

## MEETINGS OF THE ROYAL BOTANICAL SOCIETY OF THE NETHERLANDS

### MEETING OF THE SECTION FOR VEGETATION RESEARCH ON SEPTEMBER 30, 1981

H. van GILS (*Internationaal Instituut voor Luchtkartering en Aardkunde (ITC), Enschede*)  
Utilisation and management of woods in a global perspective

The following categories of land utilisation may be found in woods:

1. *Forestry*
  - 1.a timber
  - 1.b wood; underwood, firewood
  - 1.c pulp-wood
  - 1.d bark (cork, maple-sugar, turpentine, oak-bark for tan, etc.)
2. *Rangeland*
  - 2.a hunting
  - 2.b pastoralism
  - 2.c ranching
3. *Conservation*
  - 3.a erosion control; wind, water, snow erosion
  - 3.b watershed protection
4. *Culture*
  - 4.a sacred wood
  - 4.b nature reserve
  - 4.c out-door recreation
  - 4.d employment
  - 4.e decoration of estates

Many woodlands have a multiple land utilisation.

On a world-wide scale woodlands are a main component in regulating the atmospheric CO<sub>2</sub> and H<sub>2</sub>O concentrations; this aspect of woods is not discussed here.

It is noticed that this meeting deals with woods currently utilised mainly or partly for the cultural purpose of nature reserve (4.b). The present-day woods have been shaped to a large extent by their land use history. The land use history of the Netherlands woods include many of the woodland utilisation types mentioned above. Many of these historical woodland utilisation types can be studied elsewhere in the world of today. Especially the wood (1.b) and the rangeland 2.a-c) utilisation types of woodland have been driving forces for the form and composition of woods in The Netherlands.

Two theses are presented for discussion:

- (i) Current management of Netherlands woods in nature reserves tries to conserve specific successional wood stages.

Land use history, especially the former rangeland (2.a, 2.b, 2.c) and wood utilisation (1.b) of woodland, are usually not assessed in designing a wood management plan.

- (ii) Current management of Netherlands woods in nature reserves should try to create possibilities for natural processes.

Grazing of woods by both 'wild' and domestic animals and wood utilisation should be included more into wood management than is done currently.

E. C. J. OTT (*Vakgroep Vegetatiekunde, Plantenecologie en Onkruidkunde, Transitorium, De Dreijen 11, 6703 BC Wageningen*)

Coppice Management and Plant Communities

The species composition of the ground vegetation in coppice woods is known to be greatly influenced by the various systems of coppice management. Both the simple method with clearfellings at the end of each rotation and the composite coppice system with shading trees have significant effects on the microclimate under the canopy and even on the physical and chemical properties of the soil or the water balance. The early silvicultural system included preparation of the soil in order to create a suitable habitat for the species of trees that it was desired to grow.

The most important species under coppice management in the 18th. and 19th centuries was pedunculate oak (*Quercus robur*). The required conditions on dry, sandy soils were frequently achieved by digging to a substantial depth, whereas low-lying and waterlogged soils had to be drained. An example of the latter may be seen in the sequence where wet, non-loamy podzolic cover sands, poor in nutrients, change gradually into peaty soils in the more eutrophic conditions of lowland stream valleys.

The natural sequence in woodland plant communities probably consisted of birch-oakwood (*Betulo-Quercetum*) on relatively dry sites, with *Frangula alnus* and *Sorbus aucuparia*, and a ground vegetation of *Molinia caerulea* on the fairly wet sites, followed by a mixed alder-birchwood. On the peaty soils an alderwood (*Carici elongatae-Alnetum*) is normally found.

Increasing the area of the habitat for oak necessitated draining the soil by a system of ridges and furrows or ditches. The ridges were raised above the original soil level with the material from the ditches that was rich in organic substances. In contrast to the uncultivated site this created many microhabitats between the top of the ridges and the bottom of the furrows together with an increase in soil fertility.

In these circumstances former oak coppice, now neglected and mixed with other tree species, has lost its *Betulo-Quercetum* identity and now belongs to the *Holco-Quercetum* with the characteristic species *Populus tremula*, *Holcus mollis*, *Rubus fruticosus* agg. and *Dryopteris dilatata*. Under wet and somewhat eutrophic soil conditions, in which the nutrients are derived from periodic high groundwater levels in the ditches, the *Holco-Quercetum alnetosum* occurs, with *Alnus glutinosa*, *Rubus idaeus*, *Lysimachia vulgaris* and *Holcus lanatus*. Under poorer conditions can be found the *Holco-Quercetum betuletosum* with *Betula pubescens*, *Frangula alnus*, *Molinia caerulea* and *Dryopteris carthusiana*.

The *Carici remotae-Alnetum* is to be seen in those areas bordering on the ditches where water seepage occurs.

The communities mentioned above, each based on a preparation of 50 vegetation relevés, are tentatively presented as associations with sub-associations to compare them with the *Betulo-Quercetum*, the last mentioned, however, not reaching a more advanced syntaxonomic level because of the lack of characteristic species.

The average floristic composition of the communities described was derived from a variety of stands all with their own specific composition resulting from the variations in coppice management mentioned in the first paragraph of this summary.

#### H. PIEK (*Natuurmonumenten, Noordereinde 60, 1243 JJ 's-Graveland*)

##### Forest succession and nature techniques

The useful values of Dutch woodlands for men and other living organisms are based on using the protective capacity of forest ecosystems.

The functional relations between living organisms and woodlands are according to C. G. van Leeuwen to be divided in four basic functions:

- supply functions
- discharge functions
- resistance functions
- retention functions

The supply functions of forests are for example production of timber, firewood, hibernics, but also fresh air and information. The discharge functions are for example to dump waste, dungresidues or even to build houses into a woodland. The resistance functions of woodlands have to do with 'to keep off' such like refuse, wind, dust, noise, grazing animals (hedgerows!). The retention functions have to do with the concept of 'to keep in'.

Examples: to keep in rainwater and genetical information.

When men or other organisms use forests there always is some wastage. To keep the useful value up to the mark measures have to be taken, e.g. for levelling up the ecologically useful value for threatened and/or rare wild plant or animal species. This is called nature technical forest-management. This forest-management is a part of all the correcting techniques to prevent the harmful effects of the agro-techniques and urban-techniques. There are two types of nature technical measures: 1. Maintenance measures, and 2. Improvement measures.

The maintenance measures can be divided in outward and inward measures. Outward or defensive measures have to do with regulation of the maximum of admissible human influence. These measures are taken with systems with resistance and retention functions. The inward or restoration measures are taken with systems with supply and discharge functions and regulate the human influence which is minimally needed. The improvement measures use the same systems with the same functions as the maintenance measures.

The nature technical forest-management gives special attention to the threatened species.

In The Netherlands there are about 760 higher plant species threatened. About 93 of them or 12% occur in the three most common forest types (*Quercion robur-petraea*, *Alno-Padion* and *Carpinion*).

The nature technical forest-management is also interested in forest succession. The different succession stages in forest have their own characteristic plant species. Changes in the woodland vegetation include progressive and regressive successions. The climax of the succession is a forest with several sylvigenetical series. A cyclic succession is probably found in most zonal climax forests in Western Europe. A hypothetical example of a cyclic succession on an acid, poor sandy podzol soil (Veluwe) was given.

