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SHORT COMMUNICATION

THE EFFECTS OF *GIBBERELLA ZEAE* ON RNA, PROTEIN AND DRY WEIGHT IN MAIZE SEEDLINGS*

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Abstract—NaCl-soluble RNA, tris-glycine buffer soluble protein and dry weight of *Gibberella zeae* infected maize seedling shoots were altered quantitatively from that observed daily (days 5–9) for the corresponding healthy tissues.

IN STUDIES concerning different host-parasite relationships, changes in metabolite synthesis have been of great interest. Several investigators showed that synthesis of RNA,[1–3] and proteins,[4] and changes in dry weight[4] within different host-parasite relationships reached a maximum over that of noninfected tissues during a stage of advanced pathogenic development. A rapid decrease in these constituents followed as continued senescence of the host occurred.

This work is a preliminary investigation of the quantitative changes in NaCl-soluble RNA (*s*RNA), Tris-glycine buffer soluble protein (soluble-protein) and per cent dry weight in three *Gibberella zeae* infected and noninfected maize seedling lines, Ohio 45 and West Virginia 12, and their hybrid (12 × 45), during a period of days 5–9.

Similar to the investigations discussed above, quantitative changes in *s*RNA, soluble-protein and dry weight were associated with the development of the *G. zeae*-maize host-parasite relationship when compared to the daily patterns of the noninfected seedling shoots (Figs. 1–3). These constituents were greater in the infected than in the noninfected tissues with development of the fungal mycelium upon the diseased shoots and the occurrence of the symptoms of infection (after 5–6 days; depending on the rate of infection in the diseased tissues within and between experiments). These quantitative differences continued until approximately 50% water-soaked rotting was apparent (6–8 days). Upon further rotting, these constituents decreased faster in the infected than in the noninfected plants (after 8 days). For example, these developments correlated with the following statistically different changes: infected Ohio 45 had a higher *s*RNA content than the noninfected control shoots from 6–7 days (Fig. 1). The $sRNA_i$ (*s*RNA content of infected tissue) in West Virginia 12

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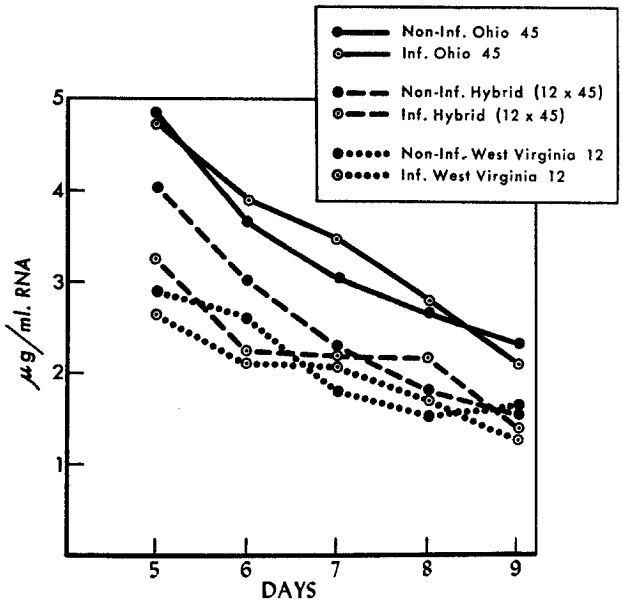

FIG. 1. COMPARISON OF *s*RNA CONTENT IN NONINFECTED (Non-Inf.) AND INFECTED (Inf.) MAIZE SEEDLING SHOOTS.
Values are the means of three replications.

and the hybrid was lower than $sRNA_n$ (*s*RNA of noninfected tissue) at 5–6 days and increased to a content greater than the latter before and after 7 days, respectively, continuing through day 8. A decline in $sRNA_i$ to below that of $sRNA_n$ occurred in all three maize lines after 8 days. The three lines of maize infected with *G. zeae* produced higher percentages

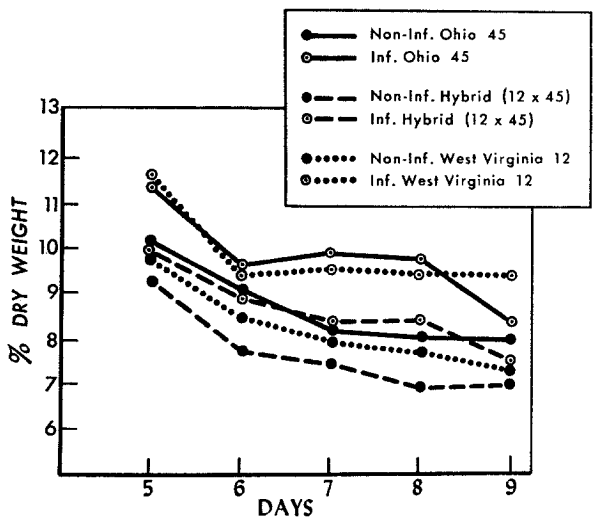

FIG. 2. COMPARISON OF DRY WEIGHT OF Non-Inf. AND Inf. MAIZE SEEDLING SHOOTS.
Values are the means of an accumulated number of experiments including those run in preparation for this analysis.

