The Genetic Basis of Plant-Pathogen Interaction

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Plant pathogens display different degrees of specificity. Some organisms are pathogenic on a broad host range, whereas others are limited to certain species. Also, more complex systems have developed that involve intricate race-cultivar relationships. Recent models by Heath (15) and by Bushnell and Rowell (4) emphasize that successful parasites have evolved the genetic capacity to overcome the host's defense mechanisms. This interaction establishes a basic compatibility between species. Further genomic interactions may evolve that produce a complex system of race-cultivar specificity. In both models, race-cultivar specificity is attributed to a single gene-for-gene relationship in which genetically determined interactions between products of the host cultivar and the parasitic race condition incompatibility. A system in which the genetic determinants of the host and parasitic race do not correspond results in compatibility. In this letter, we stress the evolutionary importance of gene regulation and its possible role in race-cultivar specificity. The point of our letter is to emphasize that major genetic changes conditioning specificity may be at regulatory loci rather than in structural genes. We define regulatory loci as those sites that control the timing and expression of other genes (16). The events governing changes in specificity may occur at random in loci that regulate the onset of the pleiotropic reactions of a resistance mechanism.

The greater significance of changes at regulatory rather than structural gene loci in the evolution of plant pathogen specificity is suggested by work in other eukaryotic systems on the molecular basis of differentiation and speciation (6,8,19,25,29,34,35). Evolutionary changes may require more than the gradual random accumulation of single mutations affecting single structural loci. The rate of base substitution at loci determining protein structure cannot account for the rate of phenotypic change for some animal lineages (33). Amino acid replacement in structural genes proceeds at similar rates in all organisms. Wilson (33) proposed that evolution at the organismal level may depend upon mutation at regulatory loci that alter the pattern of gene expression. An example of this phenomenon is found in the increased amounts of alcohol dehydrogenase detected in *Drosophila* selected through 28 generations for an increased tolerance to ethanol (21). The alcohol dehydrogenase molecules had not undergone a change in amino acid sequence, but the control of their synthesis had been altered to provide more enzyme. Studies with hybrids from crosses between *Drosophila melanogaster* and *D. simulans* suggest that related species differ in the manner in which a structural locus is regulated (30). Tepper et al (31) and Richmond and Tepper (24) showed that regulatory loci may be polymorphic within natural populations of a *Drosophila* species providing the variability required for selection.

Regulatory loci may control structural gene expression in a number of ways, including altered rates of transcription, mRNA processing, translational and post-translational modifications, or differential rates of degradation. These varied mechanisms of regulation are under intensive study. Recently, some coordinately regulated structural genes have been shown to share common regulatory sequences. Regulated cotranscription of members of one type of repetitive DNA sequence has been linked to expression of a set of developmentally related structural genes in *Dictyostelium* (38). These observations pertain to a classical model for regulation developed between 1969 and 1979 by Britten and Davidson (3,9). This model was based on the hypothesis that short sequences of repetitive DNA may have a regulating function in eukaryotic genomes (3).

The Britten and Davidson hypothesis has been used as the basis for models of plant-pathogen specificity proposed by Day (10) and modified by Callow (5) in which the host resistance genes are envisioned as sensor genes. Day (10) proposed that sensor genes regulate different combinations of producer genes, the expression of which results in a resistant response. Callow (5) also incorporated Roseman's (26) suggestion that the sensor genes function through membrane receptors. In Callow's (5) proposal, the product of the pathogen's avirulence gene, the elicitor, binds to the sensor receptor which results in a specific membrane messenger that interacts with a regulatory DNA sequence. Activation of the appropriate regulatory loci would trigger the expression of the many structural genes associated with resistance. These proposals extend an earlier model published in 1969 by Hadwiger and Schwochau (14) who suggested that products from the invading microorganism induce host resistance by eliminating certain gene control mechanisms. The regulatory mechanism in this model was likened to the repressor system displayed by the *lac* operon in *Escherichia coli*.

