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# THE EFFECT OF AIR POLLUTION ON TRANSPLANTED MOSSES

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The use of forest mosses as bioindicators was tested with transplantation experiments. One transplantation experiment was made to study effects of air pollutants on two forest moss species, *Hylocomium splendens* and *Pleurozium schreberi*. Another transplantation was used to study he nitrogen fixation capacity of blue-green algae in the *Hylocomium* and *Pleurozium* moss layers. The surface structure of the moss species was studied by scanning electron microscopy. The air pollution induced changes in the surface structure of moss cells were observable soon after the transplantation. In polluted industrial areas the fertilizing effect of air-borne nitrogen compounds increased the photosynthetic activity of mosses before their destruction. Stress respiration was also observable in polluted areas. The nitrogen fixing capacity decreased or was almost inhibited in all the air-polluted environments.

## INTRODUCTION

Two common moss species in Finnish forests, *Hylocomium splendens* (Hedw.) B. S. G. and *Pleurozium schreberi* (Brid.) Mitt., were used as bioindicators in transplantation experiments in air polluted areas. The levels of $CO_2$ fixation and $N_2$ fixation were studied in the transplanted mosses in polluted and clean environments.

Several studies have proved mosses to be very sensitive to air pollutants (i.e. GILBERT 1968, 1969, LeBLANC 1969, LeBLANC et al. 1971, WINKLER 1976, WINNER and BEWLEY 1978a and b, FERGUSON and LEE 1978). Air pollutants affect the growth and reproduction of mosses. Several species have been shown to disappear from industrial and urban environments, though suitable stands for moss growth can still be found (RAO and LeBLANC 1967, GILBERT 1968, BARKMAN 1969). The suitability of mosses to bioindication is mainly due to their primitive morphological and anatomical structure. Mosses can absorb and accumulate nutrients and pollutants directly from the atmosphere. The use of mosses as indicators of soil, water and air pollution and the importance of mosses as accumalators of metal ions have recently been reviewed by HISATSUGU 1980, LeBLANC and RAO 1974.

The nitrogenase activities in moss epiphytic blue-green algae were used as one of the indicators of air-pollution induced effects on mosses. BASILIER et al. (1978) found the nitrogenase activity of mosses to correlate with the number of blue-green algae in natural clean air areas. This is in accordance with our findings obtained during several years of study (KALLIO, unpubl.). Only few studies of pollution effects have been made earlier using nitrogenase activity measurements by asetylene reduction (KALLIO and VARHEENMAA 1974, HÄLLGREN and HUSS 1975, KALLIO 1976).

The morphology of *Hylocomium splendens* with groove-like surface structure of leaves and paraphyllia provides a good shelter for algae against drying. *Pleurozium schreberi* is a moss of somewhat dryer forests, and its water-holding capacity is not so good as that of *Hylocomium*. The effects of air pollution on moss surfaces were studied by monitoring the moss surfaces with a scanning electron microscope.

## MATERIAL AND METHODS

The transplantation of mosses was made in several industrial and urban areas of Oulu in 1977–1979 (Fig. 1.). The nitrogenase activity was tested with several transplantations in the Oulu, Kokkola and Turku areas during two years (1978 and 1979) (Fig. 2.). The material for the transplantations was obtained from natural forests at Ii 65°20′N and outside the urban areas of Oulu, Turku and Kokkola. Control moss layers were also transplanted. The mosses were detached from the ground similarly to the transplantations made in polluted areas. The size of the transplanted layers was about 40 cm x 60 cm. The earliest transplantations were made at the end of May as soon as the ground had thawed (for futher details, see KARHU 1981). Standardization of URAS 2 $CO_2$-exchange measurements: light intensity 10 000 lux, temperature +5–+6°C, $CO_2$ -air flow 20 l $h^{-1}$, moistening of moss samples in moisture chambers for 12 h before the gas exchange measurements.

The surface structures of the mosses were studied with a scanning electron microscope JEOL JSN-35. The moss material was deep-frozen. A thin 10–40 nm cold film was used. The magnifications used for the micrographs were 100x and 1000x. The tip leaves of the mosses were micrographed (the Institute of Electron Optics, Unifersity of Oulu).

