Biochemistry

 Volume 27, Number 25 December 13, 1988

Perspectives in Biochemistry

Oligosaccharides as Recognition Signals for the Expression of Defensive Genes in Plants

Clarence A. Ryan

Institute of Biological Chemistry and Program in Biochemistry and Biophysics, Washington State University, Pullman, Washington 99193

Received June 16, 1988; Revised Manuscript Received August 26, 1988

ABSTRACT: Oligosaccharides derived from cell walls of fungi and plants, including β-glucans, chitin, chitosan, and pectin, are inducers of the synthesis of a wide spectrum of defensive chemicals in plant tissues. These oligosaccharides are generated at infection or wound sites and may be early signals to activate genes whose products, such as antibiotic phytoalexins, extensin, proteinase inhibitors, pathogenesis-related proteins (PR proteins), and lignin, enhance the plants' defenses against pathogens and herbivores. Inducing activities of β-glucans, derived from fungal cell walls, have been found to reside in a heptaglucosyl oligosaccharide having β-1,3 and β-1,6 linkages. Active oligomers derived from chitin (α-1,4-acetylglucosamine polymers) and chitosan (α-1,4-glucosamine polymers) have a minimum DP of about 5. Linear oligogalacturonans (α-1,4 linkages), purified from plant cell walls with DP = 9–12, are inducers of localized defensive responses in plants against fungi. However, oligogalacturonans of DP = 2 and higher are inducers of proteinase inhibitors in plants that respond systemically to insect attacks. The available evidence suggests that an oligosaccharide-based communication system may be present in plants as an early warning to activate defensive responses. However, little is presently known of the biochemistry or molecular biology of intercellular or intracellular events that regulate the defensive genes in response to the oligosaccharide signals.

Roles for oligosaccharides as signals for the regulation of gene expression in plants have been under investigation for only about 15 years (Darvill & Albersheim, 1984; Ryan, 1987). Oligosaccharides from both fungal and plant origins have been identified that can act as chemical signals to activate a broad spectrum of plant genes that are involved in numerous and varied responses that protect the plants against predators or pathogens. a few reports have also implicated oligosaccharides of plant origin in the regulation of aspects of plant growth and development (Darvill & Albersheim, 1984), but most reports have been concerned with their roles in plant defense. Unlike the N-linked oligosaccharides that are involved in recognition systems in animals and yeast, the well-characterized carbohydrates that activate plant defensive genes are not covalently attached to proteins. They are relatively small oligomers that are hydrolytic fragments derived from cell walls of attacking pathogens or pests or from the cell walls of the plant itself (Darvill & Albersheim, 1984; Ryan, 1987). The ability of these oligosaccharides to alter gene expression patterns has stimulated renewed interest in intracellular and intercellular signaling processes in plants. At the present time little is known of the intracellular recognition mechanism for any plant hormone or how plant hormones activate gene expression. It is of fundamental importance to determine if plants have evolved an oligosaccharide-based recognition-communication system to regulate the expression of genes that have functions unique to plants, such as inducible defensive responses. Additionally, when intracellular recognition and signaling systems involving oligosaccharides are elucidated in plants, it will be of interest to know if an evolutionary relationship exists between plant recognition systems and the oligosaccharide recognition and signaling systems of animals.

Roles for carbohydrates in recognition and signaling processes in eucaryotic organisms have been the subject of intensive investigation for the past 35 years (Ashwell & Harford, 1982). Since the initial demonstration that a carbohydrate moiety on the surface of erythrocyte membranes was essential for the attachment and entry of influenza virus (Burnet, 1951), N-linked glycans have been implicated in a broad spectrum of recognition processes, including the clearance of glycoproteins from the circulation by hepatic or reticuloendothelial cells (Ashwell & Harford, 1982; Clark et al., 1987; Weigel,

