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Interaction of Abscisic Acid and Indole-3-Acetic Acid-Producing Fungi with *Salix* Leaves

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Abstract. The molds *Botrytis cinerea, Cladosporium cladosporioides,* and the yeast *Aureobasidium pullulans,* isolated from the leaves of three short-rotation *Salix* clones, were found to produce indole-3-acetic acid (IAA). Abscisic acid (ABA) production was detected in *B. cinerea.* The contents of IAA and ABA in the leaves of the *Salix* clones and the amounts of fungal propagules in these leaves were also measured, in order to evaluate whether the amounts of plant growth regulators produced by the fungi would make a significant contribution to the hormonal quantities of the leaves. The content of ABA, and to a lesser degree that of IAA, showed a positive correlation with the frequency of infection by the hormone-producing organisms. The amounts of hormone-producing fungi on leaves that bore visible colonies were, however, not sufficiently high to support the claim that either the fungal production of ABA or IAA would significantly contribute to the hormonal contents of the leaves of the *Salix* clones. It is therefore suggested that the effect of fungal IAA production on plants is limited to the rhizosphere and that *B. cinerea,* which is a known pathogen, induces ABA production by the mother plant as a response to physiological stress.

Abscisic acid (ABA) is an important endogenous plant growth regulator and is involved in the response mechanisms of plants to changing environmental conditions. ABA has, apart from this, a clearly demonstrable inhibitory action when applied externally to growing organs. It is therefore of interest when considering the plant–microbe interaction that some phytopathogenic fungi are also able to produce this growth regulator (Marumo et al. 1982, Dörffling et al. 1984, Assante et al. 1977, Oritani and Yamashita 1985, Okamoto et al. 1987). The role of ABA in the host–pathogen relationship is, however, as of yet poorly understood. There are some reports in the literature showing that in plants infected by pathogenic fungi, the level of ABA is increased. The possibility has been raised that this increase is a response of the plants to stress, being the result of infection rather than the cause of it (Pegg 1976).

Indole-3-acetic acid (IAA) is the principal auxin in higher plants. The production of extracellular IAA has been recorded in several fungi and bacteria (Navarro et al. 1975, Woehler 1990). IAA is present in soil, synthetized by the autochtonous microflora (Stevenson 1964, McLaren and Peterson 1967, Strzelczyk and Karwowska 1969, Strzelczyk et al. 1973). IAA has been isolated or auxin activity found in culture filtrates of a high percentage of heterotrophic microflora present in an environment where they might influence plant growth (Waksman 1952, Gibson 1959, Puroshothaman et al. 1974). Libbert et al. (1966) also found that the IAA in the plant is actually contributed by the microorganisms, particularly bacteria present near the root system.

It has been claimed that microbial synthesis of plant growth regulators may be an important factor in soil fertility (Kampert and Strzelczyk 1975). A change of the bacteria population can indirectly influence plant growth. It would seem likely that growth regulators, such as ABA and IAA, produced by soil-borne microorganisms would play a role in this mechanism.

IAA production in fungi and bacteria, present in

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Abbreviations: ABA, abscisic acid; ABA-Me, abscisic acid methyl ester; GC-MS-SIM, gas chromatography-selected ion monitoring-mass spectrometry; IAA, indole-3-acetic acid; IAA-Me, indole-3-acetic acid methyl ester.

the rhizosphere of plants, is well known, and it seems probable that the rhizosphere of plants is supplied with external IAA, owing to microbial production. There has yet to surface, however, a theory that would explain the role of this production in plant–microbe interactions (Mayer 1989, Gardan et al. 1992). The aim of this study was to determine whether IAA or ABA production does take place in fungi isolated from the leaves of three willow clones, in order to determine whether the phyllosphere of plants is also supplied with externally produced IAA or ABA. Furthermore, we attempted to estimate whether this production constitutes a significant addition to the total hormonal amounts in the leaves.

Materials and Methods

Strains and Growth Conditions

The fungi were originally isolated from the leaves of three willow (*Salix*) clones, aimed to be used by the chemical industry and as fuel for energy production: *S. myrsinifolia* Salisb. clone V78, *S. dasyclados* clone P6011, and *S. viminalis* clone S-HK-81-2X. The fungi were identified as *Botrytis cinerea, Cladosporium cladosporioides,* and *Aureobasidium pullulans*. The leaves were collected from plants grown in the field at Kotkaniemi Research station of Kemira, Ltd. (Vihti, Finland). The pure cultures were rotated in 1-month intervals, and stored at +5°C, and then moved to room temperature 1 day prior to inoculation.