The concept of regulatory control of resistance mechanisms is supported by the physiological events that accompany incompatibility. Resistance in the plant often involves a hypersensitive response in which there is severe metabolic disturbance of cells in the zone of contact with the challenge. Hypersensitivity is a pleiotropic response that involves such notable changes as increasing plasmalemma permeability (32), the production of low molecular weight phytoalexins and polyphenols (18), and the eventual necrosis of the responding plant cell (27). The occurrence of these responses is highly controlled. The production of any phenolic structure is energy demanding and has been demonstrated in hypersensitivity to involve de novo enzyme synthesis (18,37). Phytoalexins and polyphenols cannot be produced at random in plant cells because they are themselves phytotoxic (28). Yet the accumulation of phytoalexins and polyphenols in plant tissues can be triggered by chemicals including heavy metals (36) and several types of elicitors that are of plant and fungal origin (1,7,13,22). Ultraviolet irradiation (2) or freezing (23) of plant tissue will stimulate phytoalexin production. Plant cell necrosis, browning, and phytoalexin production also are associated with late stages of some compatible plant-pathogen interactions (17,18). Consequently, a key feature of these incompatibility responses is their relatively early initiation. To capitalize on these reactions as part of a defense system against a microbial challenge, the plant must have evolved intricate triggering systems. Therefore, we emphasize that regulation of the pleiotropic reactions of resistance are of vital significance to the control of specificity. The genes that have been demonstrated by genetic crosses to condition resistance in incompatibility could be the genes that regulate the onset of the pleiotropic response of hypersensitivity.

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Compatibility must involve the negation of potential resistance mechanisms, including the triggering of the hypersensitive response. As suggested by Bushnell and Rowell (4) and Heath (15), the pathogen in a compatible interaction may fail to initiate resistance mechanisms because it produces a suppressor of a general elicitor. Acquisition of race-specific resistance in a previously compatible host may involve the selection of a new regulatory locus that controls the induction of the hypersensitive response. The new regulatory locus results in recognition of another elicitor from the challenge and the consequential activation of the many structural genes involved in the resistance event. Bushnell and Rowell (4) suggested that the pathogen may then mutate to virulence through development of new specific suppressors that prevent the regulatory locus from initiating the expression of the structural genes. An alternative scheme is that evolution of virulence in the pathogen may involve structural alteration of an elicitor so that it no longer triggers a resistance response. Further mutation by the plant to recognize a new type of elicitor would be required to generate the resistance response. The races would differ by the nature of their specific elicitors rather than the specific suppressors (4). Independent of the mechanism of the system, we emphasize that the events governing specificity are associated with regulatory loci in the host that control expression of the structural genes functioning in the resistance response.

The mechanisms of regulatory control remain speculative. If the genes determining resistance control transcription of the structural genes, differences between isogenic cultivars may not be apparent in extracts of the unchallenged plants subjected to electrophoresis and staining for protein composition. Differences between the isogenic lines may be demonstrated only by examining DNA nuclease cleavage patterns of these cultivars or by examining untranslated RNA from the nuclei of the challenged plant tissue.

The possibility that there are several loci involved in regulation of the pleiotropic changes in the expression of resistance is consistent with the present knowledge of plant and pathogen behavior. It seems reasonable that more than one locus regulates resistance events, because some of the reactions associated with hypersensitivity also occur in responses of the plant to other stimuli. If changes at a locus can affect specificity without causing loss of the regulatory function, allelism at that locus should be possible. Regulatory loci could vary in the extent of possible allelism. Multiple loci and different levels of allelism at some of the loci for resistance have been shown for barley and wheat powdery mildew (11) and the flax-rust systems (12). Physiological studies of different incompatible race-cultivar pairs in these systems have revealed that the timing and the cellular changes involved in the hypersensitive event can be quite distinct. Littlefield (20) observed that incompatiblity in the flax-rust system conditioned by the *L* and *N* genes is highly distinct from the responses determined by the *M* and *P* genes. Lesser, but noticeable, differences were observed in the physiology of the response conditioned by the *L* rather than the *M* locus or the *M* compared to the *P* locus. Ellingboe (11) showed that the genes for resistance in barley and wheat to powdery mildew act at different times and possess distinct physiological characteristics. However, incompatible responses determined by different alleles of the same locus were not physiologically distinguishable from each other. Cultivars of bean also vary in physiological expression and in timing of the metabolic changes within the plant cells in response to incompatible races of *Colletotrichum lindemuthianum* (27). These observations support Littlefield's (20) conclusion that hypersensitivity is not a single phenomenon without variation. Clearly, the resistance response can be achieved through events that have different modes of expression and timing. Based on the observations, data, and models summarized in this letter, we have concluded that genetic changes involved in evolution of plant pathogen specificity may be at regulatory rather than structural loci.

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