resonances and continuum states, not intrinsically different from their less massive counterparts. Indeed, such a model implies some degree of correlation among clusters of arbitrary size, which makes the notion of an individual cluster slightly nebulous. But this sort of microscopic interpretation, even if correct, might not provide great economy or insight. It may be that clusters can be described most economically as collective excitations of hadronic systems with many degrees of freedom. In this event, theoretical techniques traditionally brought to bear on nonrelativistic many-body problems will supply new insights into their nature. Alternatively, it may be possible to make explicit the suggestive connection with the quark parton model description of the hadron jets observed in inelastic electron-proton collisions. This would yield a unified description of the full spectrum of clusters.

Either development would raise the clustering phenomenon to a general property of hadronic matter. It should then be observed not only in hadronic reactions but also in lepton-induced reactions in which hadronic final states are formed. First indications of clustering effects are already visible in proton-antiproton annihilation and in electron-positron annihilation into hadrons at high energies. New opportunities for experimentation with colliding electron-positron beams with CM energies up to 40 Gev, which will appear soon, undoubtedly will shed new light on the apparent similarity between many-particle reactions initiated by different beams.

In conclusion, the cluster concept has led to many important insights, and further study of the phenomenon of clustering is certain to provide clues for a deeper understanding of the strong interactions.

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1. In collisions of relativistic particles with a stationary proton target, the center-of-mass energy is $W = [2M(E_{\mathrm{lab}} + M)]^{1/2}$, where E_{lab} is the beam energy and M is the proton mass. At very high energies, W is approximately the square root of twice the beam momentum, in Gev.
2. Were particles emitted isotropically in the CM system, we should regard particle production as a three-dimensional process. There are regimes, in low-energy annihilation reactions and in the decays of massive particles, in which the three-dimensional picture seems appropriate.
3. The hydrodynamic approach was initiated by L. D. Landau [*Izv. Akad. Nauk SSSR Ser. Fiz.* **17**, 51 (1953)]. A recent survey is given by P. Carruthers [*Ann. N.Y. Acad. Sci.* **229**, 91 (1974)].
4. The multiperipheral model was introduced by D. Amati, A. Stanghellini, and S. Fubini [*Nuovo Cimento* **26**, 896 (1962)].
5. R. P. Feynman, *Photon-Hadron Interactions* (Benjamin, Reading, Mass., 1972).
6. If all effects of energy and momentum conservation may be neglected (a good approximation in the pionization region), the correlation function can depend only on the rapidity difference $|y_1 - y_2|$.
7. S. R. Amendolia *et al.*, *Nuovo Cimento A* **31**, 17 (1976).
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Coevolution in Insect Herbivores and Conifers

Scale insects adapt to intraspecific variation in pine defenses with a differentiated deme on each tree.

George F. Edmunds, Jr., and Donald N. Alstad

Sessile habits facilitate demographic studies in many conspicuous scale insects and permit a number of experimental manipulations that illustrate plant-herbivore interactions. Outbreaks of black pineleaf scale, *Nuculaspis californica* (Coleman), on ponderosa pine, *Pinus ponderosa* (Laws), are caused when dust or insecticide drift kills the *Prospaltella* (Eulophidae: Aphelininae) parasitoid, which normally controls scale abundance (*1*). Evidence from long-term observation of persistent outbreaks led to the hypothesis that ponderosa pine shows a complex intraspecific variation in its defensive characteristics, and that scale insects are selected for increasing adaptation to individual host trees (*1*).

A number of observations provide evidence that pines are resistant to the scales. One striking characteristic of an infestation is the variation of scale population density from one pine to the next; scale-free pines frequently stand for years beside trees infested with as many as ten insects per centimeter of needle, often with intertwining branches. Scale-free trees tend to remain uninfested even though first instar larvae can be seen crawling on their needles during the insect's reproductive period in July. When plots of trees are sprayed with insecticide to control the scales, trees are reinfested after 2 to 5 years to approximately their original level of infestation; that is, formerly severely infested trees become severely reinfested, previously lightly infested trees become lightly reinfested, and so forth. When twigs are reciprocally grafted from adjacent infested and uninfested trees, the grafted twigs retain their characteristics. Two of three scale-free grafts remained uninfested for 3 years (after which the trees were removed for construction of a building), and none of the infested twigs caused adjoining twigs to become infested.