The growth medium contained the following ingredients: 10 g KNO_3 (Merck), 5 g KH_2PO_4 (Merck), 0.25 g $MgSO_4$ (Merck), 0.4 g $FeCl_3$ (Merck), 20 g glucosemonohydrate (Merck), and 5 g neopeptone (contains tryptophane) (Difco) per 1000 ml of distilled water. The fungi were cultivated for 14 days at room temperature (20–23°C), on a rotary shaker (160 rpm, Certomat) in two parallel 250 ml shake flasks, containing 100 ml of medium. During the growth, the fungi were subjected to daylight, with the same light period as outside, even though the production of ABA by *B. cinerea* is enhanced in the dark and even more so when it is subjected to blue light (Marumo et al. 1982). This was done in order to simulate light circumstances in the field.

Preparation of Samples

The mycelium of *B. cinerea* and *C. cladosporioides* was separated from the growth medium by filtration (Whatman No. 3 paper). The growth medium was retained, and the mycelium washed with distilled water and frozen (−60°C), before lyophilization 48 h (Christ Alpha 2–4). The cells of *A. pullulans* were separated from the growth medium by centrifugation (2000 *g*, 5°C, 30 min), the supernatant was retained and the cells were washed twice with 10 ml of distilled water. Thereafter the cells were transferred to Whatman No. 3 filter paper, were frozen and lyophilizated. The fungal samples were stored at −60°C and the growth media at +5°C before extraction and derivatization of plant hormone.

The leaf samples were collected from the three willow clones in September 1992. Seemingly intact leaves, as well as leaves that showed visible signs of infection by fungi, were collected for the analysis of plant hormones. The leaves that were collected for the analysis of ABA and IAA were fully developed and not senescent. The leaf samples were frozen to −60°C and lyophilized (Christ Alpha 2–4) before quantitation of the plant hormones. Parallel samples of lyophilized leaf material (1 g) were weighed for the analysis. The samples were treated similarly as the myceliar samples for the extraction and derivatization of plant hormone.

Extraction of Plant Hormone

All glassware was soaked in a 5% solution of Surfacil (Pierce Chemical Co.) in hexane and oven-dried for 1 h at 100°C. After cooling, the glassware was rinsed with methanol. All solvents were of HPLC-grade. All the sample preparations were carried out under dim light. Samples and extracts were stored at +5°C when not in use. If the samples had to be stored for longer periods, they were moved to −20°C. All samples were saturated with nitrogen between preparation steps to minimize oxidative decomposition.

The fungal and the leaf samples were ground with a mortar and pestle and the samples were extracted overnight with 70% (vol/vol) acetone, containing 20 mg/L^{-1} of the anti-oxidant butylated hydroxytoluene (BDH Limited) at 4°C, in the dark, under continuous stirring. The slurry was centrifugated (2 min, 1500 *g*, 8°C), and the supernatant was retained. The pellet was reextracted for 3 h, the slurry centrifugated and the supernatants were combined. The combined supernatants were then evaporated to the aqueous phase in a rotavapor, saturated with N_2, at 35°C. The aqueous solution, containing the myceliar or the leaf-derived ABA and IAA was after this treated similarly as the growth medium, containing the extracellular ABA and IAA, for the extraction of plant hormone. A similar amount of pure growth medium as was used for the cultivation of the fungi was measured for ABA and IAA content as well.

The pH of the solution was adjusted to 8.5 with a freshly prepared half saturated solution of $NaHCO_3$. The alkaline aqueous extract was then washed three times with chloroform and the organic phase, containing pigments, was discarded. The remaining aqueous solution was acidified (pH 2.5) with 1 N H_2SO_4 and extracted three times with ether. The ethereal extract was evaporated to about 4 ml under N_2 and dried over anhydrous Na_2SO_4. The extract was retained and the Na_2SO_4 washed once with ether. The ethereal extracts were combined and evaporated to about 50 µl under N_2, after which the samples were subjected to thin layer chromatography on aluminium sheets coated with kieselgel 60 F_{254} (Merck). The eluent contained toluene, ethyl acetate, and acetic acid (50:30:4). The plates were developed, in the dark, to 15 cm. The ABA and the IAA spots were identified by comparing to standards ((±)*cis, trans*-ABA, Sigma; IAA, Sigma). The R_f of ABA was 0.29 and the R_f of IAA 0.37. The ABA and IAA spots were scraped of from the plate and extracted overnight with diethyl ether, after which the ether was retained. The remaining kieselgel was reextracted twice with diethyl ether and the combined ethereal extracts were evaporated to about 50 µl under N_2, transferred to microvials and dried in a desiccator for 2 days at +5°C in the dark. The atmosphere in the desiccator was saturated with N_2.

