Folia Geobot. Phytotax. 31: 47-56, 1996

HORMONE SENSITIVITY AND PLANT ADAPTATIONS TO FLOODING

Laurentius A.C.J. Voesenek, Minke Banga, Jan G.H.M. Rijnders, Eric J.W. Visser & Cornelis W.P.M. Blom

Department of Ecology, University of Nijmegen, Toernooiveld, 6525 ED Nijmegen, The Netherlands; tel. +31243653382, fax +31243652409, E-mail RENSV@SCI.KUN.NL

Keywords: Auxin, Ethylene, Gibberellin, *Rumex*, Submergence

Abstract: Plant hormones play a key role as mediators between environmental signals and adaptive plant responses. Auxin, ethylene and gibberellins are involved in the initiation of adaptive plant responses such as the development of adventitious roots and stimulated shoot elongation upon flooded conditions. These adaptive plastic responses in plants are frequently linked to changes in the concentrations of the hormones involved, but only rarely to shifts in sensitivity. Examples from ecophysiological research performed with species from the genus *Rumex* demonstrate the importance of the hormone sensitivity concept in plant adaptations to flooding: (a) *Rumex* species can be grouped into three response categories according to the ethylene sensitivity of the youngest petioles: positive, negative and indifferent; (b) Sub-ambient oxygen concentrations sensitize petioles of wetland *Rumex* species to ethylene; (c) Enhanced ethylene levels sensitize petioles of wetland *Rumex* species to gibberellin; (d) Auxin is the primary plant hormone responsible for the initiation of adventitious roots in wetland *Rumex* species. However, a factor related to waterlogging, possibly ethylene, is required to sensitize the root-shoot junction to endogenous auxin.

INTRODUCTION

Floodplains, swamps, peat bogs and salt marshes are habitats that expose terrestrial plants to overwet conditions. Flooding in these habitats may vary in timing, frequency and duration and may submerge a plant partially (waterlogging) or completely (ARMSTRONG et al. 1994). Many plant species from these areas have, or develop, traits that enable survival and reproduction under these adverse conditions. Most plant responses upon waterlogging can be mimicked in plants grown in de-oxygenated hydroponic cultures (DREW 1990), which demonstrates that low levels, or a complete lack, of oxygen is the primary environmental signal that induces adaptive changes in the physiology and morphology of plants. Adaptations that avoid oxygen deprivation reduce diffusion resistances within the plant and between the plant and the atmosphere. Within the plant, resistance is relieved by the development of a gas-space continuum (aerenchyma) that facilitates diffusion or convection of oxygen-containing air to organs deficient in oxygen (VOESENEK & VAN DER VEEN 1994). Aerenchyma can develop in existing tissues or in a concerted action with the formation of new organs such as adventitious roots (JACKSON 1985, VISSER et al. 1995). The plant-atmosphere resistance in submerged plants, caused by the relatively slow diffusion rate of gases in water compared to air, is relieved by enhanced internode or petiole elongation in order to reach the better illuminated and aerated zones above the water suface (RIDGE 1987, MUSGRAVE et al. 1972, VOESENEK et al. 1993a).

Plant hormones, especially ethylene, auxin and gibberellins, play an important role as mediators between flood-induced signals and adaptive plant responses (VOESENEK & VAN DER VEEN 1994). These are often linked to changes in the concentrations of the hormones involved. An example is the development of lysigenous aerenchyma in the cortex of maize roots that is causally related to enhanced ethylene concentrations. Both physical entrapment of this gas by the surrounding water (DREW et al. 1979) and high production rates (BRAILSFORD et al. 1993), induced by sub-ambient levels of oxygen, explain the enhanced levels of ethylene in the cortex of maize roots. Enhanced shoot elongation has also been linked to ethylene entrapment and increased production rates (METRAUX & KENDE 1983, VOESENEK et al. 1993a). Adventitious root formation, too, is generally related to enhanced ethylene and/or auxin concentrations (WAMPLE & REID 1979, TANG & KOZLOWSKI 1984a, 1984b). In 1981, TREWAVAS started a debate to stress the importance of changes in hormone sensitivity instead of concentrations to explain certain developmental and plastic growth patterns. WEYERS et al. (1987) quantified hormone sensitivity in terms of "sensitivity parameters" that describe concentration-response curves in mathematical terms. R_{max} represents the maximum initial rate of response, K_d the dissociation constant for the hormone-receptor complex or the hormone concentration needed for half maximum response and p or the Hill coefficient specifies the steepness of the concentration-response curve (WEYERS et al. 1987). It is important to distinguish two aspects within the more general term "sensitivity"; (a) changes in the number and affinity of receptors and (b) changes in the chain of events subsequent to the hormone-receptor interaction, ending with the capacity to respond on the level of a plant's anatomy and morphology (FIRN 1986, RIDGE 1992). The latter type can be of importance when species or different developmental stages are compared. Shifts in K_d are assumed to be caused by changes in affinity between the hormone and the receptor; sensitivity changes indicated by alteration in R_{max} are more difficult to relate to particular mechanisms. These may either result from a change in the number or availability of receptors or from a change in the cascade of events between hormone-receptor interaction and response (WEYERS et al. 1987).

