

Factors affecting components of fitness in a gall-making wasp (*Cynips divisa* Hartig)

T.A. Sitch, D.A. Grewcock, and F.S. Gilbert

Department of Zoology, Nottingham University, Nottingham NG7 2RD, UK

Summary. The galls of the agamic generation of the cynipid gall-wasp *Cynips divisa* were studied; these galls are found only on the main veins of the leaf. Galls are concentrated on the middle veins of leaves, and generally occur closer to the midrib than to the edge of the leaf. Survival of the gall-maker and fecundity of the emerging wasp depend on wasp size and position of the gall. There is a striking pattern in gall size according to the number of galls on the vein. The number of galls on a vein appears therefore to have a strong effect on potential fecundity.

Key words: Galls – Oaks – Parasitoid – Fecundity

The commonest interaction in Nature is the tripartite one between plants, their insect herbivores, and parasitoids of the latter. It is therefore one of the most important systems to understand in both applied and theoretical contexts (Price et al. 1980). Herbivores are subject to selection pressures from both defensive mechanisms of the plant and attack by parasitoids: what determines their fitness under these conditions? We study this question in the cynipine gall wasp, *Cynips divisa*. The galls of this wasp provide a superb opportunity for testing many hypotheses about the interaction of the gall maker with both its parasites and its host plant. This paper records the patterns observed in the size and distribution of galls and their parasites.

Choice of habitat (in this case, microhabitat of the oviposition site) is a vital component of realized fitness; this study shows that it plays an important role in the determination of fitness in *Cynips*. Other gall systems have provided good evidence for the assessment of habitats in terms of their likely fitness payoff. For example, in the absence of predation pressure, galling aphids (*Pemphigus betae*) adjust their settling densities so as to equalise average fitnesses across leaves of different sizes; leaves of different sizes give gross differences in fitness if only a single gall is allowed to settle (Whitham 1980). Aphid stem mothers fight for possession of a basal position at the time of settling, since within leaves the basal position results in the highest fitness (Whitham 1986).

In a study of cynipid galls, Taper et al. (1986) concluded that the most important factor in the successful emergence of *Dryocosmus dubiosus* was the level of fungal infestation,

which was itself negatively correlated with leaf tannin levels; thus it appears that leaf tannins protect the gall maker from fungal attack. This pattern also occurs at a different taxonomic scale, where cynipid species diversity is correlated positively with average tannin levels in different oak species (Taper & Case 1987). Taper et al. (1986) interpret this as evidence that cynipids take advantage of a protective role of tannins against fungal attack, but also possibly against their parasitoids or generalised leaf herbivores. However, the defensive role of tannins is controversial, since although they may be generalised protein-precipitating 'quantitative defences' (Feeny 1976), other evidence points to their being highly specific poisons (Bernays 1981; Zucker 1983).

In an evolutionary context, the role of parasitoids in the local population dynamics of their host has been studied in a cynipid gall wasp, *Xanthoteras politum* (Washburn and Cornell 1981). This study concluded that on a local scale parasitoids overexploited their host population, leading to extinction. Askew (1961) suggested that the alternation of generations seen in cynipid gall-makers is a consequence of parasitoid pressure. The host plant is also subject to evolutionary pressures from gall-makers (McCrea & Abrahamson 1985), and it is possible that the 250 to 450% increase in seed production in insecticide-sprayed oaks recorded by Crawley (1985) is entirely due to the almost total eradication of galls.

Methods

Gall wasps (Hymenoptera, Cynipoidea, Cynipinae) induce the growth of galls on a restricted range of plant species (*Quercus*, Rosaceae, Labiatae, Compositae and a few others) but by far the greatest number occur on oaks (75% of European cynipines, 88% of American species: Askew 1985). Askew (1961, 1980, 1985) has opened up the system to study by his detailed work on gall development, identifying the inhabitants, and working out food webs in galls of different species.

In 1985, leaves were collected from oak trees near Clifton, Nottingham. In 1986, collections of oak leaves were taken in autumn from oaks in Clumber Park, Nottinghamshire. The incidence of galling is very patchy, with many oaks having no *C. divisa* galls at all on their leaves. Within the tree, galls are also patchily distributed, occurring on some branches but not others. Samples were taken only from branches with galls.