GC-MS-SIM Analysis

The dried samples were methylated in capped vials with 60 µl of methyl-8 reagent (2 mEq · ml^{-1} in pyridine, Pierce) for 30 min at 90°C. The methylated samples were analyzed with a Hewlet-

Table 1. The amounts and the distribution of the fungi among the intact and visibly infected leaves of the three *Salix* clones.[a]

	S. viminalis S-HK-2X	*S. dasyclados* P6011		*S. myrsinifolia* V78	
		Intact	Infected	Intact	Infected
A. pullulans	10^4	—	—	8×10^4	3×10^4
C. cladosporioides	3×10^3	—	3.3×10^3	7×10^3	7×10^3
B. cinerea	—	—	50	—	100

[a] *S. viminalis* S-HK-2X did not bear any visibly infected leaves. The numbers refer to colonies per g fresh weight.

Packard 5890A gaschromatograph, equipped with a HP5971 mass-selective detector, a capillary inlet system and a model 7673A high-speed automatic liquid sampler with a 10 μl syringe. The conditions were as follows: J&W Scientific DB-17HT (30 m × 0.2 mm × 0.33 μm) column; carrier gas helium, column flow rate approximately 1.0 ml min^{-1}; septum purge flow-rate 1–2 ml min^{-1}; column inlet pressure 70 kPa; injector temperature 250°C; ion-source temperature 180°C; molecular separator temperature 135°C; electron energy 70 eV and trap current 300 μA. The column temperature was initially held at 80°C for 1 min, then programmed at 10°C min^{-1} to 250°C and held at 250°C for 20 min. Under these conditions, IAA-Me and ABA-Me had retention times of 16.80 and 18.60 min, respectively. When the equipment was operated in SIM mode, IAA-Me was analyzed by monitoring the molecular ion at m/z 189.15 together with the major fragment ion at m/z 130.05. ABA-Me was analyzed by monitoring the major fragment ion 190.05 m/z and the ion m/z 160.10.

The calibration curves used in the quantitation of the samples was drawn up using three external standards. The external standards were methylated with the same procedure as the samples. The error limits shown were measured by performing a double-sided student's *T* tests with 95% confidence interval on the deviation from the mean of four external standards. The external standards were subjected to the same treatments as the samples. The overall recovery of the hormones, as calculated from these four external standards were 83.7% (ABA) and 72.8% (IAA), and they were taken into account when calculating the results. The detection limits for both compounds were 100 pg μl^{-1}, and the linear range for ABA was 100 pg μl^{-1} – 100 ng μl^{-1} and for IAA 100 pg μl^{-1} – 20 ng μl^{-1}.

The sensitivity and reproducibility of the GC/MS apparatus used and the relatively small error limits of the extraction procedure (13% for ABA, 21% for IAA) enabled us to exclude the use of internal standards, such as [$^2H_4$]IAA, [$^{13}C_6$]IAA, or [$^2H_3$ABA] in the samples. Consequently, the use of error limits did not significantly affect the interpretation of our results.

Quantitative Determination of Fungi

One gram of leaf material was submerged in 99 ml of physiological salt solution and homogenized aseptically (Ultra-Turrex, IKA). Spread plates were prepared with 200 μl of diluted samples on PDA (Difco) plates, containing 0.05% ampicilline (Sigma). The formation and the amount of different colonies were determined visually. Three parallel samples were prepared. The cell number in the cultivation media of *A. pullulans* was enumerated microscopically, using the Hawksley glass counting chamber (A 70 Helber, Hawksley, Ltd., Lansing, England).

Results and Discussion

Fungal Strains

In this study, three fungi, isolated from *Salix* clones were assayed for indole-3-acetic and abscisic acid production. The fungi are all classified as *Deuteromycetes: B. cinerea* is a phytopathogenic fungus and causes serious gray mold disease of many kinds of cultivated plants; *C. cladosporioides* is a common habitat of both plant material and in soil and has been reported to produce leaf spots on certain plants (Holliday 1980, Mankarios 1978); *A. pullulans* (*Pullularia pullulans*) is a ubiquitos saprophyte that occurs commonly in the phyllosphere of plants and is believed to have a central role in initiating the natural breakdown and recycling of plant material. *A. pullulans* is a yeastlike organism, whereas *B. cinerea* and *C. cladosporioides* are molds. *C. cladosporioides* was present on the leaves of all three *Salix* clones; *A. pullulans* and *B. cinerea* could be isolated from the leaves of two of the three clones (Table 1).