Few studies relate the development of adaptive responses to flooding to changes in hormone sensitivity (see RASKIN & KENDE 1984a, RIDGE 1992).

The aim of this paper is to illustrate the importance of the plant hormone sensitivity concept in plant adaptations to overwet environmental conditions. Examples will be presented from our ecophysiological research program which uses a range of *Rumex* species as a model system to study flooding resistance.

RUMEX: A MODEL GENUS IN ECOPHYSIOLOGICAL FLOODING RESEARCH

The field distribution of the cosmopolitan genus *Rumex* L. in river floodplains is characterized by a vertical zonation of species related to the variation among species in flooding resistance. Flood-tolerant species occur on low elevated mudflats with a high intensity of flood disturbance. Flooding-susceptible species are restricted to rarely flooded field sites such as dykes and river levees. Flooding of riparian habitats in Dutch river floodplains is unpredictable in depth, frequency and duration (BLOM et al. 1994). According to the stress resistance tactics defined by FITTER & HAY (1981), wetland *Rumex* species can avoid, ameliorate and/or tolerate the severe conditions associated with flooding (VAN DER SMAN

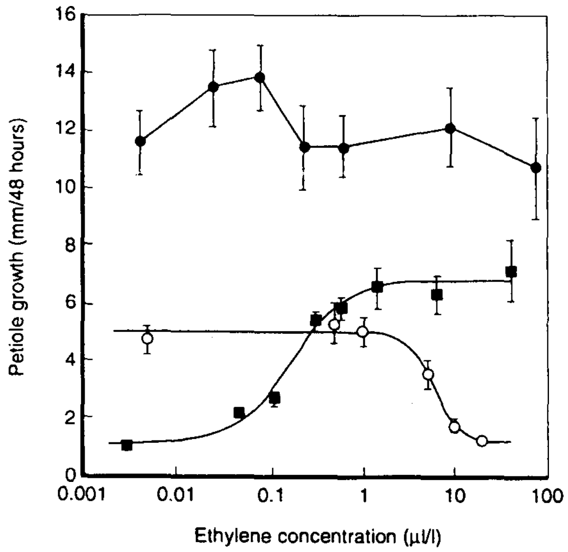

Fig. 1. Petiole and leaf (petiole + leaf blade; *R. acetosella*) growth (mean ± s.e.; n = 8-12) of three *Rumex* species exposed to a range of ethylene concentrations (● *R. acetosella*; ■ *R. palustris*; ○ *R. acetosa*). At the start of experimentation rosettes had an age of 26 days and were grown under the conditions described in VOESENEK & BLOM (1989). In *R. acetosa* and *R. palustris* a non-linear fitting procedure (PENG & WEYERS 1994) was used to construct a curve through the data. The response (R) at a given concentration ([H]) is described by the following equation:

$$R = R_{min} + (((R_{max} - R_{min}) * [H]^p)/([H]_{50}^p + [H]^p))$$

in which: R_{min}: minimum initial rate of response
R_{max}: maximum initial rate of response
$[H]_{50}$: hormone concentration giving a response of $(0.5 * (R_{max} - R_{min})) + R_{min}$
p: interaction or Hill coefficient (WEYERS & PATERSON 1992).

This revealed the following parameters:
R. acetosa: R_{max}: 1.25 ± 0.41; R_{min}: 5.09 ± 0.17; $[H]_{50}$: 5.80 ± 1.40; p: 3.10 ± 0.80.
R. palustris: R_{max}: 6.79 ± 0.31; R_{min}: 1.20 ± 0.46; $[H]_{50}$: 0.16 ± 0.04; p: 1.60 ± 0.47.

1992). *Rumex maritimus* L., a species from frequently flooded field sites, avoids the predictable winter floods by completing its life cycle between two subsequent winter floods (VAN DER SMAN 1992). Long-lived, flood tolerant seeds in a persistent seedbank bridge the gap between the growing seasons. This and other *Rumex* species from wetlands (*R. palustris* SM., *R. obtusifolius* L., *R. crispus* L.) can ameliorate the adverse effects of flooding by submergence-enhanced shoot elongation, waterlogging--induced formation of porous adventitious roots and the process of underwater photosynthesis (LAAN et al. 1989, VOESENEK & BLOM 1989, LAAN & BLOM 1990). Metabolic flood tolerance is observed in wetland *Rumex* species with the most perennial life cycle (*R. palustris* and *R. crispus*). These species produce chemical energy without oxygen by means of ethanolic fermentation (VOESENEK et al. 1993b).