H. KOOP (*Landbouwhogeschool, Wageningen en Rijksinstituut voor Natuurbeheer, postbus 46, 3956 ZR Leersum*)

Succession mosaic, spatial differentiation and spontaneous development of forests

The Neuenburger and Hasbrucher Urwald (Niedersachsen, Germany) and the National Park of Bialowieza (Eastern Poland) are not virgin forests any more. But they are three of the few places where forest vegetation has not been influenced by human activities for almost 100 years. Therefore, they are the most spontaneous forests of the Northern European Lowland. In three forest types, the Oak-beech forest (*Fago-Quercetum*), the Wood millet-beech forest (*Milio-Fagetum*) and the Oak-hornbeam forest (*Stellario-Carpinetum*) vertical and horizontal structure-analyses have been made by mapping crown projections and designing side-views of 10 metre wide strips of forest. From *Fago-Quercetum* to *Stellario-Carpinetum* the vertical structure becomes more complex and at the same time the regeneration units of the forest mosaic become smaller and the part of the forest area that is in regeneration decreases.

In the *Stellario-Carpinetum* individual trees turn over or crack down often and regularly, but bigger gaps in the canopy where the microclimate is disturbed, occur only very rarely. In the *Fago-Quercetum* on the other hand there is an irregular low frequency of occurrence of bigger gaps in the canopy. The microclimate in the *Stellario-Carpinetum* is, therefore, more constant without irregularities, which causes a vegetation mosaic that is more small-scaled and richer in plant species than in the *Fago-Quercetum*.

The size of the gaps determines the kind of tree regeneration. Different stages can be distinguished: a herb stage, which can maintain its position for several decades, consisting of *Pteridium*, *Rubus* or high herbs; furthermore a preforest-stage (*Betula*, *Populus*, *Sorbus*), a main forest type (*Fagus*, *Carpinus*, *Tilia* and *Picea*) and a terminal stage (*Ilex* and *Taxus*).

Stand structure influences the number of species and the species composition of the total vegetation. At dry sites under gaps in the canopy species of a more moisty site grow. By competition of heliophilous species the "real" forest species are superseded, especially on the moisty and richer site types.

Besides the differentiating effect of the stand structure, there are in a spontaneous forest typical small-scaled site types as a result of the uprooting of trees and lying wood. The rotting woods

and the more or less flattened mound-and-pit-microrelief are suitable places for special species that are rare for the surrounding site type. As a result of these gradients at different scale the spontaneous forests are difficult to range in a syntaxonomy that is worked out in man-made landscapes. The same problem is met by using the French-Swiss School in the natural landscapes of the Tropics.

The climax is not only a continuing, selfregulating main forest stage, although this stage gives usually the name to the potential natural vegetation. Besides the main forest type, herb stages without trees, preforest stages and terminal stages participate in a climax mosaic.

KOOP, H. (1981): Vegetatiestructuur en dynamiek van natuurlijke bossen: Het neuenburger en hasbrucher Urwald. PUDOC, Wageningen.

C. J. M. SLOET VAN OLDRIJTENBORGH (*Vakgroep Natuurbeheer, Landbouwhogeschool, Ritzema Bosweg 32a, 6703 AZ Wageningen*)

Composition and management of the shrubvegetation along the Overijsselse Vecht between Ommen and Hardenberg.

S. VAN DER WERF (*Rijksinstituut voor Natuurbeheer, Broekhuizerweg 2, 3956 NS Leersum*)

Relations between actual and potential natural forestvegetations

## MEETING OF THE SECTION FOR PHYTOPATHOLOGY ON OCTOBER 29, 1981

J. C. M. BEIJERSBERGEN and C. Th. C. VAN DER HULST (*Laboratorium voor Bloembollenonderzoek, Vennestraat 22, 2160 AB Lisse*)

Can two serotypes of tulip breaking virus be present in tulip stocks?

From leaves of two different cultivars of tulips, cv. Texas Flame and cv. Jack Laan, isolates of tulip breaking virus (TBV) were prepared. Antisera against both isolates were produced via rabbits: RD-antisera against the TBV isolate from cv. Jack Laan and RE-antisera against the TBV isolate from Texas Flame.

In ELISA the RD-antisera reacted relatively better with TBV containing bulb extracts of cv. Jack Laan and the RE-antisera better with comparable extracts of cv. Texas Flame. TBV present in naturally infected lily bulbs in ELISA reacted much better with RD-antisera than with RE-antisera.

The best explanation for this phenomenon seems to be that two serotypes of TBV can be present in naturally infected tulips. Both types can be found in stocks of tulips although the ratio in which the types occur varies greatly per individual plant of the same stock.

A. R. VAN SCHADEWIJK and J. EGGINK (*Bloembollenkeuringsdienst, p/a Laboratorium voor Bloembollenonderzoek, Vennestraat 22, 2160 AB Lisse*)

A new virus in tulips as one of the possible explanations for serological distinction between antisera against tulip breaking virus (TBV).

Using the enzyme-linked immunosorbent assay (ELISA) to detect TBV in tulips and lilies, two types of TBV-antisera available showed different serological characteristics. The existence of different serotypes of TBV and the possible involvement of a second virus was investigated. By means of immunosorbent electronmicroscopy only, a new virus with a rhabdovirus-like appearance could be detected in one of the two TBV- infected tulip stocks which were used as virus-source for antiserum production. The antiserum prepared against virus from this stock showed activity against both TBV and the unknown virus.

The differences between ELISA-results obtained with both antisera may be explained by the presence of this second virus in part of the TBV-infected tulip samples tested.

As the new virus has not been found in lilies yet, it is not clear why the antiserum with mixed

activity shows higher ELISA-absorbance values with TBV-infected lilies compared with the other antiserum, while in microprecipitin tests both antisera have equal titres against TBV from several tulip cultivars.

G. JAGER (*Instituut voor Bodemvruchtbaarheid, Oosterweg 92, 9751 PD Haren*)

Effect of origin and degree of infection with *Rhizoctonia solani* of seed potatoes on subsequent infestation and formation of sclerotia on tubers of plants grown in different soils.

When seed potatoes carrying sclerotia of *Rhizoctonia solani* were planted in a slightly acid pleistocene sandy soil similar to the one in which they had been produced, the subsequent crop stood a better chance against *R. solani* infestation than a crop from seed potatoes that had been produced on a neutral holocene clay loam (JAGER & VELVIS 1980). A similar experience was reported by a farmer who grew potatoes on a holocene clay loam, which was known to be *Rhizoctonia*-suppressive (VAN EMDEN 1967). Sclerotia on new tubers were rare when he used non-disinfected seed potatoes from his own land.

It was also found that sclerotia from the sandy soils were frequently infected with hyperparasites, mainly *Verticillium biguttatum* (GAMS & VAN ZAAVEN 1982).

This was a reason to test the hypothesis that seed potatoes would be better protected against *R. solani* when planted in a "related" soil, because of the presence of antagonists indigenous to that soil, than when planted in a "strange" soil.

