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Hyperparasitism

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Introduction

Much of the research on hyperparasitism has been of a descriptive nature, based primarily on studies with dual cultures in synthetic media. The main contributions from these investigations concern the host range of the parasite, the mode of penetration and infection, and the morphological changes of the host and the parasite resulting from parasitism.

More recent studies on hyperparasitism emphasize the effect of environmental factors, especially nutrition, on the susceptibility of the host. Research on the physiology of hyperparasitism has been limited. Nevertheless, this important aspect of the problem should continue to receive increased attention as hyperparasitism is extremely amenable to basic research dealing with the physiology of diseases in general (1).

In contrast to the voluminous literature pertaining to dual culture studies, there is little information regarding the biology of hyperparasitism in nature. Furthermore, there is a dearth of conclusive evidence to indicate that interfungus parasitism is an important factor affecting the survival of fungi in their natural habitat. The association of the purported parasite with a moribund or dead fungus host is cited as evidence that hyperparasitism may occur in nature. It has not been incontrovertibly established in most cases, however, whether hyperparasitism in nature is the cause or the effect of the diseased host. It is indeed essential to determine the existence of hyperparasitism in nature. And if the time, place, and the nature of interfungus parasitism could be ascertained, it would undoubtedly give impetus and new direction to research aimed at controlling phytopathogenic fungi through this antagonistic phenomenon.

Selected literature pertinent to the foregoing topics on hyperparasitism is included in this review. Other reviews more adequately deal with some aspects of hyperparasitism discussed in this paper (2-8).

Terminology

The terms, hyperparasitism, mycoparasitism, direct parasitism, and interfungus parasitism are used in reference to the phenomenon of one fungus parasitic on another. The pathogen of this type of parasitism is known as the hyperparasite, the mycoparasite, or simply as the parasite.

Hyperparasites may be classified into two broad groups based on the mode of parasitism and on its effects on the host. The biotrophic group of parasites secures nutrients from living cells of the host irrespective of whether they can grow on synthetic media (1, 6). Acquisition of nutrients by the biotrophic parasites is via haustoria or hyphae in close contact with host mycelia without formation of haustoria. Biotrophic parasites which kill their host or its parts during their development are called destructive parasites, whereas those that inflict little or no obvious damage to the host are known as balanced parasites. Also included with the biotrophic parasites are the obligate hyperparasites which obtain nutrients only from living cells.

The second group of mycoparasites are the necrotrophic parasites encompassing those fungi which derive nutrients from dead host cells, usually killed by the parasite before it invades the host (6). It seems that such fungi behave more like saprophytes than parasites. It is worthy of note that soilborne fungi are attacked primarily by necrotrophic and destructive hyperparasites.

Tropism

Hyperparasitism involves an intimate contact between the host and the parasite. Whether the establishment of such an association results from fortuitous contact of these microorganisms or from a tropic response of the host or the parasite, is a vexed question. The limited knowledge regarding tropism of hyperparasitism is predicated on studies made in synthetic media. Suffice it to say that results from such investigations cannot be applied forthwith to what may be occurring in a natural habitat such as the soil, the rhizosphere, and the host tissue.

The ingenious experiments of Butler (9) showed that parasitism of host hyphae by *Rhizoctonia solani* may be initiated by a tropic response involving a contact stimulus. Hyphae of *R. solani* responded thigmotropically to glass tubing, glass wool, and cotton fiber by coiling around these materials. Furthermore, the parasitic hyphae occasionally penetrated the walls of the cotton fiber. However, the intensity of coiling and penetration was considerably less sparse than that observed on host hyphae. He surmised that in addition to thigmotropism, other factors may be contributing to the abundant development of hyphal coils and infection hyphae formed by *R. solani* in contact with the host. It would be interesting to know whether contact stimulus between host and hyperparasite mediates the production of substances which enhance formation of hyphal coils and infection hyphae.

Tropic response of host hyphae toward germinated spores of the balanced mycoparasite, *Calcarisporium parasiticum*, has been reported (1). Contacts between *C. parasiticum* and the hosts, *Phyosalospora obtusa* and *Phyosalospora* spp., cannot always be ascribed to chance. In this connection, the host hyphae were stimulated to produce short, lateral branches which grew toward the germinated spores of the parasite. Less often the tips of the main hyphae of the host would depart from their normal course and grow directly toward the hyperparasite. Barnett & Lilly (1) postulated

that the tropic response of the host hyphae is elicited by a chemical substance emanating from germinating spores of the parasite. The purportedly chemical substance affecting host response is active in high dilutions and is very potent in inciting host response at distances up to 40 μ in agar media. The strength of the tropic response was dependent on the distance between host and parasite, the stage of spore germination, the composition of the substrate, and the species of the host.

In contrast to the foregoing tropic response of the host hyphae toward the parasite, the hyperparasites, *Gleocladium roseum* and *Piptocephalis virginiana*, are attracted directly to the host cells (10).

Mode of Parasitism

The mechanisms of hyperparasitism are as diversified and as complex as are those for parasitic relationships involving higher plants and microorganisms. Studies on the mode of mycoparasitism have focused attention primarily on the general morphology of the structures for securing nutrients from the host. There is a dearth of information relative to physiological aspects of mycoparasitism. It should also be stated that nearly all of the experiments on the mode of hyperparasitism were made with dual cultures grown on synthetic media. The recent findings of Siegle (11) provides a detailed account on the mode of parasitism of *Didymella exitiales* on the host fungus, *Ophiobolus graminis*, in the rhizosphere of wheat. This new approach to the problem should help to pave the way to future studies on mycoparasitism as it occurs in nature.