Enhanced petiole elongation in response to submergence

Rumex species differ in the elongation capacity of petioles in response to submergence. Species from frequently flooded habitats respond by an enhanced growth rate of petioles under water, in contrast to dryland species that are unable to respond in this way (VOESENEK & BLOM 1989). Both groups physically accumulate ethylene to levels of 5-10 µl/l, approximately 100-fold higher than control plants (VOESENEK et al. 1993a). Therefore, differences in petiole growth between *Rumex* species upon submergence cannot simply be explained by variation in ethylene concentrations. The ethylene concentration-response curve (Fig. 1) indicates that three *Rumex* species, contrasting in petiole growth upon submergence, drastically differ in their sensitivity towards exogenously applied ethylene. In the dryland *R. acetosella* L. leaf growth seems to be independent of ethylene.

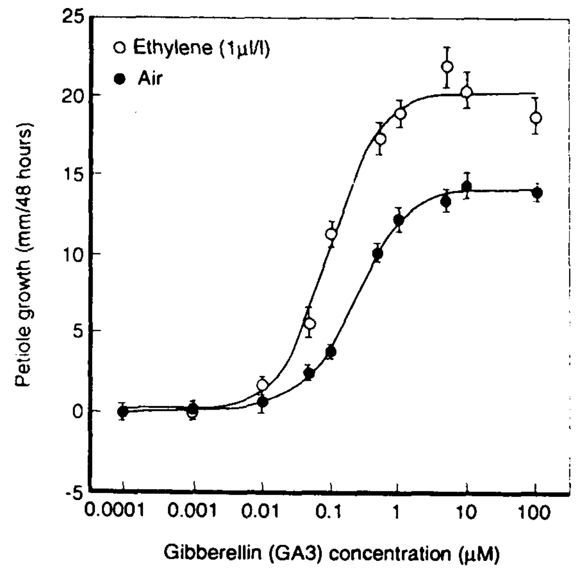

Fig. 2. Petiole growth (mean ± s.e.; n=10) of *R. palustris* upon exposure to a range of GA_3 concentrations. Rosettes had an age of 31 days, were grown under the conditions described in VOESENEK & BLOM (1989) and were pretreated with paclobutrazol (10 mL; 10^{-5} M root drench 4 days before experiment), an inhibitor of the GA biosynthesis. Curve fitting was achieved with the equation mentioned in Fig. 1. It revealed the following parameters:

Ethylene: R_{max}: 20.45 ± 0.66; R_{min}: 0.10 ± 0.75; $[H]_{50}$: 0.095 ± 0.016; p: 1.21 ± 0.24.
Air: R_{max}: 14.40 ± 0.20; R_{min}: 0.24 ± 0.19; $[H]_{50}$: 0.23 ± 0.02; p: 1.14 ± 0.07.

Therefore, no attempt was made to fit the Weyers equation on the data. *R. acetosa* L., another species from rarely flooded sites, reduced its growth rate upon ethylene exposure ($[C_2H_4]_{50}$: 5.80 ± 0.80 μl/l; Hill coefficient: 3.10 ± 1.40). *R. palustris* demonstrated a positive concentration-response curve upon ethylene exposure with a $[C_2H_4]_{50}$ of 0.16 ± 0.04 μl/l and a Hill coefficient of 1.60 ± 0.47. The response of *R. acetosa* is classical for most land plants (GOESCHEL & KAYS 1975), whereas the stimulation of growth as observed in *R. palustris* is characteristic for many aquatic and amphibious plants (RIDGE 1987, VOESENEK & VAN DER VEEN 1994). The response of *R. acetosella*, however, is very exceptional and can only be compared with the response of *Echinochloa oryzoides* (ARD.) FRITSCH coleoptiles (a weed from rice paddyfields) upon enriched ethylene environments (PEARCE & JACKSON 1991). According to these authors, the lack of response in this species might indicate a deficiency in the receptor protein responsible for ethylene binding and action. Ethylene-insensitive mutants of *Arabidopsis thaliana* (L.) HEYNH. are characterized by root and shoot hypocotyl elongation in the presence of high concentrations of ethylene (GUZMAN & ECKER 1990), by higher production levels of ethylene compared to wild-type plants and by a lack of suppression of ethylene biosynthesis upon pre-treatment with ethylene (KENDE 1993). One of them, *etr1*, is mutated in a gene probably acting very early in the ethylene signal transduction pathway as the receptor itself or as a protein interacting with the receptor (CHANG et al. 1993).