Experiments were set up on pleistocene sandy soils and on holocene loam or clay soils with seed potatoes from both origins ("sand seed" and "clay seed"). Four seed qualities were used: disinfected, clean, lightly and moderately infected with *R. solani*. Although the seeds in each class were visually equally covered with sclerotia, a larger proportion of the sclerotia on the "sand seed" proved to be dead than on "clay seed" (42% and 12% respectively). Results: Only one pleistocene soil type proved to be suppressive; the sclerotium index of tubers of plants from "sand seed" being much lower than that of "clay seed". On three other pleistocene soil types and one very light holocene soil the sclerotium index proved independent of the seed, due to a very strong *Rhizoctonia* infection from these conducive soils. Three holocene soils (pH 7.0–7.5) gave sclerotium indices for harvested tubers similar to those of the seed. The sclerotium indices of tubers grown from "clay seed" were always much higher than those from "sand seed". Also, the damage done by *R. solani* to stems and stolons of plants obtained from "clay seed" was always more serious.

The hypothesis thus was wrong. "Sand seeds" seem better protected. This may be due to the fact that the abundance of antagonists on the sclerotia and on the seed surface of "sand seed" was greater than on "clay seed".

In following field experiments, clean and infected "sand" and "clay seed" were used, treated or not treated with antagonistic fungi (*Verticillium biguttatum* and *Gliocladium roseum*).

Results: The differences in disease index between plants from infected "clay" and "sand seeds" had largely disappeared in two holocene soils when the seed had been treated with antagonists. In another holocene soil the differences still existed although less pronounced than in the pleistocene soils. Here the effect of treatment with antagonists often was negative. Plants from untreated, infected "clay seeds" still showed a distinctly higher disease index than those from "sand seeds". The damage to plants from clean seeds was relatively low, especially on the holocene soils.

With regard to the sclerotium index (of tubers harvested for seed), the effect of the origin of the seed had disappeared. Seed tubers treated with antagonists tended to yield a harvest with a lower sclerotium index than untreated seeds. This was very clear for the harvest of infected "sand seed" in holocene soils. Clean seeds, especially those treated with antagonists, produced a harvest with a very low sclerotium index.

In conclusion, the effect of a *Rhizoctonia* infection on the damage to the plants and the production of sclerotia depends more on the antagonistic microflora of the soil than on that of the seed potato. Secondly, the damage to the plants as well as the production of sclerotia can be reduced, at least in some soils, by using suitable antagonists. This is a subject for further research.

EMDEN J. H. VAN 1967. Beschouwingen over pathogene bodemschimmels. *Meded. Dir. Tuinb.* 30: 248–256.

GAMS, W. & Annemarie VAN ZAAZEN 1982. Contribution to the taxonomy and pathology of mushroom inhabiting *Verticillium* species. I. Taxonomy. *Neth. J. Plant Pathol.*, in press.

JAGER, G. & H. VELVIS 1980. Occurrence of *Rhizoctonia solani*-suppressive potato fields in the northern parts of The Netherlands. *Inst. Bodembevruchtbaarheid, Rapp. 1-80*, 62 pp. (In Dutch; headings of tables and figures and summary in English).

P. H. J. F. VAN DEN BOOGERT and H. VELVIS (*Instituut voor Bodembevruchtbaarheid, Oosterweg 92, 9751 PD Haren*)

Possibilities for biological control of *Rhizoctonia solani*, a pathogen of the potato plant

Biological control may be defined as the reduction of the harmful effects of a parasite or a pathogen through the use of other living entities.

*Rhizoctonia solani*, a pathogen to potatoes, is responsible for damage to stems and stolons and the production of sclerotia on new tubers, which strongly reduces their value as seed stock. This fungus occurs in many potato fields, possibly nearly as long as potatoes are grown.

The existence of soil properties that suppress *R. solani* in potato fields has been studied by JAGER & VELVIS (1980). The suppressive soils produce a nearly clean crop from clean seed potatoes and a rather clean crop from seed potatoes with sclerotia.

It is possible to induce suppressiveness through addition of living mycelium of *R. solani* to soil. The acquired property of an activated soil is detectable for at least 10 weeks at 15°C. Pot experiments showed that sprouts grown from contaminated seed potatoes in activated sandy, sandy loam and clay loam soils were almost unaffected; sprouts grown from contaminated seed potatoes in soils without added mycelium were strongly affected. It was also found that the mycoparasite *Verticillium biguttatum* frequently occurred in these activated soils.

It is assumed that *V. biguttatum* is an important cause of suppressiveness. The following experimental results support this hypothesis.

- 1 - Sclerotia of *R. solani* inoculated with propagules of an active strain of *V. biguttatum* were nearly all killed during an incubation period of 6 weeks at 15°C. The majority of untreated sclerotia was still alive at the end of that period.
- 2 - Sprouts grown from contaminated seed potatoes inoculated with propagules of *V. biguttatum* were less affected than sprouts grown from untreated contaminated seed potatoes.
- 3 - In a field experiment (on a soil that proved to be moderately suppressive) tubers were grown from -1 disinfected seed potatoes -2 contaminated seed potatoes and -3 contaminated seed potatoes inoculated with propagules of *V. biguttatum*. Tubers harvested from disinfected and contaminated seed potatoes inoculated with *V. biguttatum* were almost clean; tubers harvested from contaminated seed tubers were lightly infected with sclerotia of *R. solani*.

It is concluded that *Verticillium biguttatum* is a promising biological agent for control of *Rhizoctonia solani*.

JAGER, G. & H. VELVIS (1980): Onderzoek naar het voorkomen van *Rhizoctonia*-werende aardappelpercelen in Noord-Nederland (with an English summary). *Inst. Bodembevruchtbaarheid, Rapp. 1-80*, 62 pp.

LIESBETH VERVOORT, H. A. T. I. SWINKELS and H. R. VISSCHER (*Proefstation voor de Champignoncultuur, Peelheideweg 1, 5966 PJ Horst*)

Production of ethylene by mushroom mycelium and its microsymbiont in mushroom casing soil

Generative growth and fructification of mushrooms, *Agaricus bisporus* (Lge.) Imb. is thought to occur as result of symbiosis of mushroom mycelium and the microflora of the casing soil. It can be induced by lowering the CO<sub>2</sub> level of the ambient air. It was studied whether ethylene, one of the volatile metabolites of mushroom mycelium, plays a role in this process.

Ethylene was measured as produced by mushroom mycelium on sterilized compost or by sterilized or non-sterilized casing soil or by all combinations thereof.

Production of ethylene was found to be highest in full-grown compost covered by non-sterilized

casing soil. This then must be a result of symbiosis between mycelium and casing soil microflora.

Lowering of the CO<sub>2</sub> level leads to a disturbed CO<sub>2</sub>/ethylene balance. Analogous to higher plants this disturbance or the concentration of ethylene itself may cause changes in morphogenesis.