Indole-3-Acetic Acid Production

Figure 1 exemplifies a typical GC-MS-SIM gaschromatogram of a microbial sample (*B. cinerea*). For comparison, the chromatogram of a hormonal standard, containing both ABA-Me and IAA-Me, is also shown. A large peak with the same retention time (16.80 min) as authentic IAA-Me was detected. When the samples were analyzed with SCAN-detection, mass-spectras that corresponded well with the IAA-Me standard were obtained for this peak (Fig. 2). Consequently, the three fungi all produced IAA in the growth conditions used. The growth medium of *A. pullulans* contained consider-

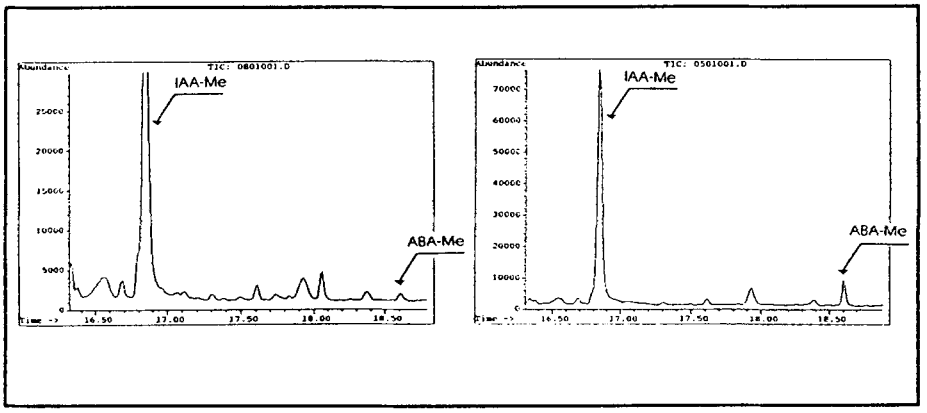

Fig. 1. The GC-MS-SIM gaschromatograms of a sample, extracted from *B. cinerea* (left), and the gaschromatogram of a hormonal standard (right).

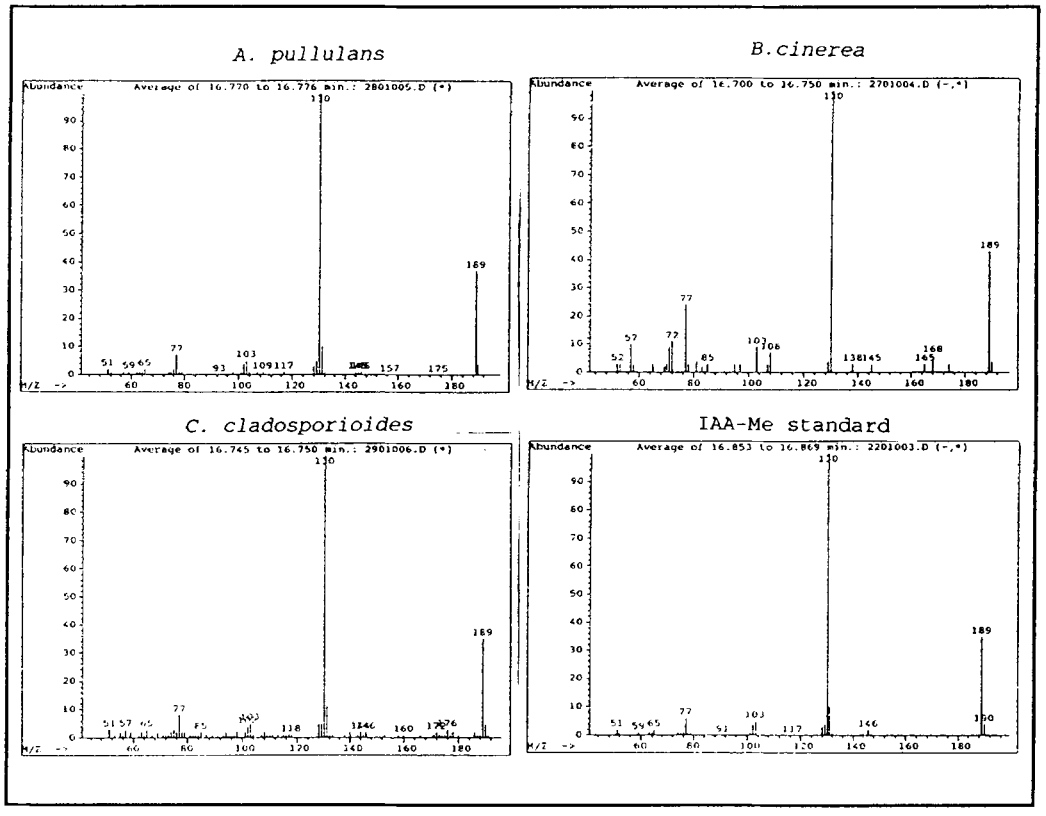

Fig. 2. The mass-spectra of an IAA-Me standard, and the spectra of IAA, extracted and derivatized from the growth medium of the three fungi.

ably more IAA than those of the two other fungi (44 ± 9 ng ml^{-1}), the media of *B. cinerea* containing least IAA (0.7 ± 0.1), even though the mycelium of this fungi contained more IAA than the mycelia of the two other fungi (Table 2).