Based on this information, it is an intriguing question whether *Echinochloa oryzoides* and/or *Rumex acetosella* are in fact species with a mutation in the ethylene signal transduction chain. Arguments for this idea are the observations that the ACC-synthase activity and the basic ethylene production of *R. acetosella* is twice as high as that of *R. palustris* and that *R. acetosella* seems to lack a negative feedback in ethylene biosynthesis (data not shown). The differences in the sensitivity of *R. acetosa* and *R. palustris* to ethylene is probably not

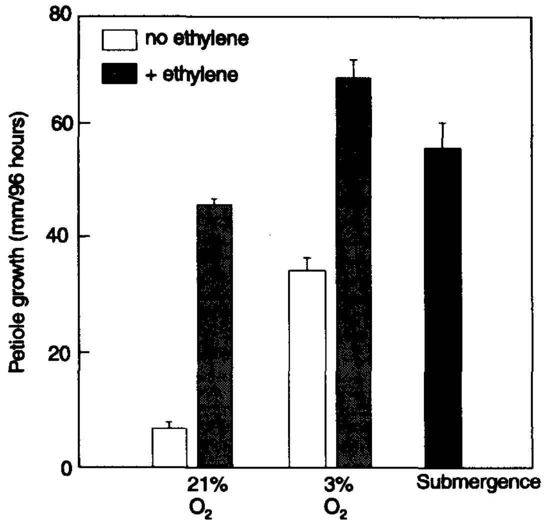

Fig. 3. Petiole growth (mean ± s.e.; *n*=18) of *R. palustris* exposed to four different gas mixtures and one submergence treatment. All gas mixtures were continuously flushed and contained 0.033% carbon dioxide. Ethylene was added at a growth saturating concentration of 5 µl/l. Rosettes were 27 days old and were grown under the conditions described by VOESENEK & BLOM (1989).

related to events early in the signal transduction pathway since both species respond to ethylene.

In *R. palustris* ethylene sensitizes the petiole tissue to a gibberellin (GA_3) (Fig. 2). Ethylene decreased the $[GA]_{50}$ from 0.23 ± 0.017 to 0.095 ± 0.016 µM and increased the R_{max} from 14.4 ± 0.2 to 20.5 ± 0.7 mm. Ethylene also induces an increase in the production of active endogenous GAs and their precursors (data not shown). To some extent this response is comparable to what is observed in partially flooded deep-water rice (SUGE 1985, HOFFMANN-BENNING & KENDE 1992). We hypothesize that in *R. palustris* ethylene stimulates the expression of genes interfering with the number of GA receptors, the affinity of GA to its receptor and the GA production. In this respect *R. acetosa* demonstrates a completely different ethylene – gibberellin interaction: ethylene de-sensitizes petiole tissue to GA_3 and has no influence on the production of active GAs (unpublished results).

However, GA is probably not the rate limiting step in the petiole growth of *R. acetosa* under water since exogenous application of GA in the presence of elevated ethylene levels (5 µl/l) does not stimulate petiole elongation in this species (data not shown). It is more likely that in *R. acetosa* ethylene exerts a direct effect on petiole cell walls that cannot be overruled by GA. In pea cells ethylene can induce a change in the orientation of microtubuli from predominantly transverse to longitudinal. This results in cell wall deposition of longitudinal cellulose microfibrils and thus in a cessation of cell elongation (SHIBAOKA 1994). SAUTER et al. (1993) demonstrated a close correlation between the rate of growth along the internode of deep-water rice and the orientation of cellulose microfibrils in the wall of epidermal cells. They also showed that GA in this species cannot cause reorientation of microtubuli and cellulose microfibrils. GA can only stimulate elongation in cells with a transverse orientation of cellulose microfibrils. This might be a model to explain the absence of petiole elongation in submerged, ethylene-rich *R. acetosa* plants exposed to high levels of GA.

Complete submergence induces a dramatic change in the endogenous concentrations of gases in plants. In general, ethylene and carbon dioxide levels increase, whereas the concentration of oxygen declines. Although ethylene in most cases seems to be the key substance in underwater growth of amphibious plants, fluctuations of other gases, especially oxygen, play an important role in submergence-induced shoot elongation (RASKIN & KENDE 1984b). The best mimic for submergence-induced petiole elongation in *R. palustris*, and thus the endogenous plant atmosphere under water, is a gas mixture containing 5 µl/l ethylene and

Table 1. Number of adventitious roots formed in *Rumex* species after 4 days of waterlogging. The age of the plants varied between 28 and 42 days (modified from BLOM et al. 1994). *n*=4.

Rumex species	Habitat	Number of roots
R. thyrsiflorus	permanently dry	8.0 ± 2.3
R. acetosa	permanently dry-moist	22.8 ± 5.8
R. obtusifolius	frequently flooded	16.3 ± 1.7
R. crispus	very frequently flooded	17.8 ± 2.8
R. conglomeratus	frequently flooded	42.5 ± 9.0
R. palustris	very frequently flooded	49.8 ± 1.7
R. hydrolapathum	permanently waterlogged	4.8 ± 0.9

3% oxygen (Fig. 3). Low levels of oxygen not only stimulate petiole growth in the presence of ethylene, but also in the absence of this gaseous growth regulator. Elevated levels of carbon dioxide (5%) have no influence on petiole elongation in *R. palustris* (data not shown).