Several different modes of hyperparasitism have been observed. The balanced mycoparasites derive nutrients from host cells in three general ways. One means is by the production of haustoria within host cells. Brefeld (12) was the first to report on this mode of parasitism in describing the development of haustoria of *Piptocephalis fresiniana*. Subsequently, haustorial formation was noted with other balanced mycoparasites, including several species of *Piptocephalis* (13, 14), and *Dispira cornuta* (15).

A second means of parasitism by balanced hyperparasites involves the dissolution of the host wall in contact with the parasites, *Parasitella simplex* and *Chaetocladium* sp. (16). In this case, the host nuclei migrate through a hole of the cell wall into a specialized basal cell of the parasite in contact with the host.

More recently, Barnett & Lilly (1) described a third means of parasitism incited by the balanced mycoparasite, *Calcarisporium parasiticum*. This unique mode of parasitism is characterized by a small, specialized, septate, contact cell called the buffer cell. Contact cells are produced at the tips of the mycoparasitic hyphae. Unlike the previously mentioned modes of parasitism, the specialized contact cell of the parasite does not pierce or dissolve the host hyphae. It is speculated that the buffer cell functions physiologically to increase permeability of the plasma membrane of the host and to absorb nutrients from the host. A similar mode of parasitism was observed (17) for *Gonatobotryum fuscum*. However, this balanced hyperparasite differs from *C. parasiticum* in that the small hypha in contact with the host lacks a discrete buffer cell.

The mode of parasitism of the destructive hyperparasites may be by direct penetration of the host, thereby establishing haustoria or internal mycelia or by coiling around the host hyphae with or without penetration. Examples of these kinds of parasitism are discussed by Butler (9) for the mycoparasite, *Rhizoctonia solani*, on various hosts. In the case of *Penicillium vermiculatum* parasitizing *R. solani*, Boosalis (18) showed that the penetration pegs develop either from hyphae in direct contact with host mycelium or from mycelium coiling around host hyphae.

Following penetration, the infection mycelium of the destructive parasites grow and may permeate parts or the entire hyphal element of the host (18). According to Butler (9), *R. solani* may infect a few host hyphae within 24 hr after contact, with maximum infection occurring at 48 to 72 hours. The rate and intensity of infection is greatly affected by environmental conditions, particularly temperature and nutrition (9, 18). Hyphal branching commonly develops within parasitized hyphae and may also occur at the junction of branching of the host hyphae. The number of hyphae produced by *P. vermiculatum* within host hyphae varies from one to four, the majority with one. Butler (9) and Boosalis (18) reported that hyphae of *R. solani* and *P. vermiculatum*, respectively, do not penetrate the hyphal wall of the host from the inside. Furthermore, the wall of the parasitized host may collapse but it does not disintegrate.

The destructive hyperparasite, *Didymella exitiales*, penetrates the hyphae of *Ophiobolus graminis* in several ways (11). In one instance, the hyphae of *D. exitiales* grow toward those of *O. graminis* and follow them for a way without parasitism being detectable. Only after a minimum incubation of 12 days, *D. exitiales* penetrates the host by means of thin hyphae which grow within the host. After 16 days incubation, the parasitized cells of *O. graminis* are devoid of protoplasm. A second method of penetration is accomplished by the tips of hyphal branches of *D. exitiales* boring through the cell wall of the host. In this case, penetration is apparently attained without mechanical pressure. Siegle (11) also noted that penetration of the host may be initiated by means of appressoria produced by *D. exitiales* as a result of certain mechanical or chemical stimuli.

Some investigators (9, 18, 19) found that destructive mycoparasites do not produce antibiotic or other deleterious materials which diffuse through the medium in advance of the hyphae. Unlike *Trichoderma lignorum* and other necrotrophic, antibiotic-producing fungi, the destructive mycoparasites do not initiate their parasitic activities at a distance. It would appear that an intimate association of the host and parasite is a *sine qua non* for the production of substances initiating direct parasitism.

A more detailed description of the foregoing mechanisms of parasitism, as well as other, more subtle host parasite relationships, are presented in recent review articles (2-8).

Host Structures Parasitized

Besides the mycelium, many of the other structures of the host are attacked by the mycoparasites. *Gliocladium roseum* parasitizes and de-

stroys the conidia of many species of fungi (19). However, only the immature conidiophores and conidia of *Helminthosporium sativum* are attacked by *C. roseum*. Campbell (20) found, on synthetic media, that the conidia of *H. sativum* were invaded and killed by *Myrothecium verrucaria*, the conidia and mycelia by *Epicoccum purpurascens*, whereas only the hyphae were invaded by *Phoma humicola*. Destructive mycoparasites grown in dual cultures on synthetic substrates, and in some instances in sterilized soil, grew within sporangiophores (9), sporangia (9), oogonia (21), chlamydospores (22), oospores (23, 24, 25), and zoospores (26) of some of the Phycomycetes. The ubiquitous mycoparasites, *Darluca filum* and *Tuberculina* spp., grow within urediospores, teliospores, and pycniospores of many of the Uredinales (7). The stroma of *Dibotryon morbosum* and other phytopathogenic fungi are parasitized by *Trichothecium roseum* (7). Tribe (27) stated that the sclerotia of *Sclerotinia trifoliorum* were parasitized by *Coniothyrium minitans*. The comprehensive review article by DeVay (3) on mycoparasitism lists additional hyperparasites and the specific host structures they affect.