of dry weight than the noninfected tissues after 5 days and continued to do so through day 9 (Fig. 2). At day 6, the soluble-protein content of the infected tissue was significantly greater than that of the noninfected shoots in all three maize lines (Fig. 3). Nonsignificant soluble-protein differences between infected and noninfected tissues were observed before and after 6 days; except for Ohio 45 at day 9. These statistically significant changes in *s*RNA, dry weight and soluble-protein correlated with the development of the fungus on, and the destruction of, the host tissues in their respective experiments.

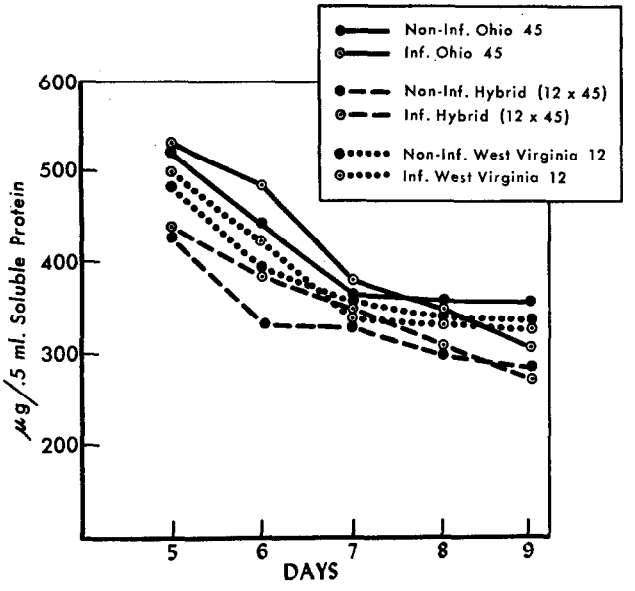

FIG. 3. COMPARISON OF SOLUBLE-PROTEIN IN Non-Inf. AND Inf. MAIZE SEEDLING SHOOTS. Values are the means of three replications.

The experiments showed that the rates of changes of the *s*RNA, soluble-protein and dry weight in the infected tissues varied within and between experiments (Figs. 1–3). Possibly, these variations could be attributed to the differences in mycelial contact with the shoot tissue (which possibly determines the rate of fungus infection), since the seedlings were grown without nutrients to eliminate metabolic variations due to varied absorption through the diseased root systems. In addition, variations in the rate of seedling growth and moisture content may have contributed further to the fluctuations between experiments.

The changes in *s*RNA, soluble-protein and dry weight may be due in part to the presence of fungal as well as plant metabolites. Other investigations of host-parasite relationships by cytological,[5] enzymatic[6] and microspectrophotometric[7,8] techniques have shown that quantitative and qualitative biochemical changes occur in both of the organisms. However, most of this activity is in the host and is induced by the parasite.[7,8] Analyses such as the latter are needed to further elucidate the biochemical changes in the host-parasite relationship of *G. zeae* and maize.

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EXPERIMENTAL

Two inbred lines of maize, West Virginia 12 and Ohio 45, susceptible and resistant to *G. zeae* in the adult earing stage, respectively, and their hybrid (12 × 45) of intermediate resistance were germinated at 26° in complete darkness on paper towels containing an inoculum of culture-grown fungus.[9] It was observed that resistance was not expressed in the seedling stage during these experiments. The noninoculated group was prepared similarly except the fungus inoculum was omitted. Shoot tissue was collected daily from days 5–9 by cutting at the base. External mycelial tissue was removed from the seedling shoots. The samples were weighed before and after drying at 80° to determine the per cent dry weights. A modified method of the combined techniques developed by Zscheile and Murray[10] and Malca *et al.*[1], was used to extract, purify and analyse the *s*RNA fraction (0·55 M NaCl-soluble). In the present study no pyrimidines were eluted from the columns of Dowex 1-X8 resin (200–400 mesh) with the pH 1·1 HCl elutions. All adsorbed bases examined in this analysis were eluted in the first 100 ml, pH 2·2 HCl fraction. Further studies are needed to elucidate this latter problem. The soluble-protein was extracted by homogenizing fresh plant material in tris-glycine buffer (0·1 M; pH 8·3) followed by centrifugation at 17,300 *g* for 30 min in a refrigerated centrifuge. The clear supernatant was then analysed for protein using the Lowry method.[11] The Duncan's New Multiple Range Test was used for statistical analysis of all data. Significant differences were taken at the 5% level. The F-test for testing the null hypothesis of no differences between replications was accepted for all studies included in this paper.

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Key Word Index—Lea Mays; Gramineae; *Gibberella zeae*; infection; ribonucleic acid; protein.