

Crossroads of Animal, Plant, and Microbial Physiological Ecology

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Animal, plant, and microbial ecophysiologicalists have diverged greatly in the last few decades in the principal research questions they address (e.g., global change versus evolution), in the methods they use, and even in the professional societies to which they belong. Two symposia in 2001 brought these diverse groups together, with presentations by researchers who study two or three kingdoms in intimate interaction. The second symposium, the subject of this report, was sponsored by the Ecological Society of America's Physiological Ecology section. Several of the presentations showed, among other things, commonalities in chemical signaling among kingdoms, as well as exploitation of such signals and other metabolic pathways by parasites and their hosts. These and other important findings from such interkingdom and interdisciplinary research can help explain why current functional groups exist.

Keywords: physiological ecology, ecophysiology, cross-kingdom, chemical signaling, functional groups

Animal and plant ecophysiologicalists used to address highly analogous questions such as, How do organisms balance fluxes of water, solutes, heat, and radiation? and How do adaptations and acclimations in such processes confer fitness? Indeed, these two groups of scientists—and a few microbial ecophysiologicalists—used to be found in the same professional societies. Today, the disciplines have diverged. A plant ecophysiologicalist is likely to measure whole-stand fluxes and aim to scale up to the globe; an animal ecophysiologicalist is more likely to take a highly molecular approach, as in the study of heat-shock proteins and their evolutionary history. Yet the two disciplines have much to offer each other, even in techniques alone. In global change, for example, the responses of bird populations to extremes of heat concurrently with human use of pesticides call for studies that join organismal physiology of animals with large-scale flux estimation that is more in the purview of plant ecophysiologicalists.

The dichotomy between the two fields is in active discussion among members of the Ecological Society of America's (ESA) Physiological Ecology section, which is largely made up of plant ecophysiologicalists, and the Society for Integrative and Comparative Biology (SICB). An evening discussion session at the ESA meeting at Snowbird, Utah, in August 2000 led to two symposia. SICB presented "Plant/Animal Physiology" in

January 2001, organized by Martin Feder, immediate past president of the society (Feder 2002). This meeting was followed in the summer by the symposium summarized in this article. Organized by the current authors and supported by the National Science Foundation, this symposium brought together six researchers whose work deals in detail with two or more kingdoms. They discussed many examples of the more well-known interactions of plants with insect and vertebrate herbivores, but their work extends to intriguing contrasts involving all five kingdoms.

Malcolm Press (University of Sheffield, United Kingdom) demonstrated many parallels between parasitism of higher plants by fungi and parasitism by other higher plants, such as *Striga*, the curse of African subsistence farmers, and mistletoe. While the parasitic vascular plants are not widely known (with the exception of mistletoes), they occur in seven orders in 16 families (Press et al. 1999). Twelve independent

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evolutionary events have been traced from free-living ancestors to obligate parasites and hemiparasites. Thus, both fungi and vascular plants provide abundant examples of contrasting free-living and parasitic nutritional modes. Vascular parasites may be larger than their hosts, as are sandalwood trees that parasitize many woody and herbaceous neighbors. Parasites may have haustoria to the shoot or to the root of the host. Remarkably, parasitism can increase water and nutrient flux to the host–parasite complex. The increase is as much as fivefold in some heath–parasite associations (Press and Whittaker 1993), though the parasite still depresses the host status. The fitness effects can be dramatic. *Cuscuta salina* parasitism on *Salicornia* in salt marshes leads to open patches in the landscape that allow invasion by other plant species such as *Arthrocnemum*.

