);
 end;
end; { procedure Partial_Pivoting }

{-----------------------------------------------------------------------}

begin { procedure CreateAndSolveEquations }

 { The following procedure computes Coefficients and    }
 { Constants of the normal equations.  The exact        }
 { solution to the square system of normal equations    }
 { will be the least squares fit to the data.           }
 ComputeNormalEquations(NumPoints, NumTerms, Basis, ZData,
                        Coefficients, Constants);
 Partial_Pivoting(NumTerms, Coefficients, Constants, Solution, Error);
 if Error = 2 then { Returned from Partial_Pivoting  }
   Error := 4; { No solution  }
end; { procedure CreateAndSolveEquations }

procedure TransformSolutionAndFindResiduals(NumPoints         : integer;
                                              NumTerms          : integer;
                                          var YData             : TNColumnVector;
                                          var Solution          : TNRowVector;
                                              Multiplier        : Float;
                                              Constant          : Float;
                                          var Basis             : TNmatrix;
                                          var YFit              : TNColumnVector;
                                          var Residuals         : TNColumnVector;
                                          var StandardDeviation : Float;
                                          var Variance          : Float);

{----------------------------------------------------------------}
{- Input: NumPoints, NumTerms, YData, Solution, Multiplier,     -}
{-        Constant, Basis                                        -}
{- Output: Solution, YFit, Residuals, StandardDeviation         -}
{-                                                               -}
{- This procedure computes the goodness of fit of the least     -}
{- squares solution.  The residuals and standard deviation of the -}
{- fit are returned.  Also, this procedure transforms the solution -}
{- according to the procedure TransformSolution in the include   -}
{- module.  See the particular module for details on the         -}
{- transformation.                                               -}
{----------------------------------------------------------------}

procedure ComputeYFitAndResiduals(NumPoints         : integer;
                                  NumTerms          : integer;
                                  Multiplier        : Float;
                                  Constant          : Float;
                              var YData             : TNColumnVector;
```

```
                    var Solution          : TNRowVector;
                    var Basis             : TNmatrix;
                    var YFit              : TNColumnVector;
                    var Residuals         : TNColumnVector;
                    var StandardDeviation : Float;
                    var Variance          : Float);

{-----------------------------------------------------------}
{- Input: NumPoints, NumTerms,                              -}
{- Multiplier, Constant, YData, Solution, Basis             -}
{- Output: YFit, Residuals, StandardDeviation               -}
{-                                                          -}
{- This procedure computes the value of the least squares   -}
{- approximation at the data points, WData.  The difference -}
{- between the approximation and the actual values are also -}
{- computed and are returned in the variable Residuals.  The -}
{- standard deviation is calculated with the formula:       -}
{-                                                          -}
{-     StandardDeviation = SQRT(Variance)                   -}
{-                                                          -}
{-                                2                         -}
{-     Variance = SUM(YData[i] - YFit[i]) /                 -}
{-                      (degrees of freedom)                -}
{-                                                          -}
{-     degrees of freedom = NumPoints - NumTerms - 2        -}
{-                                                          -}
{-----------------------------------------------------------}

var
  Index, Term : integer;
  Sum : Float;

begin
  Sum := 0;
  for Index := 1 to NumPoints do
  begin
    YFit[Index] := 0;
    for Term := 1 to NumTerms do
      YFit[Index] := YFit[Index] + Solution[Term]*Basis[Index, Term];

    { The next procedure undoes the transformation of    }
    { the YFit values.  For example, if ZData=Ln(YData)  }
    { then InverseTransform performs the function        }
    { YFit[Index] := Exp(YFit[Index]) so that YFit may   }
    { be compared to YData.                              }
    case Fit of
      Poly    : PolyInverseTransform(Multiplier, Constant, YFit[Index]);
      Fourier : FourierInverseTransform(Multiplier, Constant, YFit[Index]);
      Power   : PowerInverseTransform(Multiplier, Constant, YFit[Index]);
      Expo    : ExpoInverseTransform(Multiplier, Constant, YFit[Index]);
      Log     : LogInverseTransform(Multiplier, Constant, YFit[Index]);
      User    : UserInverseTransform(Multiplier, Constant, YFit[Index]);
    end;
    Residuals[Index] := YFit[Index] - YData[Index];
    Sum := Sum + Sqr(Residuals[Index]);
```

```
  end;
  Variance := Sum;
  if NumPoints = NumTerms then
    StandardDeviation := 0
  else
    StandardDeviation := Sqrt(Sum/(NumPoints - NumTerms));
end; { procedure ComputeYFitAndResiduals }

begin  { procedure TransformSolutionAndFindResiduals }
  ComputeYFitAndResiduals(NumPoints, NumTerms, Multiplier, Constant,
                          YData, Solution, Basis, YFit,
                          Residuals, StandardDeviation, Variance);
  case Fit of
    Poly    : PolyTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
    Fourier : FourierTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
    Power   : PowerTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
    Expo    : ExpoTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
    Log     : LogTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
    User    : UserTransformSolution(NumTerms, Solution, Multiplier, Constant,
              Solution);
  end;
end; { procedure TransformSolutionAndFindResiduals }

begin { procedure LeastSquares }
  InitializeAndFormBasisVectors(NumPoints, XData, YData,
                                Multiplier, Constant, WData, ZData,
                                NumTerms, Solution, Basis, Error);
  if Error = 0  then
    CreateAndSolveEquations(NumPoints, NumTerms, Basis, ZData,
                            Solution, Error);

  if Error = 0 then
    TransformSolutionAndFindResiduals(NumPoints, NumTerms, YData, Solution,
                                      Multiplier, Constant, Basis, YFit,
                                      Residuals, StandardDeviation, Variance);

end; { procedure LeastSquares }

end.
```

```
unit About;

interface

uses Windows, SysUtils, Classes, Graphics, Forms, Controls, StdCtrls,
  Buttons, ExtCtrls, jpeg;

type
  TAboutBox = class(TForm)
    Panel1: TPanel;
    ProgramIcon: TImage;
    Panel2: TPanel;
    ProductName: TLabel;
    Version: TLabel;
    Comments: TLabel;
    Timer1: TTimer;
    Author: TLabel;
    procedure ProgramIconClick(Sender: TObject);
  private
    { Private declarations }
  public
    { Public declarations }
  end;

var
  AboutBox: TAboutBox;

implementation

{$R *.DFM}

procedure TAboutBox.ProgramIconClick(Sender: TObject);
begin
Close;
end;

end.
```