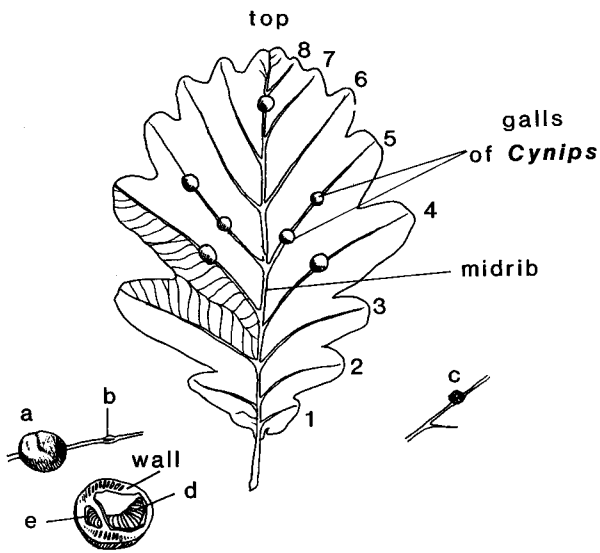


Fig. 1. Oak leaf, showing the recording scheme adopted for numbering the veins, and the gall phenotypes. Letters denote: a normal pea gall; b scar indicating a failed attempt at producing a gall, or a gall that has been knocked off; c aborted gall; d *Synergus nervosus* in the main gall chamber; e *Synergus pallicornis* in a secondary chamber in the wall

Like many cynipine species, *C. divisa* has heterogonic reproduction, i.e. an alternation between parthenogenetic and sexual generations, and each generation produces a different type of gall on oak. The parthenogenetic generation emerges from pea galls on the leaf veins of *Quercus robur* in the winter; females oviposit, and the resulting larvae produce red wart galls on the buds, which give rise to the sexual generation emerging in May. Parthenogenetic females emerge with a fully developed egg complement ready to lay, with no sign of a germarium or of any developing eggs in the ovarioles (Gilbert, unpublished work). Thus counting these will estimate the potential maximum fitness of individual females. This evades the complications of mating in the sexual generation, and enables a simple measure of maximum reproductive fitness, namely the number of eggs in the abdomen at emergence.

Galls were measured and their position on the leaf recorded according to the system shown in Fig. 1; then they were opened using a scalpel, and the contents recorded. During the autumn and early winter, adult female wasps emerge. When the gall is opened, therefore, the adult female is inside waiting to emerge, if she has escaped death from parasitism or other causes. Parasites are usually in the larval stages at this time of the year, and can be identified to genus using Askew's (1985) key. It is possible to identify some of the inhabitants at this time of the year to species (Askew, unpublished work). Thus *Synergus* inquilines in the main chamber of the gall are *S. nervosus* Hartig, whilst if in a small secondary chamber in the gall wall they are probably *S. pallicornis* Hartig; larvae identified as *Torymus* are likely to be *Torymus (Syntomaspis) cyaneus* Walker, but could be either of two other species; *Eurytoma* are *Eurytoma (Eurytoma) bruniventris* Ratzeburg; the identity of *Mesopobolus* is more uncertain, and no specific name can be assigned. Other causes of mortality include fungal attack, recognisable from the presence of fungal hyphae in the gall.

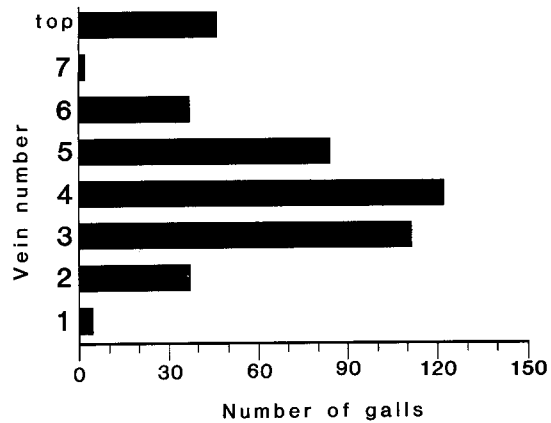


Fig. 2. Number of galls on the different veins of the leaf

Results

1. Distribution of galls

a) *Across leaves.* Galls occurred singly or multiply on leaves, with up to 27 galls on a single leaf in the samples, and even higher densities can sometimes be observed. The number of galls on a leaf is significantly correlated with leaf size ($r=0.4$, $n=43$, $P<0.01$).

b) *Within leaves.* Within a leaf, galls occur only on the primary veins running out to the lobes of the leaf. Galls also occur on the midrib, but almost exclusively on the apical part ('top') where to all intents and purposes it is identical to a primary vein (Fig. 1).