The focus is the chemical signaling between host and parasite, with its short range and high accuracy. A signal for *Striga* is the sesquiterpene strigol. It is related to many secondary metabolites that, ironically, are often defensive against fungi, though the hypothesis of their value has not been adequately tested. Strigol is mobile, moving some millimeters from the root of the potential host. The structure–function relations of the germination cues are difficult to discern because of the difficulty of isolating them and because of their complex stereochemistry. Furthermore, the recognition sites on the parasites remain unknown; much opportunity exists for chemists, molecular biologists, soil scientists, and others to clarify these important systems. Using chemical cues, a *Striga* seed germinates and forms the parasitic haustoria, which are similar in function to fungal hyphae. Other cues vary widely in chemical makeup. They include flavonoids, p-hydroxyacids, quinones, and even cytokinins. Modes of defense are unclear. Press and his colleagues are nevertheless seeking to breed *Striga*-resistant maize, a highly practical application of physiological ecology.

Sue Hartley (University of Sussex, United Kingdom) critically analyzed patterns of occurrence of plant secondary compounds providing defenses against insect herbivory. The diversity of such compounds is astonishingly great. To explain a significant part of the variation, she asks secondary compounds segregate according to plant functional groups. For example, alkaloids are found in 33 percent of herbs but only 17 percent of trees. Can plant functional groups also be classified according to insect feeding modes, or guilds? Leaf-mining and sap-sucking certainly require disparate types of defense. Phylogeny does account for some grouping of secondary defenses, though diverse life forms commonly co-occur in single families—many violets in the Southern Hemisphere are woody, for example.

Mechanistic and evolutionary requirements certainly underlie groupings of defensive compounds. These requirements include

- Biosynthetic necessity: The plant must have the beginning of the defensive pathway to evolve any defense

- Physiological correlation: Lignin required bio-mechanically can also be defensive
- Phylogeny: Plants are constrained by their evolutionary history (e.g., the diterpene lactones in *Ginkgo* or the nonprotein amino acids found in only one sub-family of the Fabaceae)

Consequently, one can even discern the phylogeny of some species by their defensive compound arrays. These three requirements can operate jointly; thus, woody plants have developed some of their defenses as offshoots of lignin biosynthetic pathways. In contrast, independent evolutionary events may occur in response to selection pressure. Silvertown and Dodd (1996) used phylogenetically independent contrasts to show that the dominance of alkaloids over tannins in defenses of herbaceous plants was not purely phylogenetic.

Insect guilds certainly group with plant functional groups, evidencing coevolution, first described by Ehrlich and Raven (1964) as a chemical arms race. Ward and colleagues (1995) looked at broader patterns in 107 plant families, showing clear groupings of woody plants from herbs and evergreens to annuals. Lawton (1983) found three-quarters of gall-formers (which need meristems and lignification potential) on woody plants, whereas aphids are most commonly found on herbs.

Overall, the functional-group approach to patterns of defense has achieved a restricted success. Further progress requires collaborations among chemists, molecular biologists, plant ecologists, and entomologists.

Heidi Appel (Pennsylvania State University) traced the elaborate chemistry of polyphenol oxidases, or PPOs. Polyphenols and their oxidases are involved across kingdoms—in flower coloration, cuticle construction, the immune responses of arthropods, human nutrition, bivalves' byssal threads, and more. In plants, their action with reactive oxygen species provides, by parallel pathways, both a greater degree of lignification and direct damage to insect herbivores. Plant PPOs can inactivate insect digestive enzymes by generating oxidizing quinones. The sequence from protonated to ionized to oxidized forms is a sequence of sequentially stronger bonds to proteins via van der Waals forces and hydrogen bonds to ionic bonds to covalent bonds.

The PPOs occur in two major pathways, producing simple polyphenols and tannins (high-molecular-weight polyphenols). Simple polyphenols participate in the creation of reactive oxygen species (some of which are defensive) and in biosyntheses, such as lignification, for structure and defense. Defense modes tied to PPOs are diverse, including inactivation of pathogen enzymes, programmed destruction of infected cells to reduce pathogen spread, distastefulness to insects, gum production to entangle insects, and reduction of digestive efficiency. The plant's defensive enzymes may retain activity inside the insect gut. Plant PPO defenses are in part constitutive but commonly inducible, with signaling occurring via the wound hormone, jasmonic acid, for one. Plants respond to an attack with upstream signaling, which was discussed in detail by Richard Bostock in a later talk. Insect

herbivores may manipulate plant PPOs to enhance their own fitness, as when gall-formation by *Andricus* wasps downregulates (reduces the expression of) plant PPOs.