Jump (1938) suggested that a phytohormone produced by *A. pullulans* may have been responsible for forking in red pine. Pentland (1965), who found this compound to stimulate the rhizomorph development of *Armillaria mellea*, described it as a volatile growth-promoting substance, the effect of which he could not mimic by the external addition of IAA. Buckley and Pugh (1971), on the other hand, reported the production of IAA by an *A. pullulans* strain isolated from sycamore trees, as indicated by the *Avena* coleoptile test and paper chromatography. Interestingly, they reported maximal IAA production (150 ng ml^{-1}) at 15°C rather than 5°

Table 2. The ABA and IAA concentrations in the growth media and the mycelia of the three fungi.

Fungi	ABA	IAA
B. cinerea		
Growth medium	1.60 ± 0.21 ng ml^{-1}	0.717 ± 0.15 ng ml^{-1}
Mycelium	196 ± 26 ng g^{-1}	128 ± 26 ng g^{-1}
C. cladosporioides		
Growth medium	Nil	6.11 ± 1.3 ng ml^{-1}
Mycelium	Nil	91.2 ± 19 ng g^{-1}
A. pullulans		
Growth medium	Nil	43.8 ± 9.1 ng ml^{-1}
Mycelium	Nil	84.0 ± 18 ng g^{-1}

or 25°C. IAA production by *C. cladosporioides* and *B. cinerea* has not been reported previously.

The content of free IAA in plants is usually between 1 and 100 ng g^{-1} fresh wt tissue (Schneider and Wightman 1974, Bandurski and Schulze 1977, Sandberg et al. 1987), which corresponds well with the present data (Fig. 3). There seemed to be a slight positive correlation between the frequency of infection by fungi and the concentration of IAA in the leaves (Fig. 3). The cell number in the cultivation media of the yeast *A. pullulans* was used to estimate whether IAA production by fungi, colonizing the leaves, could account for this correlation. At the termination of cultivation, the cell number in the growth media of *A. pullulans* was 1.94×10^8 cells ml^{-1} and the IAA concentration was 43.8 ng ml^{-1}. This gives an IAA production of 2.26×10^{-7} ng $cell^{-1}$. Given that the production of IAA by *A. pullulans* colonies, colonizing the leaves of the *Salix* clones was of the same magnitude, this would give a contribution of 0.00226–0.0181 ng g^{-1} fresh wt (Table 1) externally applied to the leaves. This amount would seem to be very low when compared to the magnitude of the IAA content in the leaves of the clones (200–500 ng g^{-1}; Fig. 3). A similar comparison cannot be made with the two molds, owing to their growth characteristics; however, when taken into account that the dry weight of all the fungi at termination of growth were similar, and since the two other fungi produced IAA in lesser amounts than *A. pullulans*, it would seem likely that neither of the two molds colonizing the leaves of the mother plant produce IAA in amounts that could have physiological significance.

Libbert and Silhengst (1970) proved in their experiments with epiphytic IAA-producing bacteria that IAA produced by microbes is taken up by plants. The present study suggests, however, that fungal-derived IAA, externally applied to the leaves of the host plant, is a neglible factor when considering the physiology of the leaves. The carrying capacity of leaf surfaces for microbes is low relative to other plant habitats, particularly roots (Fokkema and Schippers 1986). Consequently, the amounts of fungal propagules in the rhizosphere and hence the concentration of external IAA produced by these fungi is presumably higher than in the phylloplane, which would mean that if fungal production of IAA does have any role in the plant–microbe interaction, then these effects are directed to the rhizosphere. It remains to be seen, however, what the fungi have to gain from this relationship.

Abscisic Acid Production

ABA production in the given conditions was detected only in *B. cinerea* (Table 2). Previously, ABA production by a *B. cinerea* species isolated from diseased strawberries has been reported by Marumo et al. (1982). Their strain gave an ABA production of 140 μg ml^{-1} of culture media after cultivation of 7 days on PDA plates in the dark. For comparison, we measured the ABA production of our strain in similar conditions, on PDA plates, and found it to be of much lesser magnitude (5.3 ng ml^{-1}).