0006-2960/88/0427-8879$01.50/0

1987), cellular adhesion (Muller & Garish, 1978; Brodie et al., 1983; Chadwick & Garrod, 1983; Steineman & Parrish, 1987; Ziska & Henderson, 1988), hormone action (Sairam & Bhargavi, 1985), cytolysis (Ahrens & Ankel, 1987), metastasis (Dennis et al., 1987), and cell–cell recognition (Weinstock & Ballou, 1986), as well as the regulation of many cellular functions (Ashwell & Harford, 1982). Within the past 5 years, several comprehensive reviews have been written on the structure, biosynthesis, and function of N-linked carbohydrates (Ashwell & Harford, 1982; Baenziger, 1984; Goldstein et al., 1984; Schachter, 1984; Yet et al., 1987). Knowledge of the structures and biosynthesis of N-linked carbohydrates has facilitated interest in their possible roles in recognition systems in various aspects of cell growth, differentiation, and metabolism. Yet et al. (1987) recently stated "... the best one-word proposal for the function of N-linked glycans would seem to be recognition, meaning that the glycans represent specific chemical counterparts and translated into specific molecule–molecule, cell–molecule, and cell–cell interactions with associated dynamic consequences". However, both Yet et al. (1987) and Ashwell and Harford (1982) have pointed out that even though many eucaryotic systems are now known in which oligosaccharides play important roles in recognition, a unified understanding of carbohydrates as informational molecules has remained obscure.

Oligosaccharides That Signal Defensive Responses in Plants. Several types of fungal and plant cell wall derived oligosaccharide structures have been identified that induce defensive responses in plant tissues (Darvill & Albersheim, 1984; Ryan, 1987). Two structurally different types of oligosaccharides, the β-glucans and chitin fragments (or chitosan fragments), originate from the cell walls of fungi, whereas the others, α-1,4-oligogalacturonides, are parts of the plant cell wall. All of these fragments, when added to tissues of various dicotyledonous plants, have been demonstrated to induce the expression of genes that are involved in defensive responses.

Figure 1 illustrates the types and origins of the various oligosaccharides that induce defensive responses in plants. The β-glucans were the first oligosaccharides found that were inducers or "elicitors" of the synthesis of antibiotics, called phytoalexins (West, 1980; Darvill & Albersheim, 1984), in plant tissues. The phytoalexins are broad-spectrum antibiotics that are not found in healthy plant tissues but are synthesized in cells near sites of pathogen infections as part of the plant's defensive response. While studying the chemical nature of signals from fungal pathogens that caused soybean tissues or suspension-cultured cells to synthesize phytoalexins, Albersheim and his associates (Ayres et al., 1976) isolated a β-glucan-enriched fraction from the cell walls of the fungi *Phytophtera megasperma* that was a powerful inducer (elicitor) of the isoflavinoid phytoalexin glyceollin. The inducing activity was associated with the glucan fracton of the cell walls that was composed of β-1,6- and β-1,3-glucosyl linkages. The activity was abolished by incubating the fraction with exo-1,3-β-glucanase. Glucan-enriched fractions have since been isolated from cell walls of other fungi (Cline et al., 1978) and from yeast (Hahn & Albersheim, 1978) that also possess potent phytoalexin-inducing activities. Fungal cell wall derived glucans have been identified as active inducers of a variety of defensive responses in many different plant genera and species (West, 1980; Darvill & Albersheim, 1984; Ryan, 1987). Among the defensive responses is the induction of the biosynthesis of the fungal cell wall degrading β-glucanases (and chitinases) (Darvill & Albersheim, 1984; Mauch et al., 1984; Legrand et al., 1987; Rombrink et al., 1988), which can degrade fungal cell walls, producing fragments that can amplify the signaling process (Hadwiger & Beckman, 1980). The β-glucans induce a number of specific biochemical changes associated with resistance, such as increases in the concentrations of biosynthetic enzymes for the mevalonate and phenylpropanoid pathways leading to the synthesis of phytoalexins (West, 1980; Bruce & West, 1982; Dixon et al., 1983; Darvill & Albersheim, 1984; Dixon, 1986; Ryan, 1987) and lignin (Grand et al., 1987); production of the volatile plant hormone ethylene (Chappell et al., 1984); synthesis of the structural, hydroxyproline-rich glycoproteins (Showalter & Varner, 1987); and synthesis and accumulation of proteinase inhibitors (Esquerre-Tugyae et al., 1985) and pathogenesis-related (PR) proteins (Kombrink et al., 1988). To date, the biological activities of β-glucans have been associated only with