It should also be stated that the foregoing modes of penetration of host mycelia, with the exception of specialized contact cells produced by certain balanced parasites, have also been recorded in mycoparasitic studies of the aforementioned host structures.

Effect of Parasitism on Host and Parasite

The effect of parasitism on the host, ranges from no apparent damage to death (9, 15, 18). The destructive mycoparasites usually cause disintegration of the host protoplasm (9, 18). For these parasites which grow within the host hyphae, it is not always clear whether death of the protoplasm results before or after penetration of the host. It is noteworthy that following destruction of the host protoplasm the internal hyphae of some parasites undergo autolysis (22). However, the mycelia of other parasites within the host may persist long after the host protoplasm has disappeared (9, 18).

Dreschsler (23, 24) stated that after the oospores of certain hosts were emptied, the protoplasmic contents of the haustoria withdrew into the external hyphae of the parasite. These kinds of responses by the parasite and by the host increase the difficulties of detecting mycoparasitism in natural habitats.

Some of the balanced parasites produce no damage to the host, except a reduction in rate of growth (1). The most benign type of mycoparasitism exemplified by the interaction between *Dispira cornuta* and the host hyphae approaches a neutralistic relationship (15).

Stromata of *Xyteria oxyacanthae* infected by *Fusidium parasiticum* became shrunken, disintegrated and the ascospores abort (28).

The parasitic activities of *Hypomyces lactifluorum* suppressed gill formation and sporulation in fruiting bodies of *Lactarius piperatus* (2).

Marked hypertrophy of hyphae, sporangia, and septation of phycomycetous hyphae were incited by the hyperparasites, *Razellopsis* spp. (29).

Hyperparasitism may strongly stimulate sexual reproduction of the parasite. The excellent research of Haskins (30) revealed that a destructive hyperparasite *Pythium* species, probably *Pythium acanthicum*, requires some nutritional fat-solvent extractable substance(s) for development of its sexual stages. This substance was not found in the living or dead nonfruiting mycelium of the hyperparasite. However, the parasite can obtain the nutritional factor for sexual reproduction from parasitized hyphae of many species of fungi, including other species from the same genus. He also noted that this pythiaceous hyperparasite grew readily on killed mycelium of some of the hosts and produced sexual spores. It is worthy of note that the nutritional factor required for sexual reproduction is not requisite for parasitism. In this case, the mycoparasite is capable of parasitizing certain species of fungi without the production of sexual spores. These hosts evidently lacked the substance necessary for sexual reproduction but contained the factors needed to sustain parasitism. The nutritional substance is not restricted to host fungi as dead plant materials and the living plants themselves stimulated sexual reproduction of this *Pythium* sp. (30).

Factors Affecting Parasitism

It is axiomatic that the degree of parasitism is greatly affected by intrinsic (hereditary) factors and a variety of extrinsic factors. These two types of factors influencing parasitism are presented separately for reason of convenience. In reality, however, parasitism is generally influenced by the interaction of the hereditary factors of the host and parasite with the external environmental factors. Unfortunately, there is a paucity of rigorous studies on the interrelation of factors affecting mycoparasitism.

Intrinsic factors.—The effect of the intrinsic constitution of the host on parasitism is reflected in various ways. For example, the stage of development of the host may relate to its susceptibility. In this connection, Slifkin (31) showed that the initiation of sex cells or zoosporangia inhibited parasitism of *Olpidiopsis incrassata* on members of the *Saprolegniaceae*. Moreover, when the oogonia or sporangia were excised, the host reverted to the vegetative stage and became susceptible to the parasite. It was speculated that some chemical necessary for infection by the parasite is also a precursor for the reproductive stage of the host (31).

Susceptibility may also be contingent on the age of the host hyphae. Only the young, rapidly growing host hyphae are most susceptible to attack by the destructive hyperparasite, *Rhizoctonia solani* (9). On the other hand, the age of the mycelium is not a factor of susceptibility of several hosts attacked by some balanced hyperparasites (1). Studies are urgently needed on the chemical nature of the cell wall to explain the relationship of susceptibility and age of the host.

Results from studies *in vitro* indicate that the host may resist infection by forming mechanical barriers against internal mycoparasites. Infection hyphae of *R. solani* are frequently restricted by a protective sheath of wall-like material produced by the host (9). Parasitism may be restrained by some phycomycetous hosts by the formation of septa ahead of the in-

vading hyphae. Also, thickening of the inner side walls of some *Pythium* spp. at the site of appressorial attachment obstructed invasion by hyphae of another *Pythium* sp. (21).

Lysis of parasitic hyphae within host cells, and the genetic variability within a species of the parasite and the host, are two other hereditary factors affecting parasitism (9).