VISSCHER, H. R., (1979). Fructification of *Agaricus bisporus* (Lge.) Imb. in relation to the relevant microflora in the casing soil. *Mushr. Sci.* X, part I: 641–664 (*Proc. Xth Int. Congr. Sci. Cult. Edible Mushrooms*, Bordeaux 1978)

F. KLIS, M. ROOTJES, S. GROEN, C. SIGON and D. STEGWEE (*Plantenfysiologisch Laboratorium, Universiteit van Amsterdam, Kruislaan 318, 1098 SM Amsterdam*)

Formation of lesions on hypocotyledon sections of etiolated bean seedlings in response to heavy metal ions, cell wall hydrolytic enzymes and bean cell wall components

Hypocotyledons of 6-day-old dark-grown bean seedlings were wounded with a dissecting needle and on each wound a 5- $\mu$ l drop was placed. Browning was measured on an arbitrary scale of 0–5 (0 = no visible browning; 5 = very dark brown, streaky lesion). Some active compounds were 10<sup>-3</sup> M HgCl<sub>2</sub> and 10<sup>-3</sup> M AgNO<sub>3</sub>, Macerozyme R-10 and Onozuka cellulase, and wall components that had been released from isolated cell walls by Macerozyme R-10, Onozuka cellulase or heating. Application of only 0.2  $\mu$ g of cell wall components was enough to cause browning; mono- and disaccharides (Ara, Gal, Glc, Rha, Xyl; cellobiose, lactose, maltose, sucrose; 2% solutions, i.e. 100  $\mu$ g/drop) were, however, inactive. Preliminary experiments have shown that in browned tissue the incorporation of cell wall hydroxyproline was accelerated. These findings suggest that plant (cell wall) components can stimulate the synthesis of both phenolic compounds and hydroxyproline-containing wall protein.

J. VAN AARTRIJK and G. J. BLOM-BARNHOORN (*Laboratorium voor Bloembollenonderzoek, Vennestraat 22, 2160 AB Lisse*)

Effects of Virazole on the regeneration of virus-free plants from bulb-scale explants of *Lilium longiflorum* "Arai" infected with lily symptomless virus

Plants free of lily symptomless virus (LSV) can be obtained from many *Lilium* cultivars by meristem culture. For this purpose, we generally use isolated meristems formed adventitiously on bulb-scale explants *in vitro* (ASJES et al. 1974).

The present study was performed to find out whether the presence of Virazole (ribavirin; 1- $\beta$ -D-ribofuranosyl-1,2,4-triazole-3-carboxamide) in the meristem-inducing medium influences the chance of obtaining LSV-free plants from bulb-scales of *L. longiflorum* "Arai" infected with LSV. Leaf samples of the cultured plants were tested to detect the presence of LSV in two successive years by means of ELISA (BEIJRSBERGEN & VAN DER HULST, 1980), the immunodiffusion drop test (VAN SLOGTEREN 1976) and serologically specific electron microscopy (DERRICK & BRLANSKY 1976).

Addition of Virazole to the standard induction medium (final concentration 0.4, 4.0, or 40.0  $\mu$ M) did not significantly influence the regeneration process. However, in a concentration of 400.0  $\mu$ M Virazole significantly inhibited the process of adventitious formation of meristems.

The presence of Virazole (40.0  $\mu$ M) in the meristem-inducing medium led to a substantial reduction of the percentage of regenerated plants bearing LSV:

without Virazole: 38.2% LSV-infected plants (n = 75)

with Virazole: 10.8% LSV-infected plants (n = 88)

As could be expected, ELISA and serologically specific electron microscopy proved to be more sensitive than the immunodiffusion drop test for the detection of LSV in the *Lilium* plants (VAN SLOGTEREN et al. 1980).

The results are discussed in relation to the data on Virazole action in the literature.

ASJES, C. J., M. H. BUNT & D. H. M. VAN SLOGTEREN (1974): Production of hyacinth mosaic virus free hyacinths and lily symptomless virus free lilies by meristem tip culture. *Acta Horticult.* 36: 223–228.

BEIJRSBERGEN, J. C. M. & C. Th. C. VAN DER HULST (1980): Applications of enzymes during bulb

tissue extraction for detection of lily symptomless virus by ELISA in *Lilium* spp. *Neth. J. Pl. Path.* **86**: 227–283.

DERRICK, K. S. & R. H. BRLANSKY (1976): Assay for viruses and mycoplasmas using serologically specific electron microscopy. *Phytopathology* **66**: 815–820.

SLOGTEREN, D. H. M. VAN (1976): A single immunodiffusion drop test for the detection of lily symptomless virus. *Ann. Appl. Biol.* **82**: 91–95.

—, J. C. M. BEIJERSBERGEN, M. H. BUNT & C. Th. C. VAN DER HULST (1980): Detection of lily symptomless virus in leaves and bulb-scales of lily plants with the immunodiffusion drop test and with ELISA. *Acta Hort.* **110**: 91–98.

J. SALINAS CALVETE and D. H. WIERINGA-BRANTS (*Phytopathologisch Laboratorium "Willie Commelin Scholten", Javalaan 20, 3742 CP Baarn*)

The infectibility of cowpea mesophyll cells by Tobacco Necrosis Virus

The time required for infectious Tobacco Necrosis Virus (TNV) to pass through the epidermis of cowpea leaves after mechanical inoculation and to enter the mesophyll cells depended on plant age and environmental conditions. This "passage time" has been determined by removing the epidermis at different times after inoculation. At 22°C the passage time was about 3 h, but at 30°C and at water stress it was only 10 min. Water stress seemed to play a rôle in the transport of infectious virus into the mesophyll. The water stress had a positive effect only in the early stages of the infection process.

It seems that the passage of infectious virus material through the epidermis at water stress is momentary and does not depend on virus multiplication inside the epidermal cells.

It was possible to infect cowpea mesophyll cells with TNV directly by using a fine brush without carborundum.

SALINAS CALVETE, J. & D. H. WIERINGA-BRANTS (1981): The infectibility of cowpea mesophyll cells by tobacco necrosis virus. *Neth. J. Plant Pathol.* **87**: 211–216 (Meded. WCS 157).

B. J. M. VERDUIN, H. R. BLOKSMA and R. J. VANDEBRIEL (*Vakgroep Virologie, Landbouwhogeschool, Binnenhaven 11, 6709 PD Wageningen*)

Virus multiplication in cowpea leaves after differential temperature inoculation

Young cowpea (*Vigna unguiculata* (L.) Walp. CV. Blackeye Early Ramshorn leaves were "systemically inoculated" at 10°C with cowpea chlorotic mottle virus (CCMV), southern bean mosaic virus (SBMV) and a mixture of both using the differential temperature inoculation procedure (DTI) (Dawson, W.O and D. E. Schlegel (1976), *Intervirology* **7**, 284–291). Virus replication was initiated by moving the systemically inoculated leaves to a plant growth chamber at 25°C with a 14-hour photoperiod of 10.000 lux. Virus multiplication was monitored either using nucleoprotein measurements after virus isolation or Enzyme Linked Immunosorbent Assay (ELISA).

In leaves systemically inoculated at 10°C, CCMV-nucleoprotein was first isolated 24 h after the shift to 25°C and the maximum concentration was attained at 50 h. SBMV was first isolated at 12 h and remained at the same low concentration till 100 h. Viral nucleoprotein measurements demonstrated that a simultaneous inoculation of both viruses did not reduce the yield of either virus. The growth curves resembled those of the separately inoculated viruses.

Detection of viral protein by ELISA as a function of the infection period confirmed the above mentioned results. Both nucleoprotein measurements and ELISA showed a large variation in the data points, the cause of which is not yet known. The use of DTI favoured the synchronization of CCMV infection as compared to inoculation and infection under glasshouse conditions. Because of the low concentration of nucleoprotein in infected plants no such effect could be determined for SBMV.