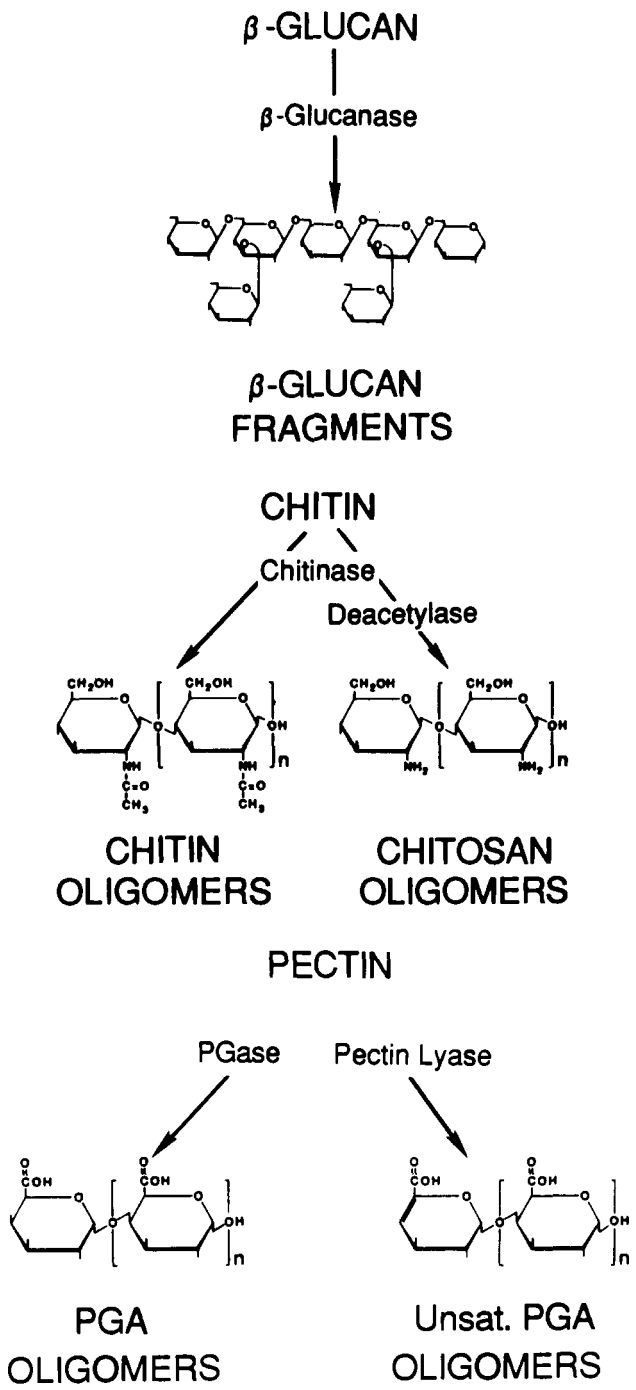

FIGURE 1: Enzymatic degradation of fungal and plant cell walls produces oligosaccharide signals that induce defensive responses in plant tissues.

the activation of defensive responses to pathogen attacks. There is no evidence to suggest that fungal-derived β-glucan fragments regulate other metabolic processes in plant cells.