Other evidence suggests that tree defenses are complex and that scales are adapted to host individuals. In severe outbreaks, where trees receive numerous potential colonists for many years, most of the scale-free trees eventually become infested. After a few scales successfully establish themselves, insect population density gradually increases in succeeding generations (unless a high winter mortality occurs). When only the lower limbs of infested trees were dipped in insecticide and 100 percent mortality was achieved, the scale populations returned to normal in two generations (2 years). In contrast, similar dipping of every twig on 4- to 5-foot (1 foot = 0.3 meter) pines that were surrounded and overgrown by densely infested trees resulted in a scale-free period of up to 10 years. The recolonization in both cases appears to be primarily by offspring falling from overhanging limbs; and, although fewer larvae reached the smaller trees, this does not account for the difference in repopulation rate. Finally, in scale outbreaks there is a strong correlation of scale population density with the

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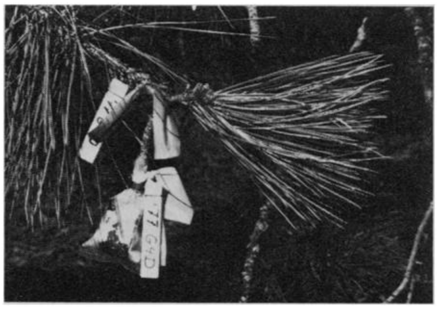

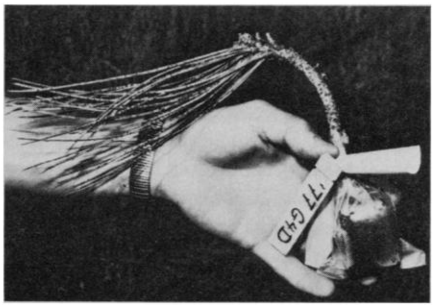

Fig. 1 (left). Infested sample tied in receptor tree. One-year-old needles of the donor sample lie in contact with the current needle crop of the receptor. Some second- and third-year receptor needles have been stripped away to clarify the photograph. Fig. 2 (right). Donor twig illustrating details of transfer sample preparation.

Table 1. Analysis of variance results. Accidental loss of some transfers resulted in unequal sample sizes in the treatment cells. Because partitioning order is important in nonorthogonal anova designs of this type, two tables with reversed main treatment order are presented. Abbreviations: SS, sums of squares; DF, degrees of freedom; MS, mean squares; F, F-ratio; and P, probability.

Source	SS	DF	MS	F	P
	Test of receptors preceding donors				
Within cells	173,091	172	1.006		
Receptors	48,976	9	5.442	5.407	.001
Donors	19,980	9	2.220	2.206	.024
Interaction	118,095	81	1.458	1.449	.023
	Test of donors preceding receptors				
Within cells	173,091	172	1.006		
Donors	20,888	9	2.321	2.306	.018
Receptors	48,068	9	5.341	5.307	.001
Interaction	118,095	81	1.449	1.449	.023

age of the host tree. Of 667 pines randomly sampled on one hillside, 10 percent of the trees under 3 meters high were heavily infested (more than 1.4 scales per linear centimeter of needle), 19 percent of the pines between 3 and 7.5 meters, and 67 percent of those over 7.5 meters were heavily infested.

From this background, we have extended and begun experimentally testing the initial hypothesis. Specifically, we propose that (i) individual trees vary in the defensive phenotypes which they present to scale insects; (ii) selection over many generations produces scale insect populations which are increasingly adapted to the defensive character of their host tree (that is, the insects track individual hosts); and (iii) selection of scale insects for maximum fitness on one tree is maladaptive with respect to the prospect of establishing colonies of offspring on trees of differing defensive phenotype. The result should be a series of semi-isolated subpopulations, or demes, on individual pine trees.