Extrinsic factors affecting parasitism.—Nutrition is considered to be one of the most important factors affecting hyperparasitism. One reason for this is that the physiological and biochemical systems of the host relating to susceptibility are apparently influenced by the quantity or quality, or both, of nutrients. In the following studies the susceptibility of the host was modified, in most instances, by varying the nutrients in various media. Results from these investigations, however, offer little information on the physiological nature of parasitism in relation to nutrition.

The kind or amount of carbohydrate may influence the incidence of parasitism. Boosalis (18, 32) showed that *Rhizoctonia solani* was severely parasitized by *Penicillium vermiculatum* when grown on sand-corn meal or potato-dextrose agar with a relatively high concentration of dextrose (20 gm/l). Parasitism was negligible, however, when the host was cultured on potato-dextrose agar containing 10 gm dextrose per liter. It appeared that the high level of dextrose increased the amount of parasitism by increasing the susceptibility of the host. Butler's findings (9, 33) indicate that the kind of carbohydrate supplied to the host and parasite greatly affected parasitism. *Mucor recurvis* is highly susceptible to *R. solani* on inorganic-salt agar media containing hexose or a di-, or polysaccharide composed of hexose residues, maltose, and on grain media of corn meal, oats, or rice. Infection was sparse or absent when the host was cultured on substrates containing pentose sugars, galactose, lactose, cellobiose, or on fresh peas or wheat germ.

Results from studies with balanced mycoparasites showed that susceptibility of the host can be altered by changing the nutritional composition of the medium. The pioneer work of Ayers (15) on the effect of nutrition on *Dispira cornuta* indicated that the kind rather than the quantity of nutrient altered the susceptibility of the host. *Dispira cornuta* was severely attacked on media containing different concentrations of proteose-peptone but not on media supplied with different quantities of dextrose as a nutrient. Slifkin (31a) studied the effect of carbon source on the parasitism of three species of *Saprolegnia* by the balanced obligate hyperparasite, *Olpidiopsis incrassata*. In general, *S. kauffmaniana*, *S. diclina*, and *S. delica* are susceptible to *O. incrassata* when grown in a medium containing hexose sugars which support good growth of the hosts. As an example, all three hosts were severely parasitized in a medium with D-glucose, β -glucose, D-levulose and D-mannose. On the other hand, although *S. delica* grew sparsely in D-galactose, it was, nevertheless, severely parasitized by *O. incrassata*. It is interesting to note that the other two species of *Saprolegnia* did not grow on the medium with D-galactose. Mannitol and D(+)-cellobiose stimulated some host growth but rendered all three hosts immune to the hyperparasite. Starch, glycogen, and xylose supported fairly good host growth and rendered the hosts susceptible.

Other investigators subsequently showed that the quality as well as the quantity of the nutrient may modify the degree of parasitism. For example, findings from studies on the balanced parasites, *Calcarisporium parasiticum* (1), *Piptocephalis virginiana* (34, 35), and *P. xenophila* (35), indicated that the degree of parasitism on some hosts is directly correlated to the amount of available nitrogen, and inversely correlated to the concentration of sugar in the medium. On the other hand, maximum parasitism of some hosts by *Gonatobotryum fuscum* was observed in media with high amounts of carbon and low amounts of nitrogen (17). Another host, *Leptographium* sp., however, was susceptible only in a substrate high in nitrogen (17).

Susceptibility is also affected by the ratio of carbon to nitrogen in the medium. The susceptibility of some hosts on media with a high carbon-nitrogen ratio is decreased (1, 35), whereas with other hosts, it is increased (17). Berry (34) discovered that the degree of parasitism was about the same for some hosts as long as the glucose-glutamic acid ratio was 3:1. With other hosts, however, the severity of parasitism varied when the amount of carbon and nitrogen changed, even though the carbon-nitrogen ratio remained the same. The susceptibility of *Saprolegnia kauffmaniana*, to the balanced hyperparasite, *Olpidiopsis incrassata*, is greatly influenced by the ratio of carbon to nitrogen (31). Generally, the higher the ratio of carbon, the greater the degree of parasitism on the three species of *Saprolegnia*. Slifkin (31) also found that although the degree of susceptibility was not necessarily the same with equal ratios of carbon to nitrogen, it was with different sources of nitrogen. Thus, when glucose was used with urea at a ratio of 11:1, *S. delica* was immune. However, at this same ratio, *S. delica* was highly susceptible when *L*-asparagine was the nitrogen source.

It was also reported that the carbon and nitrogen sources, such as different ammonia compounds and different amino acids, markedly influenced the degree of parasitism of some hosts (1, 17, 34, 35).

The findings of Shigo et al. (35) disclosed a direct correlation between the amount of nitrogen in the medium and the percentage of soluble nitrogen in the host mycelium. Furthermore, they stated that "there is a direct correlation between the amount of soluble nitrogen in the mycelium and the growth of *Piptocephalis* on its host." Interpretation of these results with respect to susceptibility, however, is difficult since it is not known whether the modification of parasitism, as estimated by growth of the parasite on the host, is in response to nutrients obtained from the host or medium, or both.

The degree of parasitism is also modified by the addition of microelements to the medium. Parasitism of some hosts was enhanced when Mn alone was added to the substrate, or when a mixture of Mn, Fe, Zn, and Ca were incorporated in media containing asparagine, phenylalanine, or potassium nitrate as the nitrogen source (35). The severest attack of *Graphium* sp. by *Gonatobotryum fuscum* occurred on a medium containing Mg and Mn (17). The role of microelements in relation to mycoparasitism is not known.