A glucan heptasaccharide composed of β-1,3 and β-1,6 linkages, having powerful inducing activity for the synthesis of the phytoalexin glyceollin in soybeans, has been isolated from acid hydrolysates of cell walls of the pathogenic fungi *P. megasperma* (Sharp et al., 1984). Of the theoretically possible 150–300 heptasaccharide structural isomers obtainable from the cell walls, only oligomers containing the structure shown in Figure 1 (top) possessed activity. This heptaglucose is the most potent inducer of phytoalexin synthesis reported to date, being active in soybean tissues at about 1 pg/g of tissue (Sharp et al., 1984). Although the heptasaccharide is highly active in inducing the antibiotic glyceollin, it is not known whether it is an active inducer of other defensive chemicals in soybeans. Whether the single oligosaccharide can activate many defensive responses, or whether different oligosaccharide structures can determine specificity to regulate complex signaling patterns in plants to recognize different pathogens, remains to be determined.

Oligosaccharide fragments derived from the fungal cell wall polymers, chitin (β-1,4-*N*-acetylglucosamine) and chitosan (β-1,4-glucosamine), are also potent inducers of defensive responses in plants (Hadwiger & Beckman, 1980). Although not as thoroughly investigated as the fungal cell wall β-glucans, chitin and chitosan fragments are active inducers of many, if not most, of the responses that are induced by the glucans (Ryan, 1987). Chitin and chitosan are large insoluble polymers within fungal cell walls but can be hydrolyzed to produce soluble fragments by the combined action of the enzymes chitin deacetylase, chitinase, and/or chitosanase or by acid hydrolysis (Hadwiger & Beckman, 1980; Hadwiger & Loschke, 1981; Walker-Simmons & Ryan, 1984). Oligomeric fragments of chitosan with DPs of 11 or less are soluble in aqueous solutions and those with DP = 6–11 have been shown to be inducers of various defensive responses in tissues of a number of plant genera (Hadwiger & Beckman, 1980; Hadwiger & Loschke, 1981; Walker-Simmons & Ryan, 1984). Incuded in these responses is the induction of the enzyme chitinase that can amplify the signaling process by releasing chitin fragments from the cell walls of attacking fungi. The similarity of signaling in plants by β-glucan and chitosan oligomers has led to speculations that both may activate fundamentally similar intracellular signaling mechanisms (Ryan et al., 1987).

Primary cell walls of plants contain pectic polysaccharides that, when enzymically or chemically fragmented, produce oligomers of galacturonic acid that can signal the activation of defensive responses (Darvill & Albersheim, 1984; Ryan, 1987). Plant cell walls are a complex mixture of polysaccharides and glycoproteins that constitute about 90% of the wall (Darvill & Albersheim, 1984). These polysaccharides are composed of about 20–30% cellulose (β-1,4-glucosyl units) and 70–80% complex carbohydrates that are a mixture of methylated galacturonic acid, rhamnose, xylose, arabinose, galactose, glucose, fucose, and other sugars present in small amounts (Darvill & Albersheim, 1984). The structural details of the plant cell wall are still mostly undefined. Oligogalacturonans derived from plant cell walls by degradation with endopolygalacturonase (PGase) or pectic lyase (PG lyase or pectin transeliminase) (Figure 1, bottom), or by mild acid hydrolysis, are inducers of many, if not most, of the defensive responses in plants that are induced by β-glucans or chitin (or chitosan) fragments (Ryan, 1987). The inducer (elicitor) activities of the fragments have been shown to be associated

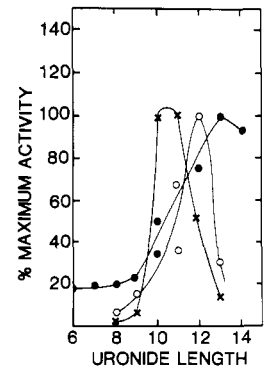

FIGURE 2: Relationships of degree of polymerization (DP) of oligogalacturonans to their activities in inducing the synthesis of the phytoalexin glyceollin in soybean cotyledons (○) (Nothnagel et al., 1983), casbene synthetase in castor bean cotyledons (●) (Jin & West, 1984), and lignin in cucumber seedlings (×) (Robertson, 1986).