All bacteria studied to date possess catecholase PPOs, but the only well-studied case is that of *Mycobacterium leprae*, which uses them to invade catecholamine-rich areas of human tissue to cause leprosy. Bacterial pathogens also manipulate host PPOs; they inhibit the latter with simple phenols such as ferulic and caffeic acids. Fungi mobilize nutrients with PPOs, while also using them defensively. Against hyphal grazers, fungi use laccase PPOs, which are copper-containing PPOs of low specificity that can oxidize both ortho- and para-hydroxyquinones. In complementary offensive fashion, the parasitic fungi often degrade host defenses with PPOs. Animals use PPOs defensively in some novel ways, such as for sclerotization of the cuticle or for melanization (defense against abiotic stress such as ultraviolet light). Their offensive uses are abundant; one example is a mosquito's use of salivary catechol oxidases to prevent vasoconstriction in a vertebrate host and obtain a blood meal.

It is apparent, then, that plants, animals, and microbes use similar PPOs defensively and offensively. This cross-kingdom use of PPOs means that the fitness consequences of particular levels of PPOs and their substrates may not be predictable unless all the players are taken into consideration. For example, when gypsy moth caterpillars damage leaves, they induce higher levels of tannins, which cause them to grow more slowly and lay fewer eggs; however, this induction also lowers mortality from a naturally occurring baculovirus, which is inhibited by PPO-mediated tannin oxidation. When organisms manipulate PPOs of their counterparts, hosts, parasites, or pathogens, the value of these manipulations can be assessed by inhibiting or overexpressing PPOs with modern molecular techniques. The important work of discerning the patterns is just beginning. Multidisciplinary research will be the key to understanding these relationships.

Sharon Strauss (University of California–Davis) examined tolerance of plants to insect damage. Because individual plants typically are attacked by herbivores, they must cope with the damage. Tolerance of herbivory, particularly the ability to regrow, must be seen as complementary to resistance to damage. In a number of species or genotypes, tolerance appears to be largely an alternative, a tradeoff against resistance. This was found in 82 percent of cases reported in the literature in which genetic background was controlled (Strauss et al. 2002).

In radishes, Strauss simultaneously examined traits associated with both tolerance and resistance. The noted tolerance traits were root mass, maximum photosynthetic rate, shoot mass, and leaf area, while the resistance traits included total glucosinolates (feeding deterrents), density of trichomes (entanglement devices on leaves), and flower size and phenology. Strauss was thus able to determine which traits were the best predictors of tolerance. Fitness was evaluated separately in each sex, as number of flowers (male plant fitness) and number of seeds (female plant fitness). Traits of root allocation did help

confer tolerance to some degree in female fitness. Male tolerance, surprisingly, included a negative correlation with allocation to leaves. Moreover, the best predictors of tolerance were, surprisingly, the resistance traits of glucosinolate content (male and female fitness) and trichome density (male fitness). Interestingly, the ability to compensate for damage through male function (flower number) was not tightly correlated with the ability to compensate through female function (seed production); that is, male and female tolerances were not significantly correlated. Investment in fruits limited flower production, and there also appeared to be a tradeoff between petal size and flower number and fruit production. Plants may specialize in their ability to tolerate damage through male versus female function. An audience member suggested this decoupling is related to the evolution of dioecy.