F. P. GEELS and B. SCHIPPERS (*Phytopathologisch Laboratorium "Willie Commelin Scholten", Javalaan 20, 3742 CP Baarn*)

Effect of seed-tuber bacterization on the yield of potatoes grown in narrow rotations



Over two hundred fluorescent *Pseudomonads* isolated from the potato rhizosphere were screened for antagonistic activity in vitro against twenty-four fungal and bacterial pathogens and saprophytes. The antagonistic effects were caused by the production of siderophores, antibiotics, or both.

Preliminary bacterization experiments on potatoes with isolates of different antagonistic capacities were carried out in continuous-potato soil, in the climate-chamber. Some isolates were capable of reducing or even eliminating substantial yield loss (30%) due to the "narrow rotation effect" (NRE).

Subsequent field trials on various locations (soil types) in both wide and narrow rotations resulted in significant yield increase in some of the treatments, up to 20%; in the narrow rotations only. It seems likely that significant reduction of the NRE can be explained by the antagonistic activity of the fluorescent *Pseudomonads* against the microbiological factor(s) responsible for the NRE with potatoes (HOEKSTRA, 1981; LAMERS, 1981).

Migration and root colonization were confirmed by the use of antibiotic-resistant mutants of the antagonists.

HOEKSTRA, O. (1981): 15 jaar "De Schreef". Resultaten van 15 jaar vruchtwisselingsonderzoek op het bouwplannenproefveld "De Schreef".

*Proefstation voor de akkerbouw en de groenteteelt in de vollegrond*, no. 11, Lelystad.

LAMERS, J. G. (1981): Continue teelt en nauwe rotaties van aardappelen en suikerbieten. *Proefstation voor de akkerbouw en de groenteteelt in de vollegrond*, no. 12, Lelystad.

M. D. DE JONG and P. C. SCHEEPENS (*Centrum voor agrobiologisch onderzoek (CABO) Postbus 14, 6700 AA Wageningen*)

#### Control of *Prunus serotina* by *Chondrostereum purpureum*

*Prunus serotina* Ehrh. was introduced into The Netherlands and other European countries only a few decennia ago. However, it soon turned out to be a nuisance rather than a valuable species. The potential of *Chondrostereum purpureum* (Pers. ex Fr.) Pouz. as a control agent is being evaluated to replace the broad-spectrum herbicides that are now being used. *C. purpureum* is a common saprophyte in dead wood. As a wound invader, it can cause silver leaf disease of *P. serotina* and other deciduous trees.

Several experiments were started in 1980 in which stubs of *P. serotina* were inoculated with mycelium of *C. purpureum*. Stubs inoculated in April and May 1980 were dead in October 1980. Most of the stubs treated in the fall of 1980 were dead or nearly by the end of September 1981. When only one stub of a shrub was treated, non-treated shoots on the same root system were also affected by the disease.

Before *C. purpureum* can be used as a regular control agent for *P. serotina*, some additional requirements have to be met:

- Fungal formulations should be stable for at least several months; application should be possible by simple, conventional techniques.
- Risks to other deciduous trees should not be enlarged by application of the fungus to *P. serotina*. Although there are many indications against this being so, additional research on host range and spore dispersal of the fungus is necessary.

P. ROSEBOOM and D. PETERS (*Vakgroep Virologie, Landbouwhogeschool, Binnenhaven 11, 6709 PD Wageningen*)

#### The development of symptoms on the foliage of beet plants after injection with beet yellows virus

Beet yellows virus is detectable in infected leaves with the ELISA test. Quantitative assays are performed with homogenates of discs 5 mm in diameter from infected leaves and 0.2 ml buffer. The virus can also be detected in a qualitative way by incubating unmacerated leaf discs with buffer in wells of ELISA plates for some hours.

With the developed ELISA technique we studied the distribution of the virus in all the leaves of beet plants. Plants with 25 leaves were inoculated by aphids either at the youngest, at one of

the middlest, or one of the oldest leaves. Analysis of the foliage of these plants at several moments after inoculation showed that virus could be detected in the inoculated leaves and in those which started to develop at the moment of infection or later. This distribution of virus over the foliage could be confirmed in tests for infectivity with aphids.

The symptoms due to systemic infection occurred in a whorl of leaves which were formed just at the moment that virus was introduced in the plant or later. This observation enables us to make an estimate of the moment the plant attracted infection. For that purpose the position of the oldest leaf with symptoms, expressed by C, in the whorl has to be determined and total number of leaves (N) on the beet has to be counted on several occasions. As the position of the oldest leaf with symptoms does not change and the total number of leaves increases the ratio C/N will decrease with time. The C/N ratios found at several moments during the growth of a plant or a group of plants form a line which can be used to estimate the date of infection when plants are available of which the date of infection is known.

The results obtained in 1981 in field experiments show that a rather exact estimate is feasible.

**F. A. VAN DER MEER** (*Instituut voor Plantenziektenkundig Onderzoek (IPO), Binnenhaven 12, 6709 PD Wageningen*)

Knobbiness of poplar, a disease caused by an unknown graft-transmissible agent.

Knobbiness of poplar is known since 1974 as a serious disease of the poplar cultivar Rap, a hybrid of *Populus trichocarpa* and *P. deltoides*. Affected trees show knobs, varying in size between 1 and 3 cm, which usually occur only locally on some parts of stems and branches. Smaller swellings of the bark and inner bark necrosis are present all over diseased trees. Knobs and smaller swellings contain tumours which often start to develop around bark fiber bundles. Opposite big knobs on the bark, tumours may also develop in the wood. Both tumours in the bark and tumours in the wood usually have a necrotic centre. Symptoms in the wood of older branches often show much similarity with "stem pitting", a symptom known from several woody hosts and often caused by viruses.

Repeated attempts to isolate bacteria or fungi from affected trees have failed so far. Recently the disease was transmitted from diseased "Rap" to healthy "Rap" by grafting, thus proving its infectious nature. Several other poplar clones without knobs, but showing inner bark necrosis and a diffuse mottle in their leaves, also induced knobbiness after grafting them to healthy "Rap".

In preliminary attempts no viruses could be detected in diseased trees. However, the type of symptoms and the transmissibility of the disease indicate that knobbiness of poplar could possibly be caused by a virus.

**F. A. VAN DER MEER** (1981): Mozaiekvirus, heksenbezem en knobbelziekte bij populier, en een virusachtige groeiremmering bij wilg (with English summary: Mosaic, witches' broom and knobbiness of poplar and a virus-like growth reduction of willow). *Populier* 18: 51-59.

**N. A. M. VAN STEEKELENBURG** (*Instituut voor Plantenziektenkundig Onderzoek, Binnenhaven 12, 6709 PD Wageningen, gestationeerd Proefstation voor Tuinbouw onder Glas, Zuidweg 38, 2671 MN Naaldwijk*)

Influence of the glasshouse climate on development of *Didymella bryoniae* on cucumber

Summary of the article:

**STEEKELENBURG, N. A. M. & J. VAN DE VOOREN** (1981): Influence of the glasshouse climate on development of diseases in a cucumber crop with special reference to stem and fruit rot caused by *Didymella bryoniae*. *Acta Hort.* 118: 45-56.