with relatively small α-1,4-galacturonic acid oligomers derived from the backbone of the soluble pectin or pectin-like materials present in the walls. Size–activity relationships of oligouronans have been determined for four different plant defensive responses: (i) the induction of the biosynthesis of isoflavinoid phytoalexin antibiotics in soybean cotyledons (Nothnagel et al., 1983); (ii) the induction of the enzyme casbene synthetase, the final step in the biosynthesis of casbene, a diterpene phytoalexin in castor bean cotyledons (Jin & West, 1984); (iii) the induction of lignin biosynthesis in cucumber seedlings (Robertsen, 1986); and (iv) the synthesis of proteinase inhibitor proteins in tomato leaves (Bishop et al., 1984). The inductions of the first three responses differ in two important aspects from the induction of proteinase inhibitors. One difference is that the synthesis of phytoalexins and lignin is localized within the general proximity of the regions of wounding or pathogen attacks (West, 1980; Dixon et al., 1983; Darvill & Albersheim, 1984; Dixon, 1986; Showalter et al., 1986), whereas the inhibitor response is systemic, occurring in cells of unwounded leaves several centimeters away from leaves on which the wound or infection occurred (Ryan, 1978). A second fundamental difference is in the minimal sizes of fragments that can induce the responses. Phytoalexins and lignin require a minimum oligouronide length of about nine sugar residues, whereas the proteinase inhibitor proteins are induced by di- or triuronides as well as larger oligomers (Bishop et al., 1984). In Figure 2 are shown the relationships of oligomers of differing lengths with their ability to induce or elicit phytoalexins and lignin. Oligouronides with sizes less than DP = 9 were essentially inactive. The maximal inducing activity resided in DPs of about 10–14. Oligouronide fragments, prepared by fragmenting poly(galacturonic acid) with pectin transeliminase, having nonreducing termini as Δ-4,5-galacturonic acid, were as fully active as the corresponding sized normal oligouronides in inducing soybean phytoalexins and proteinase inhibitors (Davis et al., 1986; Ryan, 1987), indicating that the 4-OH group of the nonreducing terminal uronide is not essential for inducing activity.

No long-distance effect other than proteinase inhibitor synthesis is known to be induced by oligosaccharides. It is not clear how the di- or trigalacturonides might be involved in the hormonal-like effects of the proteinase inhibitors, but evidence has been presented to show that radioactive pectic fragments added to wounds are not transported out of the wounded leaves (Baydoun & Fry, 1985). This implies that there is an unidentified second messenger that must be released, or synthesized and released, in response to the oligouronides. This messenger would be systemically transported throughout the

plants where it could activate the expression of proteinase inhibitor genes.

A number of oligosaccharide-induced genes that code for defensive proteins are being analyzed by various approaches in several laboratories to identify the regions of the genes that interact with putative transacting factors that are regulated by the addition of oligosaccharide fragments that are added to tissues or cells. The intracellular signaling events that take place between the release of poly- or oligosaccharide elicitors, or inducers from fungal cell walls or plant cell walls during pathogen or predator attacks, and the activation of defensive genes in the plant nuclei are poorly understood. The signaling system(s) appear(s) to be a complex network that controls the temporal appearance of different defensive proteins or enzymes (Lamb et al., 1987). Usually, several defensive genes, but not all, are expressed in response to a pathogen attack or to an exogenously applied oligosaccharide signaling molecule (Lamb et al., 1987; Ryan, 1987). Some genes, such as those that code for the biosynthetic enzymes that synthesize phytoalexins, are activated very rapidly (Lamb et al., 1987; Somssich et al., 1986), whereas others, such as those coding for hydroxyproline-rich glycoproteins or for enzymes of lignin biosynthesis, respond after a delay of several hours (Grand et al., 1987; Showalter et al., 1987). The sequence of appearance of defensive gene products in response to oligosaccharide signals can differ among plant genera or species. Nothing is known of the mechanism of the temporal regulation of these events. It is possible that the carbohydrates of all types are signaling cells nearby infection or wound sites to activate genes that produce a series of secondary signals that, in turn, are transported systemically to distal tissues to activate short-term defensive genes such as proteinase inhibitors or long-term defensive systems such as the PR proteinase (Somsich et al., 1986). Some defensive responses continue for several days or weeks (Rhodes, 1979; Kuc, 1981), suggesting that the initial signals (oligosaccharides) may also cause changes in nearby cells that mediate those long-term effects.