Richard Bostock, also with the University of California–Davis, addressed potential conflicts between the major signaling pathways in plants for defenses against insect herbivores (jasmonate) and defenses against pathogens (salicylate). Commonly, cross-resistance is found within a group; salicylate induces long-lasting resistance against fungi, bacteria, and viruses. Such cross-protection is exploited in agriculture, as by spraying tomatoes with the salicylate mimic, acibenzolar. However, in joint attacks by the animal and the microbial kingdoms, there are more reports of antagonism or tradeoffs than cross-resistance from synergy of the two pathways (Heil and Bostock 2002). The antagonism is puzzling ecologically, evolutionarily, and agriculturally. To help explain how both positive and negative interactions can occur, Bostock and his coworkers manipulated both pathways. They used genetic modifications as well as applications of natural and synthetic signals to plants. They assayed biochemical and gene-expression changes, particularly lipoxygenase, the protein PIN II (potato proteinase inhibitor), a wound-inducible herbivore defense, and a pathogen-resistance protein (P4) as markers for the pathways.

The observed responses did not always follow simple patterns. On the simpler side, salicylate (as from pathogen attack) did reduce the PIN II response against herbivores. Though the counteraction was found in cultivated tomato, it does not appear to result from agricultural selection, since the same effect was found in wild tomatoes. In the wild genotypes, however, salicylate did not affect herbivory by *Spodoptera exigua* caterpillars. Again on the simpler side, but more weakly, jasmonate reduces expression of genes for protection against microbial pathogens (Thaler et al. 1999).

Part of the inconsistency in cross-protection between attacks by different kingdoms lies in the distinction between elicitation and signaling. Negative cross-talk appears most likely when elicitors are applied at the same time or in high doses (Thaler et al. 2002). On a practical scale, both timing and dosages can be adjusted to protect crops against pests both large and small. On the ecological and evolutionary scale, opportunities for future research remain, with regard to the evolved mechanisms and the selection pressures.

Warren Porter (University of Wisconsin) turned to ecological prediction, asking, What kinds of ecosystems are possible in various climates? Are there first principles for ecosystem assembly based on energy balances and behavior of organisms? He focused on animals that, despite their small fraction of biomass, act to structure many ecosystems by activities such as herbivory and soil disturbance. Macroclimate is a given, and it is a major determinant of microclimates in a location. It therefore also determines resource availability (plants, prey animals) and predator activity envelopes.

Large animals and small animals differ in the contributions of routes for energy exchange, resulting in different energetic interpretations of the same physical environments. For large animals, convective energy exchange is poor and radiative exchange is more effective. For small animals possessing thinner boundary layers, the opposite is true. Exchange rates are also responsive to posture and insulating layers. Given a desired core temperature, one may join a model of environmental heat and radiation exchange to models of respiration and of digestive heat and mass balance. The model can be tested for its ability to reproduce known correlations of metabolic rate with animal size. More importantly for present purposes, one can predict microclimate and landscape spaces in which the animals live. A significant concern is whether or not species clump in habitat. Porter considered a three-dimensional space of discretionary mass and energy metabolism (for growth and reproduction, above basal metabolism), identical air and radiative temperature, and body mass. The model does predict clumped distribution in body size for high-digestibility foods, but virtually no clumping in body size with low-digestibility foods (Porter et al. 2000). These results show that the variety and quality of vegetation in a habitat is an important constraint on species diversity in a habitat.

More specific inquiries on single or paired species provided some surprising predictions that merit full testing. For example, in areas of burned forest, the lack of vegetative cover causes large mammals to lose much energy by radiation to the sky at night. This can greatly increase the need for food. Energy balance also appears to drive prey availability for rattlesnakes. The activity times of diurnal ground squirrels and rattlesnakes diverge in summer, while those of the snakes and of nocturnal dusky woodrats converge, giving the snakes an alternate prey base as seasons change. Topography, vegetation, and seasonal shifts in climate have important and predictable impacts on shifts in food web structure on a landscape scale (Porter et al. 2002).