The content of ABA in willow (*S. viminalis* L.) have been previously measured by Alvim et al. (1976, 1978). In their study, the ABA contents varied between 10 and 450 ng g^{-1} fresh wt tissue, the content of ABA in the leaves being of the magnitude 50 ng g^{-1} in August. These values are as well within the same range as the results in the present study for the leaves that did not bear visible signs of infection (Fig. 3). The ABA content of infected leaves were, however, somewhat higher than the previously reported amounts. The increase in the ABA content of the infected leaves, as compared to non-infected leaves, varied in quantity with the willow clone studied. In *S. dasyclados* P6011, the increase was 7.5-fold, whereas in *S. myrsinofolia* Salisb.

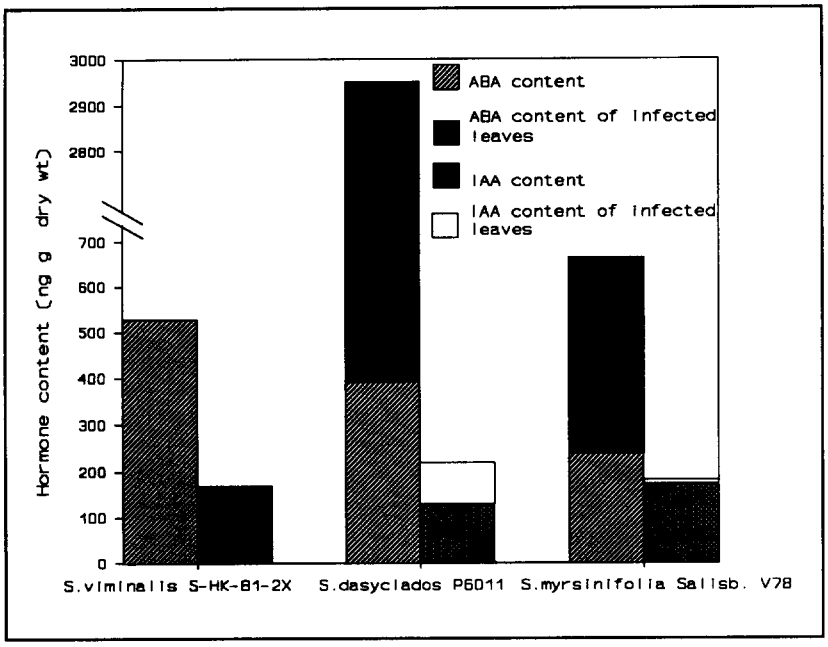

Fig. 3. The ABA and IAA content of the intact and the infected leaves of the *Salix* clones.

V78, the increase is less than threefold. The leaves were selected so that the differences cannot be attributed to senescence or dehydration, since the water content of the leaves of the different samples were similar. It could be argued then, that a correlation exists between infection by the ABA-producing fungus and the content of ABA in the leaves and, hence, that *B. cinerea* do affect the leaves of the host plant in a way that results in an increased level of ABA (Table 1, Fig. 3). Whether this increase is a result of fungal ABA production or a physical response of the leaves to stress induced by the fungus, or whether the infected leaves were damaged prior to the infection—and hence show an increased level of ABA—cannot be concluded from the data in Table 1 and Fig. 3. The cell number in the cultivation media of the yeast *A. pullulans* at the termination of growth could help in answering these questions. The ABA concentration in the growth medium of *B. cinerea* was less than the IAA concentration in the *A. pullulans* medium (Table 2), so if the arguments used above to estimate the significance of fungal IAA production when considering the physiology of the leaves is applied to the production of ABA by *B. cinerea*, then it would seem that this contribution, too, is negligent.

Botrytis cinerea was present only on the infected leaves of the clones *S. myrsinifolia* Salisb. clone V78, and *S. dasyclados* clone P6011. These two clones, as opposed to the *S. viminalis* clone S-HK-81-2X whose leaves did not contain *B. cinerea*, showed symptoms of senescense. ABA, when externally applied, causes the stomata of leaves to close within minutes of application (Mittelhauser and Van Stevenick 1969). In plants in which the stomata are closed for relatively lengthy periods or in which the degree of daily opening is curtailed, senescence occurs at a more rapid rate than in plants where the stomata are open for lengthier periods (Kuraishi and Ishikawa 1977, Thimann and Salter 1979a,b, Park and Thimann 1990). The precise way in which ABA is involved in this system has, however, not been considered. It would be tempting to state that the production of ABA by *B. cinerea* does, in combination with other factors, account for the occurrence of senescence in the leaves of the clones infected with this mold. The estimated amount of fungal-derived ABA externally applied to the leaves does not, however, support this argument. The results suggest rather, that *B. cinerea* affects the leaves in a way that enhances the ABA production of the leaves as a normal response to physiological stress and that results, eventually, in senescence.