Results from recent studies on balanced hyperparasites indicate that susceptible and immune reactions may be contingent on whether a growth

factor, required by the parasite, is present in the host and whether it is available to the pathogen. In this connection, Barnett & Lilly (1) showed that a small amount of a water extract of chopped mycelium of the host or of some immune species stimulated growth of the balanced parasite in synthetic media. A similar amount of extract from another immune species, however, did not increase growth of the pathogen. It was concluded from this, that the parasite was able to absorb the growth factor from the susceptible host but not from the immune species. The absence of the growth factor in still other species may account for their immunity. Whether this purported growth factor is present in living, intact cells of susceptible species, and whether it affects parasitism is a moot question.

Temperature.—In most studies on hyperparasitism involving balanced and destructive parasites, temperature was shown to affect the degree of parasitism. In some instances, the severity of parasitism was not altered by change of temperature. The balanced mycoparasite, *Piptocephalis virginiana*, vigorously parasitized *Thamnidium elegans* at 25° C, but did not attack the host at 15° and 20° C. With another host, however, parasitism occurred at 15° and 20° C, but not at 25° C. Both of these hosts grew moderately to well at 15° to 30° C (34). The host range of the balanced parasite, *Olpidiopsis incrassata*, was the same at temperatures of 6°, 20°, and 30° C. Moreover, changes in temperature from 10° to 25° C at intervals of 5° temperature did not alter susceptibility of the host to this parasite (31). The optimum temperature for parasitism with other balanced and destructive hyperparasites was 25° to 30° C (1, 9, 17, 33). Working with unsterilized soil, Boosalis (18) reported that the incidence of mycoparasitism on *Rhizoctonia solani* was considerably higher in green manure-amended soil at 28° than at 18° C.

Light and pH.—Although it is unlikely that light and pH are major factors affecting hyperparasitism in nature and especially in the soil, these factors, nevertheless, can influence host-parasite relations *in vitro*. Shigo et al. (17) stated that several hosts were strongly parasitized by *Gonotobotryum fuscum* in total darkness, whereas parasitism was poor under conditions of alternating light and darkness. According to Butler (9), infection of the host by *R. solani* was not influenced by diffused daylight; but it was suppressed by artificial light of a higher intensity. The absence or presence of light did not alter the degree of parasitism in studies with the balanced parasite, *Olpidiopsis incrassata* (31).

Severe infection of *Pythium debaryanum* and *P. butleri* by *R. solani* developed in a medium with a pH 5.5 and 7.1, whereas a pH 6.7 and 7.1 was not conducive to infection of *Rhizopus* spp. (9). Parasitism of *Armillaria mellea* by the necrotrophic parasites, *Trichoderma* spp., was substantially reduced by adjusting the pH of the medium below 5.1, and at pH 7.0 parasitism was inhibited (36).

The effect of pH on the obligate hyperparasite, *Olpidiopsis incrassata*, and on three species of *Saprolegnia*, the host fungi, was recently studied in a synthetic medium (31). The hosts, *Saprolegnia kauffmaniana*, *S. diclina*, and *S. delica*, tolerated a pH range of 4.3 to 8.5. The minimum and maximum levels for the hyperparasite were pH 5.0 and 8.0, respectively.

The wide pH range between these two extremes did not alter immunity or susceptibility of the three hosts to *O. incrassata*.

Other organisms.—Relatively little work has been done to determine what effect other microorganisms exert on host-parasite relation *in vitro*. Furthermore, the author is not aware of any research on this important problem in soil. The significant work of Butler (9) on this phase of hyperparasitism revealed that infection was completely inhibited, limited, or not affected by the presence of another fungus in culture with the host and parasite.

Host Range

There is an abundance of literature regarding the host range of different kinds of hyperparasites. A few of the more pertinent papers are cited on this subject. Comprehensive compilations of mycoparasitic hosts are available elsewhere (2, 3).

The varying capacity of mycoparasites to infect hosts ranges from limited to extensive. A few parasites are capable of attacking only one species (7, 18, 28). The host range of other mycoparasites is confined to a number of species of a single genus of one class (15, 17, 31). A larger number of parasites have a predilection for hosts belonging to the *Mucorales* (10, 15, 23, 34). The more diversified mycoparasites, especially those isolated from soil or diseased roots, parasitize fungi of two or more classes (1, 9, 19, 37).

A *Pythium* sp. isolated from an excised rust-infected leaf (probably *Amelanchier alnifolia*) parasitized 69 fungus species representative of the four classes of fungi (30). This destructive hyperparasite also caused browning of the root tips of several crops in pots containing unsterilized soil.

Relevance of the accumulated knowledge regarding the host range of hyperparasites has led to some interesting speculation. For example, Butler (9) hypothesized that perhaps the capacity of certain soil-inhabiting fungi to parasitize roots as well as other fungi is related to their effective survival in nature. Barnett & Lilly (1) suggested that it may be possible to use mycoparasites to identify closely related species of fungi, especially the nonsporulating groups. The fact that a hyperparasite, parasitic on phytopathogenic fungi, can be killed by other mycoparasites obviously adds to the difficulties of devising biological control measures based on interfungus parasitism.