β-Glucan elicitors and oligouronides have been shown to activate genes for various defensive responses at the transcriptional level (Chappell & Hahlbrock, 1984; Cramer et al., 1985; Graham et al., 1986; Somsich et al., 1986; Lawton & Lamb, 1987; Hedrick et al., 1988). In intact plants, mRNAs coding for defensive genes are usually found within 2 h following elicitor addition to the tissues. In parsley suspension cultures mRNAs coding for pathogenesis-related proteins could be detected within 5 min after exposure to an elicitor from the cell walls of *P. megasperma* (Somssich et al., 1986). In cultured bean cells treated with the β-glucan inducer from the walls of *Collectotrichum lindemuthanium*, mRNAs coding for three enzymes of the biosynthesis of the phenylpropanoid pathway leading to phytoalexin synthesis, and mRNA coding for chitinase, were detected in the cytoplasm within 10–20 min after addition of the inducer polysaccharides (Lamb et al., 1987). Transcripts of the mRNAs could be detected in nuclear extracts within 5 min. These examples are among the earliest transcriptional events reported for plant genes in response to an exogenous signal and resemble the kinetics found in some hormonal responses in mammals. This leaves little time for complex signal transduction events to take place between the extracellular recognition of inducer polysaccharides and the activation of the genes in the nucleus (Lamb et al., 1987).

Data concerning possible receptors for poly- or oligosaccharide signals in plants are limited. Plasma membranes of soybean have been reported that possess a single affinity class of binding sites for β-glucan preparations from the cell walls of the plant pathogenic fungus *P. megasperma* (Yoshikawa et al., 1983; Schmidt & Ebel, 1987). However, the putative receptor(s) has (have) not been isolated, nor has any biochemical event been associated with it. A possible association of Ca^{2+} with oligosaccharide signaling has been reported (Kohle et al., 1985; Staub & Ebel, 1987), and the possible involvement for ion currents (Davies, 1988; Ojalvo et al., 1987) and phoshoinositides (Kurosake et al., 1987) has been proposed. Recent data (T. F. Farmer, G. Pearce, and C. A. Ryan, in preparation) have shown that oligouronides cause the phosphorylation of a small protein in plasma membranes isolated from tomato and potato leaves. However, a clear conceptual unifying mechanism that explains the role of carbohydrates and/or other molecules in signaling the activation of plant defensive genes or of the intracellular events that regulate them remains to be established.

The availability of defined oligosaccharides, such as the heptaglucoside and small glucosamine oligomers from fungal cell walls and the small oligogalacturonides from plant cell walls, now provides materials that may be utilized for the preparation of radioactively labeled oligomers or derivatives, such as photoaffinity probes, to study their biological activities. With both the concepts and techniques that have been developed with animal hormonal systems, numerous studies are in progress to seek possible relationships between the plant signaling systems and those of other eucaryotes. Whether the signaling systems are similar in some respects to those of animals (or yeast), or are totally different, remains to be determined. In any event, it is anticipated that within the next few years a fundamental conceptual framework for oligosaccharide signaling will be elucidated in both higher and lower eucaryotic organisms.

References

Ahrens, P. B., & Ankel, H. (1987) *J. Biol. Chem. 262*, 7575–7579.

Ashwell, G., & Harford, J. (1982) *Annu. Rev. Biochem. 51*, 531–554.

Ayres, A. R., Ebel, J., Rinelli, F., Berger, N., & Albersheim, P. D. (1976) *Plant Physiol. 57*, 751–759.