The low oxygen-induced growth stimulation of *R. palustris* petioles in the absence of ethylene might have been caused by an increase of the ethylene production (VOESENEK & VAN DER VEEN 1994). This has earlier been described for internodes of deep-water rice (RASKIN & KENDE 1984b) and primary roots of maize seedlings (BRAILSFORD et al. 1993). However, it can not be ruled out that low levels of oxygen sensitize petiole tissue of *R. palustris* to ethylene. Evidence for shifts in ethylene sensitivity comes from the low oxygen-induced growth stimulation in the presence of ethylene (Fig. 3). The exogenously applied ethylene concentration (5 µl/l) saturates petiole growth in *R. palustris* (see Fig. 1). A further increase in growth under these conditions can only be achieved by an increase of tissue sensitivity towards the existing ethylene level or via an ethylene-independent growth stimulation. Preliminary evidence indicates that the effect of low oxygen on petiole growth acts via ethylene; application of inhibitors of ethylene biosynthesis (aminoethoxyvinyl glycine, AVG) and action (norbornadiene) demonstrated a significant reduction in growth under 3% oxygen levels compared to growth under sub-ambient oxygen concentrations without inhibitors. The effect of AVG was highly specific since it could be overruled when 1 mM 1-aminocyclopropane-1-carboxylic acid (ACC) was added (data not shown). In the presence of 10 µl/l ethylene, rice coleoptiles had higher growth rates in 0.5% oxygen than in an air environment. These data also indicate that the sensitivity to ethylene is higher at sub-ambient oxygen levels (HORTON 1991).

Adventitious root formation in response to waterlogging

Rumex species vary strongly in the number of adventitious roots formed upon waterlogging (Tab. 1). The development of these porous roots is almost absent in species from permanently dry (*R. thyrsiflorus* FINGERH.) and permanently waterlogged (*R. hydrolapathum* HUDS.) habitats. An intense development of adventitious roots is observed in *Rumex* species from habitats with a more or less fluctuating water table. According to the literature, formation of adventitious roots is related to an arrest of auxin transport in oxygen-deficient roots, resulting in the accumulation of auxin at the root-shoot junction (PHILLIPS 1964, WAMPLE & REID 1979). In *Rumex*, both natural and synthetic auxins induce the formation of adventitious roots under well aerated conditions on the upper parts of the tap-root. However, the endogenous concentration of auxin at the root-shoot junction does not explain the variation in root numbers between *Rumex* species. When *R. thyrsiflorus*, a species hardly able to form adventitious roots, is exposed to high levels of a synthetic auxin (1-naphthaleneacetic acid: 1-NAA) no

Table 2. Number of adventitious roots (mean ± s.e.; *n*=3-6) formed in two *Rumex* species (age 4 weeks) 4 days after treatment with a stagnant agar solution (hypoxia), or applying two different amounts of synthetic auxin to the shoot. No adventitious roots were formed under aerated conditions.

Species	Treatment		
	Hypoxia	1-NAA (10 nmol)	1-NAA (100 nmol)
R. palustris	26 ± 3	18 ± 2	38 ± 3
R. thyrsiflorus	8 ± 3	3 ± 1	6 ± 3

extra roots are formed: auxin concentration is not the rate limiting factor for the formation of these roots in this species (Tab. 2).

We think that the low sensitivity to form adventitious roots in this species is related to the root forming capacity on the level of the developmental stage, anatomy and/or morphology of the tissue at the shoot-root junction.

R. palustris, a species with a high capacity to form adventitious roots, forms them upon exposure to 1-NAA in a dose-dependent way (VISSER et al. 1995). Auxin seems to be the limiting factor for this process. This conclusion implies that under waterlogged conditions there are two routes to induce formation of adventitious roots: an increase in the auxin concentration as suggested by WAMPLE & REID (1979) and/or an increase in the sensitivity towards the existing auxin concentration by some factor related to waterlogging (see LIU & REID 1992). In *R. palustris*, adventitious root formation can be induced when hydroponically grown plants are placed in an unstirred hypoxic agar solution (hypoxia treatment; Tab. 3). When together with hypoxia the auxin transport inhibitor N-1-naphthylphthalamic acid (NPA) was applied, root initiation was reduced. The endogenous auxin concentration did not change under hypoxic conditions, but strongly decreased when NPA was added to the hypoxic agar solution (Tab. 3). We conclude that during waterlogging auxin induces adventitious root formation, although its concentration remains unchanged. Apparently, a factor closely associated with waterlogging and hypoxic roots sensitizes the root/shoot junction to auxin and thus triggers the plant to form adventitious roots. Many studies describe an important role for ethylene in the process of adventitious root formation (ZIMMERMAN & HITCHCOCK 1933, TANG & KOZLOWSKI 1984a, TANG & KOZLOWSKI 1984b, BLEECKER et al. 1987). Waterlogged plants of *R. palustris* produce more ethylene and probably contain higher endogenous levels of this gas than control plants (VOESENEK et al. 1990). LIU & REID (1992) demonstrated for cuttings of *Helianthus annuus* L. seedlings that auxin is the primary controller of adventitious root formation, but that ethylene was involved in increasing the tissue sensitivity towards auxin. In pea cuttings, an increase of the endogenous auxin concentration is also no prerequisite for the development of adventitious roots, although a certain minimum level is required (NORDSTROM & ELIASSON 1991). We hypothesize that ethylene might be the unknown factor involved in the sensitization of the root-shoot junction in *R. palustris* for auxin.