There is no difference in the distribution of galls on veins on either side of the midrib (Kolmogorov-Smirnov test, $D=0.07$, n.s.), but galls are not evenly distributed on the different veins (Fig. 2). If we assume random oviposition, we might expect that the number of galls on a vein is simply proportional to vein length; thus vein lengths were used to generate an expected distribution of galls: the observed distribution is highly significantly different from this expected pattern ($D=0.15$, $P\leq 0.001$) with galls occurring more often on veins 3 and 4 and less often on the others than expected on the basis of their length.

Galls are also generally closer to the midrib than to the leaf edge (Fig. 3), with 78% of galls being positioned less than halfway along the vein from the midrib.

There is no difference in the distribution of single-, double-, triple- or quadruple-gall veins on the different veins of the leaf (chi-squared = 24.2, d.f. = 18, $P=0.15$).

2. What influences gall diameter?

Some parasites may influence gall size (see below), but here we analyse size without reference to gall contents.

Different leaves produce different mean gall sizes ($F_{42,383}=2.31$, $P\leq 0.001$), but this may not be due to density effects. There is no significant effect of leaf gall density on mean gall size, nor on the size of the first gall on any vein, nor on gall size on veins with only a single gall. Analysis of variance demonstrates no effect of vein number on gall size (for veins 3, 4 and 5: $F_{2,298}=0.17$, n.s.), but a very significant effect of gall order on the vein on a multiply-galled vein ($F_{2,298}=5.97$, $P<0.003$). Thus we suggest that for gall sizes, differences between leaves are likely to be due both to different leaf chemistry (not assessed here)

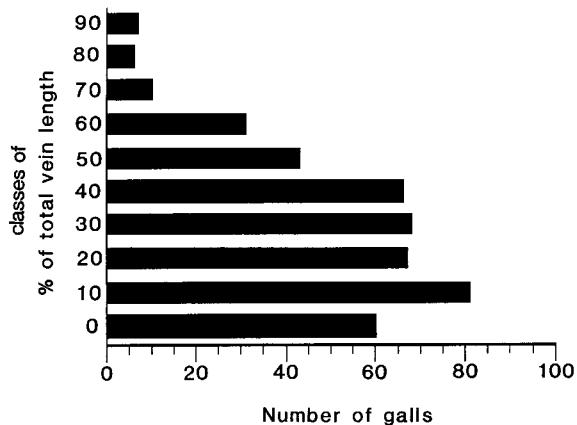


Fig. 3. Number of galls at various distances from the midrib; distances are converted to percentages of the total vein length, to allow for veins of different lengths

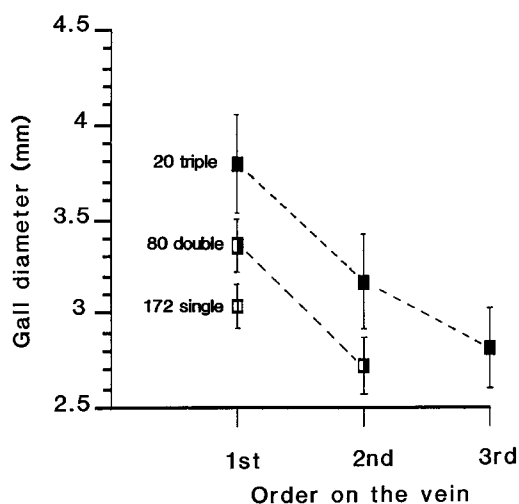


Fig. 4. Pattern of gall size according to the number of galls and their order on a single vein. Numbers next to the points indicate the sample size upon which the data are based

and to the fact that some leaves have galls dispersed singly on veins, whereas others have them clustered multiply on veins.