Occurrence of Hyperparasitism in Nature

Systematic studies have not been pursued to ascertain the frequency of mycoparasitism in soil and other habitats. Consequently, it is not possible to assess accurately the importance of interfungus parasitism in nature. The occurrence of mycoparasitism in some habitats, however, is well documented.

Hyperparasitism has been associated frequently with above-ground diseased tissues of plants. The mycoparasite, *Gonatobotryum fuscum*, was found to parasitize *Graphium* sp. growing in oak trees (17). This parasite also infected *Ceratocystis* spp. located under loose bark of a felled beech

tree (38). Similar natural habitats of mycoparasitism have been noted by other investigators (19,28,39,40). According to Matta (41), *Cercospora unamunoi*, parasitic on leaves of pepper grown in the greenhouse, was parasitized by the destructive hyperparasite, *Botrytis yuae*. The plant disease induced seemed to be alleviated by the presence of *B. yuae*.

Most of the evidence for the natural occurrence of hyperparasitism in soil is circumstantial. For instance, mycoparasitism was established on glass slides buried in sterilized soil infested with pure cultures of the host and parasite (42). This, in itself, certainly does not constitute adequate proof for the existence of mycoparasitism in soil. In another study, it was found that sublethal dosages of carbon disulfide applied to natural soil stimulated *Trichoderma viride* to invade and kill *Armillaria mellea* (43). The infection of species of *Phythium* by *Chytridiaceous* parasites was observed in natural soil (44). Drechsler (23) described destructive mycoparasitism of oospores of *Phythium graminicolum* on leaf mold. Boosalis (18) reported that *Rhizoctonia solani*, introduced into unsterilized soil amended with green manure and kept at 28° C, was parasitized by soil-inhabiting fungi. Even under such favorable environmental conditions, however, only about 18 percent of the host hyphae were parasitized. In subsequent studies with *R. solani*, Boosalis (unpublished data) discerned only six parasitized hyphae out of 15,000 screened in June and September from natural soil collected from eight fields cropped with sugar beet.

Hyperparasitic Control of Phytopathogenic Fungi

Attempts to control fungus diseases of plants through hyperparasitism have met with very little success. Investigations on mycoparasitic control of fungi have dealt primarily with soil-borne pathogens (18). In essence, the attempted methods of biological control consisted of adding copious amounts of the mycoparasite to the soil. In some instances, the pH of the soil was adjusted (45), or the soil was amended with nutrients with the aim of stimulating mycoparasitism. Such treatments are not suitable for the control of fungus diseases of plants because they do not give consistent protection and are commercially impractical.

Summary and Discussion

Much of the work on hyperparasitism has been done on synthetic media. In most instances the experimental procedure entailed the transfer of pure cultures of the parasite and the host fungus on an agar or broth substrate, allowing them to grow until parasitism was established. Whatever the shortcomings of these procedures, considerable information on certain etiological and physiological aspects of mycoparasitism was derived from dual culture studies.

Microscopic observations of the initial stages of hyperparasitism in synthetic media have revealed the devious and intricate means by which the destructive mycoparasites penetrate and invade the host cells. The mode of parasitism by the more benign balanced hyperparasites has also been resolved through similar investigations. Microscopic detection of

structural changes of some hosts in response to infection contributes to a better understanding of the nature of resistance. Considerable information on the effect of the environment on the susceptibility of the host has resulted from research with dual cultures. With respect to nutrition, it was shown that the degree of parasitism may be contingent on the source and concentration of nitrogen, sugar, or both, contained in the medium. The diversity of parasitism is clearly illustrated in dual culture studies on the host range of different hyperparasites. Some parasites are capable of infecting only one host species, whereas others have a wider host range attacking species of several classes of fungi. The foregoing facts, together with other information on interfungus parasitism gained from dual culture studies, provided the framework for subsequent investigations on mycoparasitism.

Research on the physiology of hyperparasitism was initiated recently to define the underlying biochemical systems associated with the sequence of events leading to infection. Exploratory investigations with dual cultures in synthetic media and in the rhizosphere of wheat have yielded clues on the physiology of mycoparasitism which warrant further study. The pioneer work of Barnett & Lilly (1) and of Shigo (17) with balanced mycoparasites, indicates that susceptibility is correlated with the presence of growth-promoting substances and with high amounts of soluble nitrogen within the host mycelium. These workers also found that susceptibility was increased by adding microelements to the substrate. The role of the unidentified growth-promoting substances, as well as that of the soluble nitrogen material and the microelements, is unknown. The research of Siegle (11) sheds some light on the chemical and physiological nature of penetration of the host fungus by the destructive parasite, *Didymella exitialis*. The results from studies on hyperparasitism in the rhizosphere of wheat are significant in showing that destruction of *Ophiobolus graminis* results from penetration and subsequent invasion of the hyphae as well as from amino acids produced by the parasite.