Survival of Intertree Transfers

We have performed a series of field experiments designed to test these hypotheses. Each experiment was based on a series of artificial scale insect transfers from an infested tree (the donor tree) to uninfested trees in another area (the receptor trees). Infested twigs were cut from donors just prior to scale reproduction and hung in the limbs of receptors (Figs. 1 to 4). First instar larvae established themselves on the current needle crop of the limb into which the transfer twig was tied (*2*). A week later, after hatching and establishment of the young scales were complete, each transfer was reexamined, and the number of successful colonists was quantified by estimating population sizes and assigning them to one of six ranks. Identical censuses made at subsequent intervals allowed us to calculate a survival index (the ratio of final rank abundance to the initial rank abundance) for each transfer. For example, a transfer whose rank abundance fell from an initial rank 4 to rank 2 after 9 months (overwintered second instar) had a survival index of 0.5. Control data show that we were unable to move a constant number of offspring with each transfer, and the survival indices reduce the impact of this variance.

One transfer experiment involved ten donor trees and ten receptors. Each possible combination was repeated in triplicate, resulting in a total of 300 transfers. Initial and 9-month (overwinter) abundance ranks were used to calculate a 10 by 10 by 3 array of survival indices suitable for the two-way analysis of variance (hereafter "anova") (*3*). Anova results are given in Table 1. This statistical procedure allows us to test three important aspects of the data set. The first two, the main treatments, examine differences between rows and between columns, corresponding in this case to the donor and receptor, respectively. Both results are highly significant, showing, for the rows, that the variance between scale demes originating from different donors is much greater than variance within scales from a single host. Some scale demes are significantly better colonists than others, and this presumably is a result of their adaptation to different donor trees. Likewise, the receptor trees (columns) show statistically significant variation in "resistance." Scales do not survive as well on some trees as they do on others. The third test in the anova, for interaction variance, is also significant; this is designed to detect exceptions to the main treatment patterns. Interaction would be suspected, for example, if insects from a relatively poor colonizing deme did especially well on a very resistant receptor tree. Our hypothesis predicts that the success of scale transfers should correlate with the defensive similarity of the donor and receptor trees, and the result, in the final data, should be significant interaction variance. It implies that resistance can only be quantified in the context of the insect genotypes against which a defense is mounted.

We continued to observe these transfers until they reached reproductive age the following July, and it appears that black pineleaf scale may be even more sensitive to the identity of its host than our analysis of the 9-month survival indicates. The best measure of compatibility is the ability of the transferred scales to reproduce. Our data are unfortunately very incomplete, but of 81 transfers examined with surviving second, third, and fourth instars, 28 (36 percent) completely failed to produce young, and in only two cases did the number of offspring exceed the total number of mature potential parents.

While our anova results are consistent with the interpretation that selection causes differentiation of scale insect demes on individual pine trees, a competing counterhypothesis remains. The strong positive correlation of tree age and scale population density could result simply from age-related weakening of tree defenses. To investigate this possibility, we conducted a transfer experiment on ten donor trees and ten receptors, with two replicates of each transfer. Receptors spanning a broad range of sizes were chosen, and age was determined to range from 13 to 78 years. After 9-month survival indices were calculated, means of the indices from each receptor were correlated with receptor tree age. No statistically significant correlation resulted. It is concluded that the strong relation between tree age and scale population density observed in outbreaks is not caused by a decline of resistance as the trees grow older.

Intratree Transfers

At the time of each intertree transfer experiment, we also made replicated intratree control transfers to scale-free isolated branches that had been dipped in insecticide. In the anova fieldwork and a concurrent experiment, one batch of transfers, which included the controls, was accidently mishandled and very few offspring resulted; but replicated controls were established successfully for the ten donors of the experiment on tree age defense. Of the 20 donor tree transfers, six were subsequently destroyed by a construction project and other causes; data for the remaining 14 are contrasted with age defense data in Fig. 5. Although the variance resulting from unequal original establishment is high in the numerical counts, within-tree transfers show 16 times as many larvae surviving to the second instar as do between-tree transfers. The highly skewed distribution of survivorship on intertree transfers shown in Fig. 5 is consistent with our hypothesis that survival success of transferred scales is related to defensive similarity of donor and receptor trees.