CONCLUDING REMARKS

The examples demonstrate that variation in hormone sensitivity plays an important but not an exclusive role in the adaptations of wetland *Rumex* species to waterlogged and submerged conditions. Submerged *Rumex* shoots accumulate ethylene, but differ in their capacity to elongate petioles upon flooding. *Rumex* species can be grouped into three response categories

Table 3. Number of adventitious roots (mean ± s.e.; n=6) formed and the endogenous concentration free IAA (mean ± s.e.; n=3) in the root/shoot junction of *R. palustris* after treatment. Number of roots was measured 7 days after treatment; IAA concentration after 12 hours of treatment. Amount of NPA was 150 nmol per shoot; plant age 4 weeks.

	Number of roots	Concentration IAA (nmol/g fw)
Control	0.2 ± 0.2	58.6 ± 6.4
Hypoxia	81.0 ± 5.9	51.9 ± 5.3
Hypoxia + NPA	52.2 ± 13.1	25.0 ± 1.3

according to their ethylene sensitivity of the leaf elongation response: positive, negative and indifferent.

Other gases and hormones also play a role in the petiole elongation response of wetland *Rumex* species. Oxygen, ethylene and gibberellin are involved in a cascade of sensitivity changes culminating in stimulated petiole growth. Due to the very slow diffusion rate of oxygen in water and the consumption in the shoot, endogenous levels of this gas decline in petioles and leaf blades upon submergence. Sub-ambient concentrations of oxygen sensitize the petiole tissue to ethylene. Enhanced ethylene levels, caused by physical entrapment, subsequently sensitize the petioles to gibberellin. This growth regulator in its turn stimulates cell elongation via still unknown mechanisms. Waterlogging induces in wetland *Rumex* species an abundant formation of adventitious roots at the root-shoot junction. Although auxin is the primary phytohormone responsible for the initiation of these roots in *Rumex*, its concentration in the root-forming zone did not change upon exposure to initiating conditions. Another factor, possibly ethylene, is required to sensitize the rooting zone in *Rumex* to endogenous auxin.

Changes in hormone sensitivity in relation to adaptations to flooding have also been described for the submergence-induced petiole elongation in *Ranunculus sceleratus* L. and the internode elongation in *Callitriche platycarpa* KÜTZ. and deep-water rice. In these three plant species, ethylene sensitizes the tissue to gibberellin (MUSGRAVE et al. 1972, MUSGRAVE & WALTERS 1973, RASKIN & KENDE 1984a). Another example of the importance of shifts in hormone sensitivity is the increase in sensitivity to ethylene of N or P starved roots of *Zea mays* L. Deprivation of N and P enhances the sensitivity of ethylene-responsive cells of the root cortex, leading to aerenchyma formation (HE et al. 1992).

Recently, DAVIES et al. (1994) published a hypothesis concerning the hormonal regulation of stomatal closure in dynamic environments. The long-term regulation of stomatal behaviour is mediated by a stable root signal (abscisic acid, ABA). The supply of ABA to the shoot is a function of the access of the plant to soil water. The response to evaporative demand, intercellular CO_2 level and temperature is determined by short-term variation in the sensitivity of the stomata to ABA. We suggest that such a model might also be applicable to the hormonal regulation of adaptations to flooding. The coarse regulation might take place on the level of hormone concentration. The production levels and concentrations of ethylene, indeed, correlate strongly with the flooding regime (waterlogging vs submergence). Fine-tuning and response to short-term environmental variation (streaming under water, light conditions, oxygen levels) will in this model be regulated by means of sensitivity variation.

Acknowledgements: The authors would like to thank Francis Huitink and Gerard Bögemann for technical assistance, Marret Smekens, Ingrid van Loon and Ite Nieboer for carrying out some of the experiments and Gerard Barendse and Wim Vriezen for their comments on draft versions of this paper.

REFERENCES

ARMSTRONG W., BRÄNDLE R. & JACKSON M.B. (1994): Mechanisms of flood tolerance in plants. *Acta Bot. Neerl.* 43: 307-358.

BLEECKER A.B., ROSE-JOHN S. & KENDE H. (1987): An evaluation of 2,5-Norbornadiene as a reversible inhibitor of ethylene action in deepwater rice. *Pl. Physiol.* 84: 395-398.

BLOM C.W.P.M., VOESENEK L.A.C.J., BANGA M., ENGELAAR W.M.H.G., RIJNDERS J.H.G.M., VAN DE STEEG H.M. & VISSER E.J.W. (1994): Physiological ecology of riverside species: Adaptive responses of plants to submergence. *Ann. Bot.* 74: 253-263.