Plant Hormone Production in Interactions Between Microbes

Besides the effect of fungal IAA and ABA on the host plant—and the effect, if any, of this production on the IAA producing organism itself—the effect of the hormonal production on other strains inhabiting the same leaf surface has to be taken into account. Inhibitory effect of IAA when applied to the growth medium has been reported with at least *Cylindro-*

carpon destructans (Kriesel 1987). IAA has, however, also been reported as having a stimulatory effect on the germination and growth of certain fungi (Michniewicz and Rozej 1984, Stopinska and Michniewicz 1988). ABA has been reported to depress the germination of *B. cinerea* (Blakeman 1975). A stimulatory effect of ABA on the germination of the following species has been reported: *B. cinerea* Pers and *Gloeosporium album* Osterw. (Borecka and Pieniazek 1968), *Cylindrocarpon destructans* (Zins. Scholt.) (Michniewicz et al. 1986), *Fusarum culmorum* (W.G.Sm.) Sacc. (Michniewicz et al. 1987), and *Ceratocystis fimbriata* Ell. et Halst. (Stopinska and Michniewicz 1988). These effects on the germination and growth of microbes are usually concentration-dependent. A stimulatory or inhibitory effect is usually present in a narrow concentration interval. The production of ABA and IAA by fungi or bacteria colonizing the leaf could thus have a selective effect on the microbial population.

References

Alvim R, Hewett EW, Saunders PF (1976) Seasonal variation in the hormone content of willow. I. Changes in the abscisic acid content and cytokinin activity in the xylem sap. Plant Physiol 57:474–476

Alvim R, Thomas S, Saunders PF (1978) Seasonal variation in the hormone content of willow. II. Effect of photoperiod on growth and abscisic acid content of trees under field conditions. Plant Physiol 62:779–780

Assante G, Merlini L, Nasini G (1977) (+)-ABA, a metabolite of the fungus *Cercospora rosicola*. Experientia 33:1556

Bandurski RS, Schulze A (1977) Concentration of indole-3-acetic acid and its derivates in plants. Plant Physiol 60:211–213

Blakeman JP (1975) Germination of *Botrytis cinerea* conidia in vitro in relation to nutrient conditions on leaf surfaces. Trans Br Mycol Soc 65:239–247

Borecka H, Pieniazek J (1968) Stimulatory effect of abscisic acid on spore germination of *Gloeosporium album* Osterw. and *Botrytis cinerea* Pers. Bull Ac Pol Biol 16:657–661

Buckley NG, Pugh GJF (1971) Auxin production by phylloplane fungi. Nature 231:332

Dörffling K, Petersen W, Sprecher E, Urbasch I, Hanssen HP (1984) Abscisic acid in phytopathogenic fungi of the genera *Botrytis, Ceratocystis, Fusarium* and *Rhizoctonia*. Z Naturforsh Teill C 39:683–684

Fokkema NJ, Schippers B (1986) In: Fokkema NJ, Heuvel J (eds) Microbiology of the phyllosphere. Cambridge University Press, Cambridge, pp 137–159

Gardan L, David C, Morel M, Glickmann E, Abu-Ghorrah M, Petit A, Dessaux Y (1992) Evidence for a correlation between auxin production and host plant species among strains of *Pseudomonas syringae* subsp. *savastanoi*. Appl Environm Microbiol 58:1780–1783

Gibson J (1959) Nutritional aspects of microbial ecology. In: Microbial ecology, 7th symposium of general microbiology. London, p 22

Holliday P (1989) A dictionary of plant pathology. Cambridge University Press, Cambridge, p 62

Jump JAA (1938) A study of forking in red pine. Phytopathology 28:798–811

Kampert M, Strzelczyk E (1975) Synthesis of auxins by fungi isolated from the roots of pine seedlings (*Pinus sylvestris* L.) and from soil. Acta Microbiologica Polonica Ser B 7:223–230

Kriesel K (1987) The role of growth regulators in the host-parasite relationship of *Cylindrocarpon destructans* (Zins. Scholt.). I. Auxins. Bull Ac Pol Biol 35:153–160

Kuraishi S, Ishikawa F (1977) Relationship between transpiration and amino accumulation in *Brassica* leaf disks treated with cytokinin and fusiococcin. Plant and Cell Physiol 18:1273–1279

Libbert E, Silhengst P (1970) Interactions between plants and epiphytic bacteria regarding their auxin metabolism. VIII. Transfer of ^{14}C-indoleacetic acid from epiphytic bacteria to corn coleoptiles. Physiol Plant 23:480–487

Libbert E, Wichner S, Schiewer U, Risch H, Kaiser W (1966) The influence of epiphytic bacteriae on auxin metabolism. Planta 68:327–334