Our results provide evidence for the hypothesis that selection produces differentiated demes of black pineleaf scale on individual ponderosa pines. We have demonstrated the following. (i) Individual pines show variation that results in significantly differing survival success of scale insects artificially transferred to their branches. (ii) Scale insects originating from separate donor trees show significant differences in the ability to survive on a variety of receptor trees. (iii) Interaction variance in the transfer data is consistent with the hypothesis that survival success is correlated with the similarity of donor and receptor trees. (iv) There is no evidence of age-related decline in tree defenses effecting insect survival. (v) Finally, scale insect survival after intertree transfers is much lower than that of intratree control transfers.

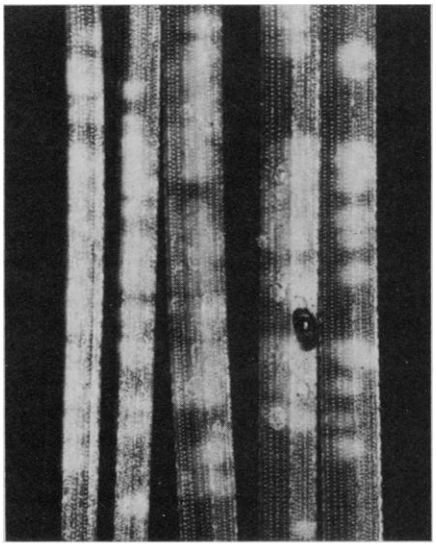

Fig. 3 (left). Needle fascicle from a receptor, showing transferred black pineleaf scale which have survived into the third instar. Fig. 4 (right). Receptor needles. One surviving third instar scale is visible. In addition, scale covers of numerous insects which died in the first instar can be seen. At each site where a first instar scale larva settled and began feeding, the needle is banded with a chlorotic lesion.

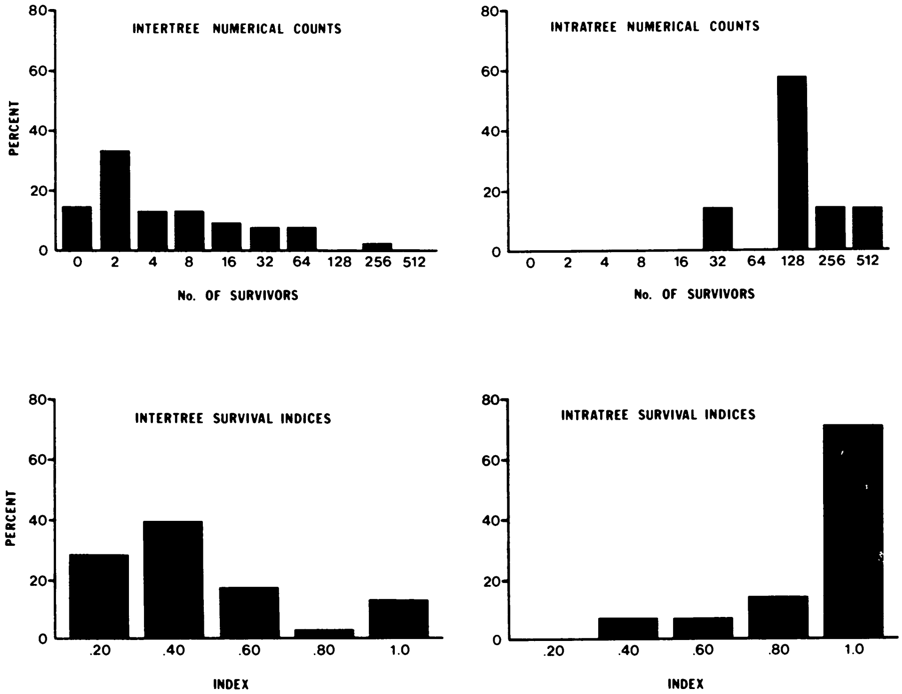

Fig. 5. Control and age dependence data. The histograms contrast insect survival from the age dependence experiment (intertree) with that observed in control transfers back to the original donor (intratree). The two upper graphs give the percentage of 9-month survivor counts in ten numerical categories; lower graphs show similar distributions of 9-month survival indices which eliminate the effects of unequal establishment. Scales on the abscissa indicate the upper limit of values included in each histogram bar.