BRAILSFORD R.W., VOESENEK L.A.C.J., BLOM C.W.P.M., SMITH A.R., HALL M.A. & JACKSON M.B. (1993): Enhanced ethylene production by primary roots of *Zea mays* L. in response to sub-ambient partial pressures of oxygen. *Pl. Cell Environm.* 16: 1071-1080.

CHANG C., KWOK S.F., BLEECKER A.B. & MEYEROWITZ E.M. (1993): *Arabidopsis* ethylene-response gene ETR1: Similarity of product to two-component regulators. *Science* 262: 539-544.

DAVIES W.J., TARDIEU F. & TREJO C.L. (1994): How do chemical signals work in plants that grow in drying soil? *Pl. Physiol.* 104: 309-314.

DREW M.C. (1990): Sensing soil oxygen. *Pl. Cell Environm.* 13: 681-693.

DREW M.C., JACKSON M.B. & GIFFARD S.C. (1979): Ethylene-promoted adventitious rooting and development of cortical air spaces (aerenchyma) in roots may be adaptive responses to flooding in *Zea mays* L. *Planta* 147: 83-88.

FIRN R.D. (1986): Growth substance sensitivity: The need for clearer ideas, precise terms and purposeful experiments. *Physiol. Pl.* 67: 267-272.

FITTER A.H.Q. & HAY R.K.M. (1981): *Environmental physiology of plants*. Academic Press, London.

GOESCHEL J.D. & KAYS S. (1975): Concentration dependencies of some effects of ethylene on etiolated pea, peanut, bean and cotton seedlings. *Pl. Physiol.* 55: 670-677.

GUZMAN P. & ECKER J.R. (1990): Exploiting the triple response of *Arabidopsis* to identify ethylene-related mutants. *Pl. Cell* 2: 513-523.

HE C.J., MORGAN P.W. & DREW M.C. (1992): Enhanced sensitivity to ethylene in nitrogen- or phosphate-starved roots of *Zea mays* L. during aerenchyma formation. *Pl. Physiol.* 98: 137-142.

HOFFMANN-BENNING S. & KENDE H. (1992): On the role of abscisic acid and gibberellin in the regulation of growth in rice. *Pl. Physiol.* 99: 1156-1161.

HORTON R.F. (1991): The effect of ethylene and other regulators on coleoptile growth of rice under anoxia. *Pl. Sci.* 79: 57-62.

JACKSON M.B. (1985): Ethylene and responses of plants to soil waterlogging and submergence. *Annual Rev. Pl. Physiol.* 36: 145-174.

KENDE H. (1993): Ethylene biosynthesis. *Annual Rev. Pl. Physiol. Pl. Molec. Biol.* 44: 283-307.

LAAN P., BERREVOETS M.J., LYTHE S., ARMSTRONG W. & BLOM C.W.P.M. (1989): Root morphology and aerenchyma formation as indicators of the flood tolerance of *Rumex* species. *J. Ecol.* 77: 693-703.

LAAN P. & BLOM C.W.P.M. (1990): Growth and survival responses of *Rumex* species to flooded and submerged conditions: the importance of shoot elongation, underwater photosynthesis and reserve carbohydrates. *J. Exp. Bot.* 41: 775-783.

LIU J.H. & REID D.M. (1992): Auxin and ethylene-stimulated adventitious rooting in relation to tissue sensitivity to auxin and ethylene production in sunflower hypocotyls. *J. Exp. Bot.* 43: 1191-1198.

METRAUX J.P. & KENDE H. (1983): The role of ethylene in the growth response of submerged deep water rice. *Pl. Physiol.* 72: 441-446.

MUSGRAVE A., JACKSON M.B. & LING E. (1972): *Callitriche* stem elongation is controlled by ethylene and gibberellin. *Nature* 238: 93-96.

MUSGRAVE A. & WALTERS J. (1973): Ethylene-stimulated growth and auxin transport in *Ranunculus sceleratus* petioles. *New Phytol.* 72: 783-789.

NORDSTROM A.C. & ELIASSON L. (1991): Levels of endogenous indole-3-acetic and indole-3-acetylaspartic acid during adventitious root formation in pea cuttings. *Physiol. Pl.* 82: 599-605.

PEARCE D.M.E. & JACKSON M.B. (1991): Comparison of growth responses of barnyard grass (*Echinochloa oryzoides*) and rice (*Oryza sativa*) to submergence, ethylene, carbon dioxide and oxygen shortage. *Ann. Bot.* 68: 201-209.

PENG Z.Y. & WEYERS J.D.B. (1994): Stomatal sensitivity to abscisic acid following water deficit stress. *J. Exp. Bot.* 45: 835-845.

PHILLIPS I.D.J. (1964): Root-shoot hormone relations. II. Changes in endogenous auxin concentrations produced by flooding of the root system in *Helianthus annuus*. *Ann. Bot.* 28: 37-45.