## MEETING OF THE NETHERLANDS SOCIETY FOR PLANT CELL AND TISSUE CULTURE ON NOVEMBER 6, 1981

**P. MALIGA** (*Institute of Plant Physiology, Biological Research Center Hungarian Academy of Sciences, Szeged, P.O.Box 521, H-6701 Hungary*)

### Protoplasts of flowering plants and their use in cell genetic manipulation

*Nicotiana plumbaginifolia* is a suitable model species for basic studies concerning the use of protoplasts in cell genetic manipulations. Haploid and diploid protoplasts are being used for selecting auxotrophic, pigment-deficient and antibiotic-resistant lines. Protoplast-derived cultures provide certain advantages for screening mutants over other culture types. Cell fusion serves to characterize the selected lines and to rescue mutations from non-morphogenetic cells. Methods to construct improved plants include somatic hybridization, transfer of organelles and chromosomes, and protoplast-protoplast and cytoplasm-protoplast fusion (the cytoplasmic male sterility factor).

**L. J. W. GILISSEN, J. BLAAS and P. A. Th. J. WERRY** (*Stichting ITAL, Postbus 48, 6700 AA Wageningen*)

### Induction and selection of mutants for chromosome transplantation in *Haplopappus*

Chromosome markers are necessary tools in the development of chromosome transplantation techniques. On the cellular level these markers (stable chromosome mutations) can be induced and selected biochemically (THOMAS 1981).

A general survey has been given of our mutation research with cell suspension cultures of *Haplopappus gracilis* ( $2n = 4$ ). Attempts to isolate mutants resistant to the antibiotics emetine, kasugamycin and cycloheximide resulted in a number of cell lines resistant to cycloheximide. But this character appeared to be unstable. Besides, the toxic concentration of the herbicides picloram and 2,4-D, at which resistant cell lines can be selected, were defined. Furthermore, selection procedures are in progress to obtain cell lines which are tolerant to sodium chloride or which are deficient in their nitrate reductase activity. Finally, zinc-tolerant cell lines were selected at the toxic concentration of  $7 \cdot 10^{-3}$  mol zinc. The tolerance appeared to be stable after culturing the cells in absence of zinc during 20 successive cell cycles. Therefore, the character of zinc tolerance might be useful as a chromosome marker, if its inheritance appears to be monofactorial and dominant.

THOMAS, B. (1981): Bibliography of mutant isolation from plant cell cultures. *Plant Molecular Biology Newsletter* 2: 77-89.

**G. S. BOKELMANN and S. ROEST** (*Stichting ITAL, Postbus 48, 6700 AA Wageningen*)

### Regeneration of plants from protoplasts of potato

A method is described of plant regeneration from shoot-derived protoplasts. The procedure was published by BINDING et al. (1978) for dihaploid potato clones and adapted to the dutch tetraploid cultivar Bintje.

Shoots were cultured on Murashige and Skoog (MS)-medium, in the absence of hormones and supplemented with sucrose 1%. The upper part of the shoots was cut into small fragments in a few droplets of the isolation medium (cellulase 1% and macerozyme 0.2% in 0.6 M mannitol). Glass bottles containing 50 ml enzyme solution per gram of plant material were placed on a roller (Rollacell RC-42) for 4 hours at 26°C and 1000 lux. After the enzyme treatment the material was filtered, washed several times by centrifugation, purified in sucrose 0.6 M, washed and cultured in liquid medium (V-KM without  $\text{NH}_4\text{NO}_3$ ) at a protoplast titre of  $5 \cdot 10^4$  per ml (1.6 ml culture medium in a 5 cm plastic petridish). The osmolality of all solutions and the culture medium was fixed at 650-700 mOsm. Cell wall regeneration and the first cell divisions were observed after 3 days. Small cell aggregates (10-50 cells) were formed after a culture for about 2 weeks, after which period the aggregates were cultured in a semi-solid (0.2% agar) V-KM medium for another 2-3 weeks. Calli were produced and transferred to a solid MS-medium for further growth and, finally,

subcultured on MS-medium in the presence of zeatin (1 mg/l), NAA (0.01 mg/l) and GA<sub>3</sub> (0.03 mg/l) about 2 months after protoplast isolation. The first adventitious shoots were initiated within 3 weeks and complete plantlets were produced after root formation of excised shoots on MS-medium supplemented with IAA at 0.1 mg/l.

The whole procedure from protoplast isolation till plant production takes 3–4 months, but up till now the reproducibility has to be improved before this technique can be used for genetic manipulation.

H. BINDING, R. NEHLS, O. SCHIEDER, S. K. SOPORY & G. WENZEL (1978): Regeneration of mesophyll protoplasts isolated from dihaploid clones of *Solanum tuberosum*. *Physiol. Plant.* **43**: 52–54.

A. J. KOOL<sup>1</sup> and G. A. M. VAN MARREWIJK<sup>2</sup> (<sup>1</sup>*Afdeling Genetica, Vrije Universiteit, De Boelelaan 1087, 1081 HV Amsterdam*; <sup>2</sup>*Instituut voor Plantenveredeling (IVP), Lawickse Allee 166, 6709 DB LH-Wageningen*)

Molecular basis and transfer of cytoplasmic male sterility in *Petunia hybrida*

The maternally inherited trait of cytoplasmic male-sterility (CMS) is of great economic importance because it can be used in the commercial production of hybrid seed to eliminate self fertilization of the seed parent plant. To increase the applicability of this trait, which is as yet limited because this trait is only found in a small number of plant species, we study the molecular basis of CMS in *Petunia hybrida* and investigate the possibility of the transfer of CMS from *Petunia* to other, economically important plant species by somatic cell hybridization. The molecular basis of CMS was determined by analysis of chloroplast (cp) and mitochondrial (mt) DNA by restriction endonucleases. We observed that the enzymes Bam HI, Bgl I or Sal I did not show any variation among cpDNA isolated from the fertile and CMS *Petunia hybrida* cultivars "Rosy Morn", "Blue Bedder" and "Snow Ball", which makes a possible role of chloroplasts in CMS less likely. Restriction endonuclease patterns obtained after Bam HI digestion of mtDNA from CMS and fertile *Petunia hybrida* cv. "Rosy Morn" showed a number of variations. Also other restriction enzymes indicated the presence of differences between mtDNA from the two cytoplasmic types. Therefore those results indicate that in *Petunia hybrida* mitochondria, rather than chloroplasts, are the coding site of factors conditioning cytoplasmic male-sterility. Present research is focussed on determining whether the observed differences between fertile and CMS mtDNA result in different expression patterns. For this purpose in vitro protein synthesis is studied in mitochondria isolated from fertile, CMS and restored fertile *Petunia hybrida* lines.