Baenziger, J. U. (1984) in *The Plasma Proteins* (Putnam, F. W., Ed.) Vol. IV, pp 271–315, Academic, New York.

Baydoun, E. A.-H., & Fry, S. C. (1985) *Planta 165*, 269–276.

Bishop, P. D., Makus, D., Pearce, G., & Ryan, C. A. (1984) *J. Biol. Chem. 259*, 13172–13177.

Brodie, C., Klein, C., & Swierkoz, J. (1983) *Cell (Cambridge, Mass.) 32*, 1115–1123.

Bruce, R. J., & West, C. A. (1982) *Plant Physiol. 69*, 1181–1188.

Burnet, F. M. (1951) *Physiol. Rev. 31*, 131–150.

Chadwick, C. M., & Garrod, D. R. (1983) *J. Cell Sci. 60*, 251–266.

Chappell, J., & Hahlbrock, K. (1984) *Nature (London) 311*, 76–78.

Chappell, J., Hahlbrock, K., & Boller, T. (1984) *Planta 161*, 475–480.

Clark, B. L., Oka, J. A., & Weigel, P. H. (1987) *J. Biol. Chem. 262*, 17384–17392.

Cline, K., Wade, M., & Albersheim, P. (1978) *Plant Physiol. 62*, 918–921.

Cramer, C. L., Ryder, T. B., Bell, J. N., & Lamb, C. J. (1985) *Science (Washington, D.C.) 227*, 1240–1243.

Darvill, A. G., & Albersheim, P. (1984) *Annu. Rev. Plant. Physiol. 35*, 234–275.

Davis, K. R., Darvill, A. G., & Albersheim, P. (1986) *Z. Naturforsch., C: Biosci. 41C*, 39–48.

Dennis, J. W., Laferte, S., Waghorne, C., Breitman, M. L., & Kerbel, R. S. (1987) *Science (Washington, D.C.) 236*, 582–586.

Dixon, R. (1986) *Biol. Rev. 61*, 239–291.

Dixon, R., Dey, P. M., & Lamb, C. J. (1983) *Adv. Enzymol. Relat. Areas Mol. Biol. 53*, 1–36.

Esquerre-Tugae, M. T., Mazsau, D., Pelissier, B., Roby, D., Rumeau, D., & Toppan, A. (1985) in *Molecular Strategies for Crop Protection* (Arnzen, C. J., & Ryan, C. A., Eds.) pp 375–397, Liss, New York.

Goldstein, J. L., Brown, M. S., Anderson, R. G. W., Russell, D. W., & Schneider, W. J. (1984) *Annu. Rev. Cell Biol. 1*, 1–39.

Graham, J. S., Hall, G., Pearce, G., & Ryan, C. A. (1986) *Planta 169*, 399–405.

Grand, C., Sarni, F., & Lamb, C. J. (1987) *Eur. J. Biochem. 169*, 73–77.

Hadwiger, L., & Beckman (1980) *Plant Physiol. 66*, 205–211.

Hadwiger, L., & Loschke, D. E. (1981) *Phytochemistry 71*, 756–762.

Hahn, M., & Albersheim, P. (1978) *Plant Physiol. 62*, 107–111.

Hedrick, S. A., Bell, J. N., Boller, T., & Lamb, C. J. (1988) *Plant Physiol. 86*, 182–186.

Jin, J. D., & West, C. A. (1984) *Plant Physiol. 74*, 989–992.

Kohle, H., Jeblick, W., Poten, F., Blaschek, W., & Kauss, H. (1985) *Plant Physiol. 77*, 544–551.

Kombrink, E., Schroeder, M., & Hahlbrock, K. (1988) *Proc. Natl. Acad. Sci. U.S.A. 85*, 782–786.

Kuc, J. (1981) in *Plant Disease Control* (Staples, R., Ed.) pp 259–272, Wiley, New York.

Kurosake, F., Tsurusawa, Y., & Nishi, A. (1987) *Plant Physiol. 85*, 601–604.