Figure 4 shows the striking pattern of gall diameter and order on veins with different number of galls. On a 3-gall vein, the first gall (closest the midrib) is larger than the second, which in turn is larger than the third. On a 2-gall vein the same pattern holds, with the first larger than the second. Considering only the first gall, the more companions it has on the vein, the larger it is. A two-way analysis of variance on the first two galls on 2- and 3- gall veins shows a highly significant effect of order ($F_{1,197} = 14.3$, $P < 0.001$) and a significant effect of vein gall-number ($F_{1,197} = 4.35$, $P < 0.05$). This effect of gall order is not due to the influence of distance from the midrib: in a multiple regression of gall diameter (dependent) with order and distance (independent variables), only order has a significant effect.

3. How do parasites distribute themselves in galls?

No differences in parasitism rates were found in relation to leaf categories of low (0-5), medium (6-10) or high gall (> 10) densities per leaf (chi-square = 5.2, d.f. = 2, $P > 0.05$). The total number of parasites in a single gall depends on

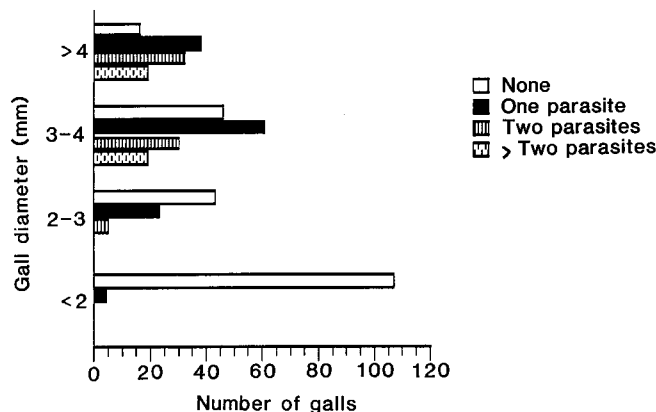


Fig. 5. Total number of parasites in galls of different sizes

gall diameter, since more are found in larger galls (chi-square = 195.5, $P \ll 0.001$; Fig. 5). The occurrence of parasites on veins is significantly non-random (chi-square = 12.9, d.f. = 5, $P < 0.05$), entirely due to the unexpectedly few parasitized galls found on the 'top' vein (Fig. 1).

Synergus nervosus occurred in 40% of the galls opened, and were generally found in the larger galls (in 67% of those > 4 mm). This is puzzling since Askew (1961) states that attack by *S. nervosus* occurs early and leads to stunting of the gall. More *S. nervosus* were found in the first gall on a vein than in the second or third (chi-square = 6.8, d.f. = 2, $P < 0.05$). *Synergus pallicornis* forms a small chamber in the wall of the gall. It occurred in 19% of the galls opened, and only those in which *S. nervosus* was present. *Eurytoma brunniventris* was found in 15% of galls; it also was generally found in larger galls, again in contrast with Askew's findings.

In contrast, fungal attack was concentrated in the smaller galls, and was common (20%). It is not possible to tell whether the fungus was the cause or consequence of the death of the gall occupant, but it was strongly associated with the absence of evidence for parasitism (chi-square = 54, d.f. = 1, $P \ll 0.001$). This may be because mould obscures the evidence for parasitism, or that the mortality occurs earlier than parasite attack.

4. What affects the survival and fecundity of the gall-maker?

Since in 1986 only 2 of 443 galls contained living *C. divisa* (0.45%), this section uses less comprehensive data collected from another site during 1985, when survival varied from 0 to 67% on different leaves.

The percentage survival of *C. divisa* was uncorrelated with either leaf size ($r = -0.14$, $n = 22$, n.s.) or with gall density between leaves ($r = -0.1$, n.s.), but although the scatter is large, we believe that there is a negative trend that would probably be significant with a much larger sample size. Figure 6 shows that survival is highly correlated with gall density within leaves.

Fecundity of individual females varied from 115 to 241 eggs. The highest values occurred at intermediate leaf sizes (10-11 cms long) and at intermediate gall densities.