H. J. WICHERS (*Laboratorium voor Farmacognosie en Galenische Farmacie, Rijksuniversiteit, Antonius Deusinglaan 2, 9713 AW Groningen*)

Preparation, purification and cytology of protoplasts derived from in vitro cultures of *Symphitum officinale*

H. WINTER, C. J. LEURS and P. K. WIERSEMA (*Centraal Isotopen-Laboratorium, Biologisch Centrum, Kerklaan 30, 9751 NN Haren (Gr)*)

Preparation, purification and ion-transport of mesophyll protoplasts of pea

R. WIJNSMA (*Laboratorium voor Farmacognosie en Galenische Farmacie, Rijksuniversiteit, Antonius Deusinglaan 2, 9713 AW Groningen*)

The production of catecholamines in in vitro cultures of *Mucuna pruriens*

## MEETING OF THE NETHERLANDS SOCIETY FOR PLANT CELL AND TISSUE CULTURE ON MARCH 12, 1982

R. L. M. PIERIK (*Laboratorium voor Tuinbouwplantenteelt, Haagsteeg 3, 6708 PM Wageningen*)

### Critical evaluation of vegetative propagation in vitro

Over the last ten years the number of papers on in vitro propagation has grown enormously. At present about 800 plant species can essentially be cloned in vitro. The number of genera propagated in The Netherlands in numbers of more than 100,000 per year is only nine (*Lilium*, *Cymbidium*, *Gerbera*, *Anthurium*, *Nephrolepis*, *Davallia*, *Solanum*, *Saintpaulia*, *Philodendron*). This indicates that there is still a wide gap between our knowledge from scientific publications and its application for mass propagation in vitro. In this paper special attention will be paid to the causes of this gap. In The Netherlands the number of research workers bridging the gap between science and application is too small.

Mass cloning of plants in vitro is limited by a number of factors. We do not understand the fundamental causes of the occurrence of mutations, including the possible role of growth regulators. Basic knowledge of the formation of organs and embryos, particularly under the influence of growth regulators, is rather limited and not much is known about the loss of the ability to form organs. The impossibility to regenerate roots on shoots of adult shrubs and trees is a serious handicap. Possibilities to induce rejuvenation are limited because induction of adventitious buds is often extremely difficult. Relatively little attention is being paid to the influence of physical growth factors, in particular to the gas phase in the test tube. The building up of brownish substances as a result of the oxidation of phenols and also the physiological disease "vitrification" are very difficult to counteract. In vitro produced plants often are not well adapted to the climate outside the test tube. Mass propagation in vitro is hampered by a number of problems which can, essentially, be solved. Too many cultures are lost by infections; if one is confronted with internal infections, meristem culture is the only way out. In vitro propagation systems often are not fully optimized, resulting in too high a cost price. Frequently timing of production is not ideal so that labour and laboratory space and equipment are not efficiently used. Too often model studies are carried out with only one genotype, which restricts the validity of the system. Propagation of chimaeric plants in vitro is very dangerous because adventitious bud formation leads to the uncovering of chimaeras.

Application of the axillary branching system often results in plants with a strong branching habit. To improve rooting and to decrease the cost price of test tube plants, in vitro formed shoots might be subjected to conventional or alternative methods of rooting outside the test tube.

E. A. ZANDVOORT and G. STARITSKY (*Vakgroep Tropische Plantenteelt, postbus 341, 6700 AH Wageningen*)

### In vitro culture of tropical woody crops in connection with vegetative propagation and genetic conservation

Clonal propagation is an important starting point for the improvement of production and quality of tropical woody crops, but can lead to genetic impoverishment at the same time. Therefore it is very important to conserve the genes that underlie the genetic variability of the crop.

In vitro culture offers possibilities for vegetative propagation and for preservation of plants as well. The endeavour for optimum results is not very useful in a practical sense, as it would lead to a separate, refined treatment of every crop and every cultivar. Therefore, research at the department is aimed at the development of a simple method, suitable for general application, for the propagation and conservation of tropical woody crops in vitro.

Isolated buds of young and old trees are used as starting material. As a rule the same basic medium is always used, whereby at most the concentration of growth regulators is varied.

At the moment research is being done with about 50 species and cultivars of the following genera: *Anacardium*, *Artocarpus*, *Camellia*, *Cinchona*, *Cinnamomum*, *Citrus*, *Coffea*, *Cola*, *Derris*, *Eu-*

*calyptus, Eugenia, Macadamia, Mangifera, Olea, Persea, Piper, Richardella, Simmondsia and Theobroma.*

The initial results of the experiments are promising in most cases.

**P. A. T. J. WERRY** (*Directie Akker- en Tuinbouw, Ministerie van Landbouw en Visserij, Postbus 20401, 2500 EK 's-Gravenhage*)

The application of tissue culture in horticultural practice

**P. M. A. VAN WELL** (*Hogere Agrarische Scholen, Baden Powellstraat 1, 5212 BW 's-Hertogenbosch*)

A tissue culture course at the Horticultural College at 's-Hertogenbosch

**J. MARINUS** (*Centrum Agrobiologisch Onderzoek, Postbus 14, 6700 AA Wageningen*)

The propagation of potatoes in vitro: limitations and results

**J. KOSTER** (*Botanisch Laboratorium, Nonnensteeg 3, 2311 VJ Leiden*)

Shoot formation on leaf explants of *Nautilocalyx lynchii*

**J. VAN AARTRIJK** (*Laboratorium voor Bloembollenonderzoek, Postbus 85, 2160 AB Lisse*)

Indications for a role of ethylene in the process of adventitious shoot formation on lily tissue

# **AUTUMN MEETING OF THE ROYAL BOTANICAL SOCIETY OF THE NETHERLANDS AND MEETING OF THE SECTION FOR VEGETATION RESEARCH ON DECEMBER 15, 1981**

**I. S. ZONNEVELD** (*International Institute for Aerial Survey and Earth Science, Enschede*)

Zonneveld about Westhoff

**V. WESTHOFF** (*Afdeling Geobotanie, Toernooiveld, 6525 ED Nijmegen*)

Impressions of Australis and Capensis

**A. J. DEN HELD** and **P. H. M. A. CLAUSMAN** (*Provinciale Planologische Dienst van Zuid-Holland*)

Flora and vegetation of the agricultural polder landscape of the province of South-Holland, in particular of the grasslands

In the study of vegetation in The Netherlands usually little attention is paid to the agricultural grassland landscape. This is probably the cause of the relatively low appreciation of these areas in valuation procedures related to town and country planning (ANONYMUS 1976, 1981).

It was only 1976, when an investigation was started of the vegetation of the grasslands of the province of South-Holland, including the ditches and their banks. These grasslands cover an area of about 1500 km<sup>2</sup>; ditches and banks have a total length of over 30,000 and 60,000 km, respectively. In 1976-1980 the area has been sampled by means of relevés according to the French-Swiss School (about 20-30 relevés per km<sup>2</sup>). This method has been chosen mainly because of the fact that relevés offer data which can be analysed in many different ways, and because of the lack of vegetation classifications from which adequate descriptive units can be derived.

Some preliminary results are presented here.

The vegetation of the grasslands and their banks often shows a gradual change, which parallels a gradient in the grazing and manuring pressure, decreasing from the surroundings of the farms to the hinterland. In this last zone there are still many meadows with a bank vegetation that is often very rich in species.