Ecological investigations on soil-borne mycoparasites have been neglected primarily because methods for this type of research have been lacking. Consequently, there is little information on the occurrence of hyperparasitism and on the biology of mycoparasites in soil. There is little evidence to show that many of the mycoparasites isolated from soil or diseased plant tissues and tested for pathogenicity in dual cultures are truly mycoparasitic in soil. This may partly explain why the additions of fungi to soil, shown to be mycoparasitic in dual cultures only, fail to parasitize the host fungus in the soil milieu. The survival of fungi in soil *per se* does not appear to be greatly affected by mycoparasitism. In this connection, Boosalis (unpublished data) did not detect any appreciable amount of parasitized hyphae of *R. solani* screened from several fields with different cropping histories. This does not necessarily mean, however, that hyperparasitism is not significant in certain microhabitats of the soil. Evidence is presented by Siegle (11) that mycoparasitism may be common in the rhizosphere of plants. With the recently devised methods for detecting and isolating fungi from soil (46-50) it should be possible to undertake systematic studies to

ascertain whether mycoparasitism occurs in the various soil microhabitats, such as plant residue, and in the rhizosphere of plants.

It is indeed imperative to know when and where the hyperparasitic wars occur before pursuing studies to learn what factors can affect the outcome of the struggle. Knowledge gained on the ecology of mycoparasites, coupled with a better understanding of the physiology of hyperparasitism, should suggest the needed strategy to turn the tide of battle. And if the tide can be directed in favor of the hyperparasites through various permutations of crop rotation, soil amendments, cultural practices, or through some other means, man will reap the spoils of hyperparasitic victories.

Literature Cited

1. Barnett, H. L., and Lilly, V. G., Parasitism of *Calcarisporium parasiticum* on species of *Phylospora* and related fungi, *West Va. Univ. Agr. Expt. Sta. Bull.*, **420T**, 36 pp. (1958)
2. Buller, A. H. R., *Researches on fungi*, **2**. (Longmans, Green & Co., London, 492 pp., 1922)
3. DeVay, J. E., Mutual relationships in fungi, *Ann. Rev. Microbiol.*, **10**, 115-40 (1956)
4. Sanford, G. B., Root-disease fungi as affected by other soil organisms, in *Plant Pathology, Problems and Progress 1908-1958*, 367-76 (Holton, C. S., et al., Eds., Univ. Wisconsin Press, Madison, Wis., 1959)
5. Weindling, R., Role of parasitism in microbial antagonisms, in *Recent Advan. Botany, Inter. Botan. Congr., IX, Montreal*, 623-26 (1959)
6. Barnett, H. L., Plant disease resistance, *Ann. Rev. Microbiol.*, **13**, 191-210 (1959)
7. Darpoux, H., Biological interference with epidemics, In *Plant Pathology, an Advanced Treatise*, 3, 521-65 (Horsfall, J. G., and Diamond, A. E., Eds., Academic Press, New York, 1960)
8. Brian, P. W., Antagonistic and competitive mechanisms limiting survival and activity of fungi in soil, *The Ecology of Soil Fungi*, 115-29 (Parkinson, D., and Waid, J. S., Eds., Liverpool Univ. Press, Liverpool, 1960)
9. Butler, E. E., *Rhizoctonia solani* as a parasite of fungi, *Mycologia*, **49**, 354-73 (1957)
10. Berry, C. R., and Barnett, H. L., Mode of parasitism and host range of *Piptocephalis virginiana*, *Mycologia*, **49**, 374-86 (1957)
11. Siegle, H., Über mischinfektionen mit *Ophiobolus graminis* und *Didymella exitialis*, *Phytopathol. Z.*, **42**, 305-48 (1961)
12. Brefeld, O., *Mucor, Chaetocladium* and *Piptocephalis*, *Botan. Unters., über Schimmelpilze*, **6**, 41-45 (1872)
13. Dobbs, C. G., and English, M. P., *Piptocephalis xenophila* sp. nov., parasitic on non-murcorine hosts, *Trans. Brit. Mycol. Soc.*, **37**, 375-89 (1954)
14. Leadbeater, G., and Mercer, C., *Piptocephalis virginiana* sp. nov., *Trans. Brit. Mycol. Soc.*, **40**, 461-71 (1957)
15. Ayres, T. T., Parasitism of *Dispira cornuta*, *Mycologia*, **27**, 235-61 (1935)
16. Burgeff, H., Untersuchungen über Sextalitat und Parasitismus bei Mucoriceen I., *Goebel's Botan. Abhandl., Heft IV* (1924)
17. Shigo, A. L., Parasitism of *Gonatobotryum fuscum* on species of *Ceratocystis*, *Mycologia*, **52**, 584-98 (1960)
18. Boosalis, M. G., Effect of soil temperature and green-manure amendment of unsterilized soil on parasitism of *Rhizoctonia solani* by *Penicillium vermiculatum* and *Trichoderma* sp., *Phytopathology*, **46**, 473-78 (1956)
19. Barnett, H. L., and Lilly, V. G., A destructive mycoparasite, *Gliocladium roseum*, *Mycologia*, **54**, 72-77 (1962)
20. Campbell, W. P., The influence of associated microorganisms on the pathogenicity of *Helminthosporium sativum*, *Can. J. Botany*, **34**, 865-74 (1956)
21. Drechsler, C., Two species of *Pythium* occurring in southern states, *Phytopathology*, **33**, 261-99 (1943)
22. Godfrey, R. M., Studies of British species of *Endogone*, II, Fungal parasites, *Trans. Brit. Mycol. Soc.*, **40**, 136-44 (1957)
23. Drechsler, c., Two hyphomycetes parasitic on oospores of root-rotting oomycetes, *Phyto-*