Nearly all the data on fecundity were gathered from females who were the sole survivors on their vein: only in two cases were two survivors on a vein found. Clearly these data are very scanty. However, there is an intriguing

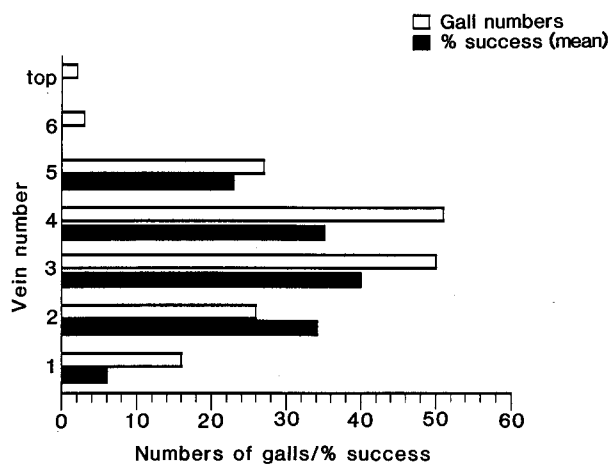


Fig. 6. Success of the gall-maker (% survival) and the number of galls on different veins of the leaf. Note that the x-axis represents both percentages (mean survival) and number of galls

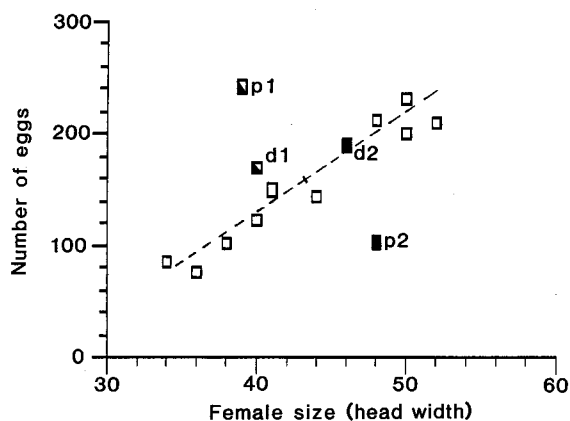


Fig. 7. Plot of size (head width in microscope units) and potential maximum fecundity (number of eggs at emergence) of the gall-maker. Filled symbols represent proximal-distal pairs on veins with two survivors; all other data including the regression line concern veins with only a single survivor. *p* proximal; *d* distal

contrast in single- and double-surviving females, seen in Fig. 7. Where a female has a companion on a vein, the size-fecundity relationship is altered substantially. The alteration occurs in opposite directions in the two cases at hand: the proximal female had either abnormally large (P1) or small (P2) numbers of eggs for her size (t-test for the admittedly a posteriori test shows that both are very significantly different from the predicted values, $P < 0.01$), whereas the distal females are not different from the data forming the line.

Discussion

The interaction between galls and their host plants is currently under intensive research (Weis and Abrahamson 1986; McCrea et al. 1985), and the great abundance and diversity of parasitoids in gall systems allows consideration of the third trophic level in these miniature ecosystems (Price et al. 1980; Weis and Abrahamson 1985). This study presents patterns seen in the galls of the agamic generation of *Cynips divisa* which are susceptible to experimental study, and sets the scene for investigations into the evolutionary biology of this 3-level system.

While in our study in 1985 some 20% of galls contained live adult female *Cynips*, dissection of more than 440 galls in 1986 disclosed only 2 surviving gall makers. This may indicate that in *C. divisa* at this latitude populations are evanescent, colonising and dying out as their parasitoids catch up with them. This shifting pattern of populations may be common in insects (Taylor 1986) and may characterise biological control by parasitoids (Murdoch et al. 1985). Washburn & Cornell (1981) described the local extinction of a gall-wasp population caused by its parasitoids, and a similar situation may exist in *C. divisa*. Only long-term data can clarify these ideas. However, it is dangerous to consider *C. divisa* in isolation from other galls; its parasitoids are polyphagous, and therefore will survive in an area in the absence of pea galls. This argues against the interpretation of shifting pattern of populations as Taylor (1986) suggests.

Ovipositing females of the sexual generation obviously tend to lay on the middle veins of leaves. This could be either due to the timing of oviposition, or because of site selection by females. Without observational data it is difficult to be sure, but Askew (1961) notes that galls first appear during mid-June, when leaves are already fully developed. Thus it seems likely that females select veins for oviposition. We have as yet no data on the consequences of this choice to wasp fitness.