Lamb, C. J., Bell, J. N., Corbin, D. R., Lawton, M. A., & Mehdy, M. C. (1987) in *Molecular Strategies for Crop Protection* (Arntzen, C. H., & Ryan, C. A., Eds.) pp 49–58, Liss, New York.

Lawton, M. A., & Lamb, C. J. (1987) *Mol. Cell. Biol. 7*, 335–341.

Legrand, M., Kauffman, S., Geoffroy, P., & Fritig, B. (1987) *Proc. Natl. Acad. Sci. U.S.A. 84*, 6750–6754.

Mauch, F., Hadwiger, L. A., & Boller, T. (1984) *Plant. Physiol. 76*, 607–611.

Muller, K., & Garish, G. (1978) *Nature (London) 274*, 445–449.

Nothnagel, E. A., McNeil, M., Albersheim, P., & Dell, A. (1983) *Plant. Physiol. 71*, 916–926.

Ojalvo, I., Rokem, J. S., Navon, G., & Goldberg, I. (1987) *Plant Physiol. 85*, 716–719.

Rhodes, D. F. (1979) in *Herbivores, Their Interaction with Secondary Plant Metabolites* (Rosenthal, G. A., & Janzen, D. H., Eds.) pp 155–220, Academic, New York.

Robertsen, B. (1986) in *Biology and Molecular Biology of Plant–Pathogen Interactions* (Bailey, J., Ed.) pp 177–183, Springer-Verlag, Berlin.

Ryan, C. A. (1978) *Trends Biochem Sci. (Pers. Ed.) 5*, 148–150.

Ryan, C. A. (1987) *Annu. Rev. Cell Biol. 3*, 295–317.

Sairam, M., & Bhargavi, G. N. (1985) *Science (Washington, D.C.) 229*, 65–68.

Schachter, H. (1984) *Clin. Biochem. (Ottawa) 17*, 3–14.

Schmidt, W. E., & Ebel, J. (1987) *Proc. Natl. Acad. Sci. U.S.A. 84*, 4117–4121.

Sharp, J. K., Valent, B., & Albersheim, P. (1984) *J. Biol. Chem. 259*, 11312–11320.

Showalter, A. M., & Varner, J. (1987) in *Molecular Strategies for Crop Protection* (Arntzen, C. J., & Ryan, C. A., Eds.) pp 375–397, Liss, New York.

Showalter, A. M., Bell, J. N., Cramer, C. L., Bailey, J. A., Varner, J. E., & Lamb, C. J. (1987) *Proc. Natl. Acad. Sci. U.S.A. 82*, 6551–6555.

Somssich, I., Schmelzer, E., Bollmann, J., & Hahlbrock, K. (1986) *Proc. Natl. Acad. Sci. U.S.A. 83*, 2427–2430.

Staub, M. R., & Ebel, J. (1987) *Arch. Biochem. Biophys. 257*, 416–423.

Steineman, C., & Parish, R. W. (1978) *Nature (London) 286*, 621–623.

Walker-Simmons, M., & Ryan, C. A. (1984) *Plant Physiol. 76*, 787–799.

Weigel, P. H. (1987) in *Vertebrate Lectins* (Olden, K., & Parent, B., Eds.) pp 65–91, Van Nostrand Reinhold, New York.

Weinstock, K., & Ballou, C. E. (1986) *J. Biol. Chem. 261*, 16174–16179.

West, C. A. (1980) *Naturwissenschaften 68*, 447–457.

Yet, M.-G., Shao, M.-C., & Wold, F. (1987) *FASEB J. 2*, 22–31.

Yoshikawa, M., Keen, N. T., & Wang, M. C. (1983) *Plant Physiol. 73*, 497–506.

Ziska, S. E., & Henderson, E. J. (1988) *Proc. Natl. Acad. Sci. U.S.A. 85*, 817–821.

Zook, M. N., Rush, J. S., & Kuc, J. (1987) *Plant Physiol. 84*, 520–525.