- pathology*, **28**, 81-103 (1938)
24. Drechsler, C., Another hyphomycetous fungus parasitic on *Pythium* oospores, *Phytopathology*, **33**, 227-33 (1943)
 25. Drechsler, c., A slender-spored *Dactylella* parasitic on *Pythium* oospores, *Phytopathology*, **53**, 1050-52 (1963)
 26. Barrett, J. T., Development and sexuality of some species of *Olpidiopsis* (Cornu) Fisher, *Ann. Botany (London)*, **26**, 209-38 (1912)
 27. Tribe, H. T., On the parasitism of *Sclerotinia trifoliorum* by *Coniothyrium minitans*, *Trans. Brit. Mycol. Soc.*, **40**, 489-99 (1957)
 28. Bachus, M. P., and Stowell, E. A., A *Fusidium* disease of *Xylaria* in Wisconsin, *Mycologia*, **45**, 836-47 (1953)
 29. Karling, J. S., A synopsis of *Rozella* and *Rozellopsis*, *Mycologia*, **34**, 193-208 (1942)
 30. Haskins, R. H., Morphology, nutrition, and host range of a species of *Pythium*, *Can. J. Microbiol.*, **9**, 451-57 (1963)
 31. Slifkin, M. K., Parasitism of *Olpidiopsis incrassata* on members of the *Saprolegniaceae*, I, Host range and effects of light, temperature and stage of host on infectivity, *Mycologia*, **53**, 183-93 (1961)
 - 31a. Slifkin, M. K. Parasitism of *Olpidiopsis incrassata* on members of the *Saprolegniaceae*, II, Effect of pH and host nutrition, *Mycologia* **55**, 172-82 (1963)
 32. Boosalis, M. G., *Penicillium* sp. parasitic on *Rhizoctonia solani*, Abstr., *Phytopathology*, **44**, 482 (1954)
 33. Butler, E. E., and King, T. H., Fungus parasitism by *Rhizoctonia*, Abstr., *Phytopathology*, **41**, 5 (1951)
 34. Berry, C. R., Factors affecting parasitism of *Piptocephalis virginiana* on other Mucorales, *Mycologia*, **51**, 824-32 (1959)
 35. Shiga, A. L., Anderson, C. D., and Barnett, H. L., Effects of concentration of host nutrients on parasitism of *Piptocephalis xenophila* and *P. virginiana*, *Phytopathology*, **51**, 616-20 (1961)
 36. Aytoun, R. S. C., The genus *Trichoderma*: its relationship with *Armillaria mellea* (Vahl ex Fries) Quel and *Polyporus schweinitzii* Fr., together with preliminary observations on its ecology in woodland soil, *Trans. Botan. Soc. Edinburgh*, **36**, 99-114 (1953)
 37. Weindling, R., *Trichoderma lignorum* as a parasite of other soil fungi, *Phytopathology*, **22**, 837-45 (1932)
 38. Vincent, M., A *Chalaropsis* on beech, *Nature*, **172**, 963-64 (1953)
 39. Barnett, H. L., A new *Calcarisporium* parasitic on other fungi, *Mycologia*, **50**, 497-500 (1958)
 40. Shiga, A. L., Fungi isolated from oakwilt trees and their effects on *Ceratocystis fagacearum*, *Mycologia*, **50**, 757-69 (1958)
 41. Matta, A., Segnalazione della *Cercospora unamunoi* (Unam.) E. Castel. e di. un suo parasitica su peperone in serra, *Ann. Fac. Sci. Agrar. Univ. Studi Torino*, **1**, 307-12 (1962)
 42. Warren, J. R., An undescribed species of *Papulospora* parasitic on *Rhizoctonia solani* Kuhn, *Mycologia*, **40**, 391-401 (1948)
 43. Darley, E. F., and Wilbur, W. D., Some relationships of carbon disulfide and *Trichoderma viride* in the control of *Armillaria mellea*, Abstr., *Phytopathology*, **44**, 485 (1954)
 44. Butler, E. J., An account of the genus *Pythium* and some Chytridiaceae, *India Dept. Agr. Mem. Botan. Serv.*, **VI**, 1, No. 5 (1907)
 45. Weindling, R., and Fawcett, H. S., Experiments in the control of *Rhizoctonia* damping-off of citrus seedlings, *Hilgardia*, **10**, 1-16 (1936)
 46. Baker, R., and Martinson, C., The isolation of fungi by means of the soil microbiological sampling tube, *Prot. Intern. Botan. Congr.*, IX, 2, 15 (1959)
 47. Boosalis, M. G., and Scharen, A. L., Methods for microscopic detection of *Aphanomyces euteiches* and *Rhizoctonia solani* and for isolation of *Rhizoctonia solani* associated with plant debris, *Phytopathology*, **49**, 192-98 (1959)
 48. Durbin, R. D., Techniques for the observation and isolation of soil microorganisms, *Botan. Rev.*, **27**, 527-60 (1961)
 49. Johnson, L. F., Curl, E. A., Bond, J. H., and Fribourg, H. A., *Methods for studying soil microflora disease relationships* (Burgess Publ. Co., Minneapolis, 178 pp., 1959)
 50. Menzies, J. D., The direct assay of plant pathogen populations in soil, *Ann. Rev. Phytopathol.*, **1**, 127-42 (1963)