RASKIN I. & KENDE H. (1984a): Role of gibberellin in the growth response of submerged deep water rice. *Pl. Physiol.* 76: 947-950.

RASKIN I. & KENDE H. (1984b): Regulation of growth in stem sections of deepwater rice. *Planta* 160: 66-72.

RIDGE I. (1987): Ethylene and growth control in amphibious plants. In: CRAWFORD R.M.M. (ed.), *Plant life in aquatic and amphibious habitats*, Blackwell Scientific Publications, Oxford, pp. 53-76.

RIDGE I. (1992): Sensitivity in a wider context: Ethylene and petiole growth in *Nymphoides peltata*. In: KARSSEN C.M., LOON L.C. & VREUGDENHIL D. (eds.), *Progress in plant growth regulation*, Kluwer Academic Publishers, Dordrecht, pp. 254-263.

SAUTER M., SEAGULL R.W. & KENDE H. (1993): Internodal elongation and orientation of cellulose microfibrils and microtubules in deep-water rice. *Planta* 190: 354-362.

SHIBAOKA H. (1994): Plant hormone-induced changes in the orientation of cortical microtubules: Alterations in the cross-linking between microtubules and the plasma membrane. *Annual Rev. Pl. Physiol. Pl. Molec. Biol.* 45: 527-544.

SUGE H. (1985): Ethylene and gibberellin: Regulation of internodal elongation and nodal root development in floating rice. *Pl. Cell Physiol.* 26: 607-614.

TANG Z.C. & KOZLOWSKI T.T. (1984a): Ethylene productions and morphological adaptation of woody plants to flooding. *Canad. J. Bot.* 62: 1659-1664.

TANG Z.C. & KOZLOWSKI T.T. (1984b): Water relations, ethylene production and morphological adaptation of *Fraxinus pennsylvanica* seedlings to flooding. *Pl. & Soil* 77: 183-192.

TREWAVAS A.J. (1981): How do plant growth substances work? *Pl. Cell Environm.* 4: 203-228.

VAN DER SMAN A.J.M. (1992): *Flooding resistance and life histories of short-lived floodplain herbs*. PhD. Thesis, University of Nijmegen.

VISSER E.J.W., HEIJINK C.J., VAN HOUT K.J.G.M., VOESENEK L.A.C.J., BARENDSE G.W.M. & BLOM C.W.P.M. (1995): Regulatory role of auxin in adventitious root formation in two species of *Rumex*, differing in their sensitivity to waterlogging. *Physiol. Pl.* 93: 116-122.

VOESENEK L.A.C.J., BANGA M., THIER R.H., MUDDE C.M., HARREN F.J.M., BARENDSE G.W.M. & BLOM C.W.P.M. (1993a): Submergence-induced ethylene synthesis, entrapment, and growth in two plant species with contrasting flooding resistances. *Pl. Physiol.* 103: 783-791.

VOESENEK L.A.C.J. & BLOM C.W.P.M. (1989): Growth responses of *Rumex* species in relation to submergence and ethylene. *Pl. Cell Environm.* 12: 433-439.

VOESENEK L.A.C.J., HARREN F.J.M., BÖGEMANN G.M. & BLOM C.W.P.M. (1990): Ethylene production and petiole growth in *Rumex* plants induced by soil waterlogging: The application of a continuous flow system and a laser driven intracavity photoacoustic detection system. *Pl. Physiol.* 94: 1071-1077.

VOESENEK L.A.C.J. & VAN DER VEEN R. (1994): The role of phytohormones in plant stress: too much or too little water. *Acta Bot. Neerl.* 43: 91-127.

VOESENEK L.A.C.J., VAN OORSCHOT F.J.M.M., SMITS A.J.M. & BLOM C.W.P.M. (1993b): The role of flooding resistance in the establishment of *Rumex* seedlings in river flood plains. *Funct. Ecol.* 7: 105-114.

WAMPLE R.L. & REID D.M. (1979): The role of endogenous auxins and ethylene in the formation of adventitious roots and hypocotyl hypertrophy in flooded sunflower plants (*Helianthus annuus*). *Physiol. Pl.* 45: 219-226.

WEYERS J.D.B. & PATERSON N.W. (1992): Quantitative assessment of hormone sensitivity changes with reference to stomatal responses to abscisic acid. In: KARSSEN C.M., VAN LOON L.C. & VREUGDENHILL D. (eds.), *Progress in plant growth regulation*, Kluwer Academic Publisher, Dordrecht, pp. 226-236.

WEYERS J.D.B., PATERSON N.W. & BROOK R.A. (1987): Towards a quantitative definition of plant hormone sensitivity. *Pl. Cell Environm.* 10: 1-10.

ZIMMERMAN P.W. & HITCHCOCK A.E. (1933): Initiation and stimulation of adventitious roots caused by unsaturated hydrocarbon gases. *Contr. Boyce Thompson Inst. Pl. Res.* 5: 351-359.