Mankarios AT (1978) Tissue degrading enzymes associated with *Cladosporium* leaf spot on *Psidium Guajava*. Proc Egypt Acad Sci 31:167–172

Marumo S, Katayama M, Kokori E, Ozaki Y, Natsume M, Kondo S (1982) Microbial production of abscisic acid by *Botrytis cinerea*. Agric Biol Chem 46:1967–1968

Mayer AM (1989) Plant-fungal interactions: A plant physiologist's viewpoint. Phytochemistry 28:311–317

McLaren AD, Peterson GH (eds) (1967) Soil biochemistry, Vol I. Marcel Dekker, New York, 509 pp

Michniewicz M, Czerwinska E, Lamparska K (1986) Stimulatory effect of abscisic acid on the growth and development of *Cylindrocarpon destructans* (Zins.) Scholt. Bull Ac Pol Biol 34:54–59

Michniewicz M, Michalski L, Rozej B (1987) Control of growth and development of *Fusarium culmorum* (W.G.Sm) Sacc. by abscisic acid under unfavourable pH values of the media and temperature. Bull Ac Pol Biol 35:143–515

Michniewicz M, Rozej B (1984) Control of growth and development of isolates of *Fusarium culmorum* (W.G.Sm.) Sacc. of different pathogenicity to wheat seedlings by plant growth regulators, V. Auxins. Acta Physiol Plant 6:189–195

Mittelhauser CJ, Van Stevenick RMF (1969) Stomatal closure and inhibition of transpiration induced by (RS) abscisic acid. Nature 221:281–282

Navarro B, Jose M, Montoya E (1975) Review of plant hormone production by microorganisms. An Edafol Agrobiol 34: 1075–1096

Okamoto M, Hirai N, Koshimizu K (1987) The biosynthesis of abscisic acid in *Cercospora pini. densiflorae*. Abstr Bull Plant Growth Reg Soc Am 15(4)

Oritani T, Yamashita K (1985) Biosynthesis of (+)-abscisic acid in *Cercospora cruenta*. Agricol Biol Chem 49:245–249

Park J, Thimann KV (1990) Senescence and stomatal apperture as effected by antibiotics in darkness and light. Plant Physiology 92:696–702

Pegg GF (1976) Endogenous inhibitors in healthy and diseased plants. In: Heitefuss P, Williams H (eds) Physiological plant pathology. Springer-Verlag, Berlin, pp 607–616

Pentland GD (1965) Stimulation of rhizomorph development of *Armillaria mellea* by *Aureobasidium pullulans* in artificial culture. Can J Microbiology 11:345–350

Purushothaman D, Marimuthu T, Venkataramanan CV, Kesavan R (1974) Role of actinomycetes in the biosynthesis of indole acetic acid in soil. Current Science 43:413–414

Sandberg G, Crozier A, Ernstsen A, Sundberg B (1987) High performance liquid chromatography and the analysis of indole-3-acetic acid and some of its decarboxylated catabolites in Scots pine (*Pinus sylvestris* L). In: Linskens HF, Jackson JF (eds) High performance liquid chromatography in plant sciences. Modern methods of plant analysis, New series, Vol 5. Springer, Berlin, pp 72–91

Schneider EA, Wightman F (1974) Metabolism of auxin in higher plants. Ann Rev Plant Physiol 25:487–513

Stevenson IL (1964) Biochemistry of soil. In: Bear FE (ed) Chemistry of the soil. Von Nostrand Reinhold Co, New York, 242 pp

Stopinska J, Michniewicz M (1988) Control of growth and development of *Ceratocystis fimbriata* Ell et. Halst. by plant growth regulators I. Auxins Bull Ac Pol Biol 36:259–264

Strzelczyk E, Kampert M, Dahm H (1973) Production and decomposition of indoleacetic acid (IAA) by microorganisms isolated from the root zone of two crop plants. Acta Microbiologica Polonic Ser B 5:71–79

Strzelczyk E, Karwowska JM (1969) Microbial degradation of indoleacetic acid (IAA) in soil. Polish Journal of Soil Science 2:59–64

Thimann Z, Salter Z (1979a) Relation between senescence and stomatal closure: Senescence in light. Proc Nat Acad Sci USA 75:2295–2298

Thimann Z, Salter Z (1979b) Relation between senescence and stomatal closure: Senescence in darkness. Proc Nat Acad Sci USA 75:2770–2773

Waksman SA (1952) Soil microbiology. J Wiley, New York, 356 pp

Woehler I (1990) Auxin-indole-3-acetic acid: Formation, distribution, effects, biochemistry, and methods of detection in microorganisms, plants and soils. GSF-Ber, Munich, 143 pp