These results emphasize two questions that require further clarification.

1) Our use of the word "deme" in reference to the differentiated insect subpopulations on individual hosts implies that the differentiation is genetic. From the data at hand, this is a sound inference, but transfer experiments do not prove that we are observing genetic phenomena in the scales. Because all of the experiments involve two generations of scales, mechanisms other than genetic differentiation seem unlikely.

2) The specific mechanisms responsible for the differential survival of transferred scales on ponderosa pine are unknown. The differences might be explained by variation in nutritional quality

of the pines or in densive compounds. The pines contain a variety of terpenes, nonvolatile terpene acids, polyphenols, and tanninlike substances. The diversity and intertree variation of monoterpenes has led us to speculate that they are the most likely cause of the observed scale population variation. They are known to be defensive toxins (*4, 5*) and they are under strong genetic control (*5*). Gardner and Sanders have found that monoterpenes are the most abundant volatile compounds in the leaves of ponderosa pine in the plots under study, and that they vary among trees both in quantity and quality (*6*). At least ten molecular species are present, and their fractional contribution to the total varies from tree to tree. Smith (*4*) and Hanover (*5*) note that terpenes are extremely variable, and the latter states that terpene variation gives "almost every tree a unique individuality."

Discussion

Plant defensive compounds have been divided into two functional classes by Feeny (*7*) and Rhoades and Cates (*8*). One group, exemplified by glucosides in the Cruciferae (*9*), may be toxic to insects at low concentration, but is subject to various countermeasures by coevolved specialist herbivores. Because selection can produce insect populations tolerant of most toxins, the utility of each compound will depend on its frequency in the plant community, with rare toxins conferring the greatest protection. These pressures have resulted in a great diversity of toxic compounds. They are best known in short-lived or widely scattered plants whose lack of predictability in time and space offers them an additional escape from specialist pests. The second class of compounds functions by forming complexes with plant proteins and reducing their digestibility for the herbivore. Because the target of these tanninlike defenses is the plant protein itself, even specialist pests have few opportunities to intercede and must pay the price of reduced growth rate on a low-quality resource or feed only on young tissues whose tannin concentrations are minimal. The tannin defense is more characteristic of plants that are "apparent" or "bound to be found" (*7*) by specialist herbivores because of their longevity, spatial distribution, temporal predictability, and the like (*7, 8*).

Because ponderosa pines are long-lived, closely spaced, and large, they are presumably "apparent" to specialist herbivores; one would expect them to maintain a tanninlike defense, and protein-complexing compounds are well known in conifers (*8, 10*). Intraspecific variation demonstrated in our experiments suggests that the frequency dependence characteristic of coevolving toxin-herbivore interactions is involved, and that ponderosa pines are using both tannin- and toxinlike defenses.

The use of antiherbivore toxins in a long-lived perennial has a number of interesting coevolutionary consequences. Individual trees will benefit by diverging from the defensive pattern of their neighbors, and the array of toxins should be diverse. If short-lived plants had a large number of toxins, specialist herbivores would be forced to detoxify this entire array to maintain mobility between host individuals. But in pines, where host longevity can span 200 insect generations, pests theoretically can drop superfluous detoxification mechanisms, and differentiated populations will develop on individual trees. Such differentiation would reduce the fitness of the offspring on hosts of differing defensive phenotype. As a result, the toxin defenses of apparent plants should be variable and diverse (*11*).