Ecological applications are potentially large. Porter consulted with the American Bird Conservancy, which was concerned about die-offs of endangered bird species in Florida, where a pesticide spray program against mosquitoes was in progress. He reconstructed seasonal changes in microclimates across the state in winter and summer and used the data to compute daily respiratory air volumes of endangered bird species exposed to pesticide spraying. Concentrations of ultrafine-droplet, airborne pesticides had already been calculated, so respiratory exposures could then be computed. The organophosphate

spraying program against mosquitoes was thus indicated as deleterious to endangered birds on San Marcos Island, because it was performed when their respiratory rates were maximal.

All these researchers extend the questions and the methods of both plant and animal traditions of ecophysiological research. Whereas plant ecophysiologicals emphasize the measurement of landscape fluxes and the interpretation in terms of extant vegetation functional groups, these crossover specialists ask what structures the ecosystems. Animals, fungi, bacteria, and even protists—although generally a minor component in a community's biomass—exert strong controls via herbivory, parasitism, and pathogenicity, both with plants and among themselves. Scientists must appreciate these interactions to explain why we have the functional groups that exist today, as well as to predict some large-scale responses to global change. These researchers delve into complex biotic interactions, well beyond the past focus of animal ecophysiology on fluxes of water, energy, and other simple substances. They apply the detailed biochemical and molecular approaches used by many current animal ecophysiologicals to study the larger-scale questions of how the biosphere is structured. Such interdisciplinary inquiry must surely attract increasing numbers of researchers in the very near future.

References cited

- Ehrlich PR, Raven PH. 1964. Butterflies and plants: A study in coevolution. *Evolution* 18: 586–608.
- Feder ME. 2002. Plant and animal physiological ecology, comparative physiology/biochemistry, and evolutionary physiology: Opportunities for synergy. An introduction to the symposium. *Integrative and Comparative Biology* 42: 409–414.
- Heil M, Bostock RM. 2002. Induced systemic resistance (ISR) against pathogens in the context of induced plant defenses. *Annals of Botany* 89: 502–512.
- Lawton JH. 1983. Plant architecture and the diversity of phytophagous insects. *Annual Review of Entomology* 28: 23–39.
- Porter WP, Budaraju S, Stewart WE, Ramankutty N. 2000. Calculating climate effects on birds and mammals: Impacts on biodiversity, conservation, population parameters, and global community structure. *American Zoologist* 40: 597–630.
- Porter WP, Sabo J, Tracy CR, Reichman J, Ramankutty N. 2002. Physiology on a landscape scale: Plant–animal interactions. *Integrative and Comparative Biology* 42: 431–453.
- Press MC, Whittaker JB. 1993. Exploitation of the xylem stream by parasitic organisms. *Philosophical Transactions of the Royal Society of London, B* 341: 101–111.
- Press MC, Scholes JD, Barker MG, eds. 1999. *Physiological Plant Ecology*. Oxford (United Kingdom): Blackwell Science.
- Silvertown J, Dodd M. 1996. Comparing plants and connecting traits. *Philosophical Transactions of the Royal Society of London, B* 351: 1233–1239.
- Strauss SY, Rudgers JA, Lau JA, Irwin RE. 2002. Direct and ecological costs of resistance to herbivory. *Trends in Ecology and Evolution* 17: 278–285.
- Thaler JS, Fidantsef AL, Duffey SS, Bostock RM. 1999. Trade-offs in plant defense against pathogens and herbivores: A field demonstration of chemical elicitors of induced resistance. *Journal of Chemical Ecology* 25: 1597–1609.
- Thaler JS, Karban R, Ullman DE, Boege K, Bostock RM. 2002. Cross-talk between jasmonate and salicylate plant defense pathways: Effects on several plant parasites. *Oecologia* 131: 227–235.
- Ward LK, Hackshaw A, Clarke RT. 1995. Food-plant families of British insects and mites: The influence of life-form and plant family. *Biological Journal of the Linnean Society* 55: 109–127.