We do not yet know whether a large gall produces a large (and therefore fecund) wasp, since gall sizes were not recorded in 1985 when survival was good. However, the pattern of gall development on multi-gall veins is consistent with a resource-interception hypothesis; plant resources demanded chemically by distal galls for growth are partially intercepted by proximal galls, leading to larger sizes. Larger galls may make parasitoid attack less likely (Askew 1961). We have some evidence (Fig. 7) that such extra resources can be channelled into altering size and/or fecundity, probably via resource allocation patterns of the wasp. We consider that the allometric relationship between size and fecundity has great potential as a sensitive probe for detecting growth conditions (cf. Collins 1980). Tests of these ideas will require the exclusion of parasites and manipulation of gall densities on veins, both easily accomplished in this system.

The role of the fungus-tannin interaction has been explored for an oak-gall system by Taper et al. (1986), and their hypothesis may well apply here too. *C. divisa* females may choose their oviposition site by detecting tannin levels (cf. Zucker 1982), as an evolutionary response to high mortality from fungal attack. In this study, fungus was probably the second most important cause of death to gall occupants (20%).

Acknowledgements. We thank Dr. J.C. Hartley, David Conway and the Insect Ecology class of 1986 for help with this work; we also thank Dr. R.R. Askew, who made many valuable comments on the manuscript.

References

- Askew RR (1961) On the biology of the inhabitants of oak galls of Cynipidae (Hymenoptera) in Britain. *Trans Soc Br Ent* 14:237-268
- Askew RR (1980) The diversity of insect communities in leaf mines and plant galls. *J Anim Ecol* 49:817-829

- Askew RR (1985) The biology of gall wasps. In: Ananthkrishnan TN (ed) 'The biology of gall insects'. Oxford, pp 223-271
- Bernays EA (1981) Plant tannins and insect herbivores: an appraisal. *Ecol Ent* 6:353-360
- Collins NC (1980) Developmental responses to food limitation as indicators of environmental conditions for *Ephydra cinerea* Jones (Diptera). *Ecology* 61:650-661
- Crawley MJ (1985) Reduction of oak fecundity by low-density herbivore populations. *Nature* 314:163
- Feeny PP (1976) Plant apparency and chemical defence. *Rec Adv Phytochem* 10:1-40
- McCrea KD, Abrahamson WG (1985) Evolutionary impacts of the goldenrod gallmaker on *Solidago altissima* clones. *Oecologia* 68:20-22
- McCrea KD, Abrahamson WG, Weis AE (1985) Goldenrod ball gall effects on *Solidago altissima*: C14 translocation and growth. *Ecology* 66:1902-1907
- Murdoch WW, Chesson J, Chesson PL (1985) Biological control in theory and practice. *Am Nat* 125:344-366
- Price PW, Bouton CE, Gross P, McPherson BA, Thompson JN, Weis AE (1980) Interaction among three trophic levels: influence of plants on interaction between insect herbivores and natural enemies. *Ann Rev Ecol Syst* 11:41-65
- Taper ML, Case TJ (1987) Interaction between oak tannins and parasite community structure: unexpected benefits of tannins to cynipid gall wasps. *Oecologia* 71:254-261
- Taper ML, Zimmerman EM, Case TJ (1986) Sources of mortality for a cynipid gall wasp (*Dryocosmus dubiosus* (Hymenoptera, Cynipidae)): the importance of the tannin-fungus interaction. *Oecologia* 68:437-445
- Taylor LR (1986) Synoptic dynamics, migration and the Rothampsted insect survey. *J Anim Ecol* 55:1-38
- Washburn JO, Cornell HV (1981) Parasitoids, patches and phenology: their probable role in the local extinction of a cynipid gall wasp population. *Ecology* 62:1597-1607
- Weis AE, Abrahamson WG (1985) Potential selective pressures by parasitoids on a plant-herbivore interaction. *Ecology* 66:1261
- Weis AE, Abrahamson WG (1986) Evolution of host-plant manipulation by gall makers: ecological and genetic factors in the *Solidago-Eurosta* system. *Am Nat* 127:681-695
- Whitham TG (1980) The theory of habitat selection examined and extended using *Pemphigus* aphids. *Am Nat* 115:449-466
- Whitham TG (1986) Costs and benefits of territoriality: behavioural and reproductive release by competing aphids. *Ecology* 67:139-147
- Zucker WA (1982) How aphids choose leaves: the roles of phenolics in host selection by a galling aphid. *Ecology* 63:972-981
- Zucker WA (1983) Tannins: does structure determine function? An ecological perspective. *Am Nat* 121:335-365

Received November 16, 1986