A number of more or less rare taxa, growing under lightly manured, damp environment, are at least as numerous in the agricultural area as in the nature reserves within this area, e.g. *Cirsium dissectum*, *Potentilla anglica*, *Pedicularis palustris*, *Carex panicea*, *C. ovalis*, *Fritillaria meleagris*, *Chrysanthemum leucanthemum*, *Menyanthes trifoliata*, *Ranunculus lingua*. Some even do not occur (any longer) in these reserves, e.g. *Viola persicifolia* ssp. *persicifolia*, *Alchemilla vulgaris*, *Taraxacum* sections *Palustria* and *Spectabilia*, *Veronica scutellata*. Also, a number of more common species have their main distribution in the agricultural area, e.g. *Triglochin palustris*, *Carex disticha*, *Ranunculus flammula*, *Apium nodiflorum*. This indicates the importance of lightly cultivated parts of the polder landscape, which on the one hand are threatened by a more intense management in the agricultural area itself, but which, on the other hand, are disappearing in the nature reserves because of the decreasing nutrient contents and the disappearance of grazing and/or mowing as environmental factors.

As for the water vegetation, there are many ditches with little or not polluted, eutrophic water, in which often a richly structured vegetation occurs; species like *Ranunculus circinatus*, *Elodea canadensis*, *Potamogeton trichoides*, *P. natans*, *Zannichellia palustris*, *Myriophyllum spicatum*, *Hottonia palustris*, *Nymphoides peltata*, *Stratiotes aloides*, *Butomus umbellatus*, *Alisma gramineum*, Characeae, etc., are common here. An unexpected phenomenon is the occurrence of a vegetation type with plants of meso- to oligotrophic conditions, like *Juncus bulbosus*, *Scirpus fluitans*, and *Echinodorus ranunculoides*, mixed with species like *Potamogeton natans* and *trichoides*, *Elodea nuttallii* and *canadensis*, *Nymphaea alba*, and *Sparganium emersum*, in isolated ditches mainly in the eastern parts of the reclaimed polders, on acid older marine clay deposits, poor in calcium; this vegetation type also occurs on some places in the peat areas.

Not only are there great differences in vegetation within one region (as Vijfheerenlanden, Alblas-serwaard, Krimpenerwaard, Midden-Delfland, Voorne), but also between the regions. Each region has a different, characteristic vegetation, most probably caused by differences in soil, salt contents of the water, and management.

One of the most important conclusions is that the agricultural grassland landscape of South-Holland or, in general, of all the lower parts of The Netherlands, has an until now highly underestimated botanical significance in regional and in national respect. From an international point of view, this importance even is much greater, because of the fact that the plant communities concerned are extremely characteristic of The Netherlands, and are consequently rare. Therefore it should be emphasized that this vegetation is severely threatened by the still accelerating increase of agricultural pressure. Serious study of the vegetation of the polder landscape may contribute to a more careful attitude.

ANONYMUS (1976): Samenvatting Landelijke Milieukartering. *Studierapporten Rijksplanologische Dienst* 5.3.A. Den Haag.

ANONYMUS (1981): *Structuurschema Natuur- en Landschapsbehoud*. Ministerie van Cultuur, Recreatie en Maatschappelijk Werk; Ministerie van Volkshuisvesting en Ruimtelijke Ordening. Den Haag.

WESTHOFF, V. & A. J. DEN HELD, (1969): *Plantengemeenschappen in Nederland*. Thieme. Zutphen.

A. J. M. ROOZEN (*Afdeling Geobotanie, Toernooiveld, 6525 ED Nijmegen*)

Vegetation changes in salt marshes; 28 years PQ-research on Terschelling

Succession in salt marsh plant communities on the Boschplaat of Terschelling (The Netherlands) is described by means of vegetations relevés from 40 permanent plots, arranged in three transects. In total 489 relevés were made in the period 1953–1980. These data were analysed with numerical methods, whereafter a succession diagram was constructed.

The succession is strongly related to the altitude of the soil surface. In the salt marsh four zones can be distinguished with a different development of the vegetation, corresponding to the main successional lines. 1. Low level salt marsh (around mean high water): *Puccinellietum maritimae typicum* – community of *Limonium vulgare* and *Halimione portulacoides* – *Halimionetum portulacoides*. 2. Medium level salt marsh (up to mean high water at spring tide): *Puccinellietum maritimae*, transition from *typicum* to *parapholietosum* – *Plantagini-Limonietum* – *Artemisietum maritimae*. 3.

High level salt marsh (only inundated at storm tides): *Puccinellietum maritimae parapholietosum* – *Junco-Caricetum extensae* – *Armerio-Festucetum* – *Artemisietum maritimae*. 4. Drift-line zone: Species-rich communities with elements of the *Ammophiletea* and *Violo-Corynephoretea* – *Atriplici-Agropyretum pungentis*. The most important environmental process in the covered period is the sedimentation of fine particles on the formerly sandy soil. In low level salt marsh a better aeration of the soil, due to raising of the soil surface by sedimentation, is of major importance. In medium and high level salt marsh the increasing salt and nutrient content of the soil by the formation of a silt layer is probably most relevant.

In the drift-line zone the deposition of plant litter is responsible for the described changes.

During the succession the differences between plots decrease. An increased homogeneity over the zonation is also detectable in the converging development of the different successional lines in the *Artemisietum maritimae*. At a continuing sedimentation a strong extension of this community can be expected.

H. W. J. VAN DIJK (*Vakgroep Milieubiologie, Kaiserstraat 63, 2311 GP Leiden*)

Activity-impact-relationships of water supply by infiltration in Dutch coastal dunes; especially relationships between water quality and spontaneous vegetations on banks

Research at the Department of Environmental Biology of the University of Leiden concentrates on quantifying the effects of human activities on natural environment and on proposing and developing alternatives for human activities threatening environment and nature.

In the project described effects were studied of infiltration of polluted river water on spontaneous vegetation in water catchment areas in the Dutch coastal dunes. By nature this vegetation is rich and diverse, containing many rare species. Infiltration influences the vegetation in various ways: ground works, disturbance and fluctuation of water level and very high supply of nutrients. Some results of the study of effects of the unnaturally high supply of macronutrients were mentioned in relation to general problems with quantifying activity-impact relationships.

On banks of directly infiltrated lakes as well as of less influenced seepage lakes, vegetation after 25 years of infiltrating is mostly dominated by ruderal plant species ("competitors" after J. P. Grime, "Stickstoffzeiger" after H. Ellenberg). Their dominance may be explained by the high nutrient content of the infiltrated water, especially of orthophosphate. A much better explanation is provided by the continuous supply – instead of the content – of macronutrients, as calculated from the measured speed of ground water flow and the concentration of the limiting macronutrients N, K and P in bank infiltration water. For example the cover of the extremely ruderal *Urtica dioica* increases from less than 20% to nearly 100% with an increase of the phosphate supply from 0.2 to 0.6 grams P-orthophosphate/m<sup>2</sup>.day. Comparable results were found with other, less extremely ruderal species.

These and other results were used to illustrate general problems in quantifying activity-impact relationships, such as selection of activity and effect parameters; determination of the most relevant scale of time and space; generalization of the relationships between activities and effects; assessment of the human impact; choice of management, within proposed activity or alternatives (possibly zero), to limit damage of nature and environment.

On the basis of quantified activity-impact relationships calculations could be made for predicting how the environmental impact of water infiltration in the dunes might be limited. Such knowledge is significant for the choice between continuation and expansion of water infiltration in the dunes or alternative water supply plans.