Insects that track individual tree defenses should show adaptations which confine a large proportion of their progeny to a host individual and reduce the flow of genes between insects on different trees. Female scale insects are completely sessile, and most of the newly hatched larvae crawl only a few centimeters to new needles on the same twig. Gene flow between demes occurs only when these larvae are blown and settle on another tree, or by interdemic mating of the winged males. Although females probably issue a sex pheromone that can attract mates from some distance, the extent of interdemic mating should vary with insect density. If a tree is heavily infested, a large fraction of the males within potential mating distance will be members of the same deme. In contrast, insects surviving at low densities on their host should be much more prone to outcrossing and gene exchange. Because increased gene flow will be a major factor restraining the differentiation of insect demes on individual trees, outbreak situations in which scales largely escape density reduction by parasitoids may promote unusually close host tracking. All of the insects used in our transfers were taken from persistent high-density outbreaks, but long-term survival of insect pests at high densities is relatively rare. If more interdemic mating occurs in natural low-density situations, the resulting gene flow should be more disruptive of the tracking of individual hosts.

As a result of these interactions, we perceive a number of advantages for the pine tree. Intraspecific defensive variation forces the coevolved herbivore into an evolutionary dichotomy. A clear trade-off exists between colonizing ability, and adaptation to a single host; results of our transfer experiments indicate that, for the pine-scale system, interdemic and kin selection pressures for colonizing ability are weaker than the individual pressures favoring tight host tracking. In strongly outcrossing pines, this will impede the spread of insects from a tree to its offspring. However, as differentiation of insect demes becomes more extreme, demographic characteristics of the population will increasingly favor the group pressures (*12*). Host tracking could ultimately result in insects so specialized that their extinction would be inevitable with the death of a tree. Winged males and the resulting potential for interdemic mating indicate that the opposing tensions are balanced short of this extreme, and the continuing scale mortality that we observe even on heavily infested trees is probably evidence of the introduction of maladaptive genotypes.

Conclusions

Our transfer experiments have demonstrated the differentiation of scale insect demes on individual host pines. The interaction has a number of defensive benefits for the host. Recombination in outcrossing pines will produce a variety of defensive patterns and minimize the probability of passing highly preadapted pests from parent to offspring; similarly, insects will be forced to compromise the tracking of individual hosts with the necessity of colonizing new trees.

These arguments are general, and we anticipate that the intraspecific variation of defenses will be more widely recognized in long-lived plants as field biologists become sensitive to the pattern of insect densities and the correlation of infestation with plant age which first attracted our attention. The occurrence and variation of monoterpenes in xylem, cortex, and leaves of pines suggests that these substances are general antiherbivore or antipest compounds. We expect that differentiation on host individuals will be demonstrated in a variety of insects, and perhaps in some pathogens and parasites (such as dwarf mistletoe on pine) which coevolve with toxin-defended, long-lived plants. We have seen this pattern in gall aphids (*Adelges* spp.) on white, blue, and Norway spruces, and Riek (*13*) reports similar observations with eurymelid leafhoppers (Homoptera) on *Eucalyptus*. Reproductive behaviors which confine the offspring to a single tree also suggest the possibility of host tracking. The female spruce budworm [*Choristoneura fumiferana* (Clemens)] travels to other trees, but "does not fly until she has deposited one or two egg masses" (*14*). Douglas-fir tussock moths (*Orgyia pseudotsugata* McD.) have wingless females that deposit their eggs on the cocoon and winged males. The first instar larvae are transported by the wind. We had predicted that the Douglas-fir tussock moth would also show differentiation on host individuals, and Cates (*15*) has indicated that they are selective of host individuals and may be differentiated.

In trees that produce tanninlike defenses, resistance to insect attack is determined by the concentration of compounds that reduce digestibility in the tissue. The coevolved nature of toxin-herbivore interactions makes them quite different, and, in this case, resistance per se has little meaning. We stress that in coevolved systems resistance can only be quantified in the context of the genotypes of the insect population against which a defense is mounted.

In view of these interactions, breeding and cloning programs for rapid-growth conifers may need reevaluation. Both cloning and selection can greatly narrow the variation in the pine gene pool. This reduction of defensive variation will destabilize the coevolutionary equilibrium and increase vulnerability of the trees to insect damage. In addition, raising the trees in monocultural plantations will further increase their "apparency" (*7*). With conscious spatial management of tree strains differing in defensive phenotype, however, these same interactions could be used to silvicultural advantage. The maintenance or enhancement of defensive heterogeneity in plants should be recognized as a major objective in agricultural genetics.

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