The nitrogenase activities in the moss blue-green algae were measured in early September in the years 1978 and 1979 under laboratory conditions for minimizing the variable light, moisture and temperature effects. The samples (top parts of the newest moss growth) from the different areas were collected and sent to the laboratory of the Department of Biology of the Turku University. Acetylene reduction analyses were performed in laboratory conditions after one night pretteatment at constant moisture, temperature and light. Commercial acetylene gas (AGA) was used as a substrate for nitrogenase, and the results are expressed as ethylene (nmol reduced from acetylene) per one gram of moss dried at 105°C. Moss sample incubation with acetylene was carried out at 20°C for one or two hours.

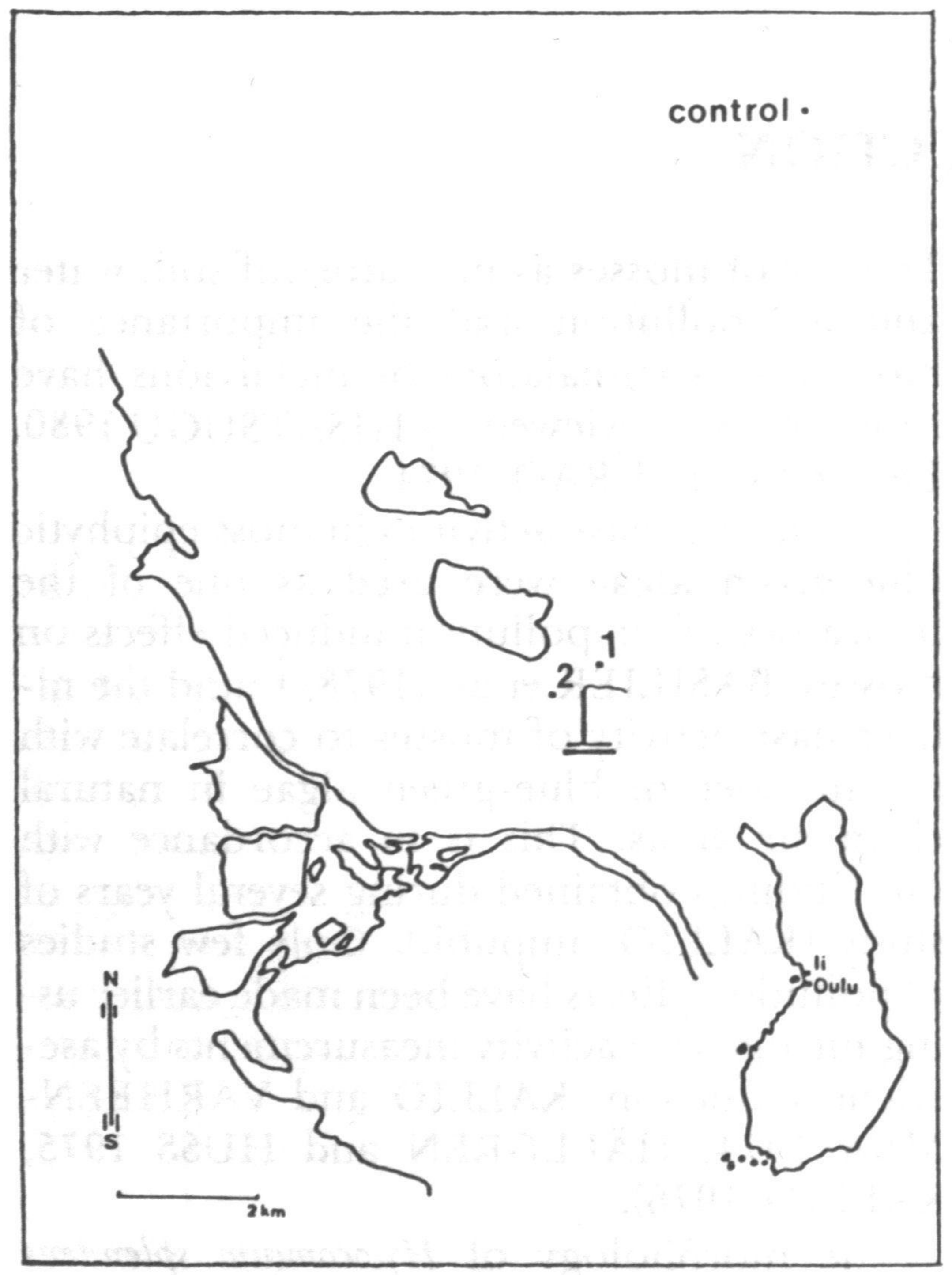


Fig. 1. Location of the moss transplantation sites in Oulu during 1977–1979 (sites 1 and 2), and location of the control transplantation (control). Part of the material was obtained from Ii (in small map).

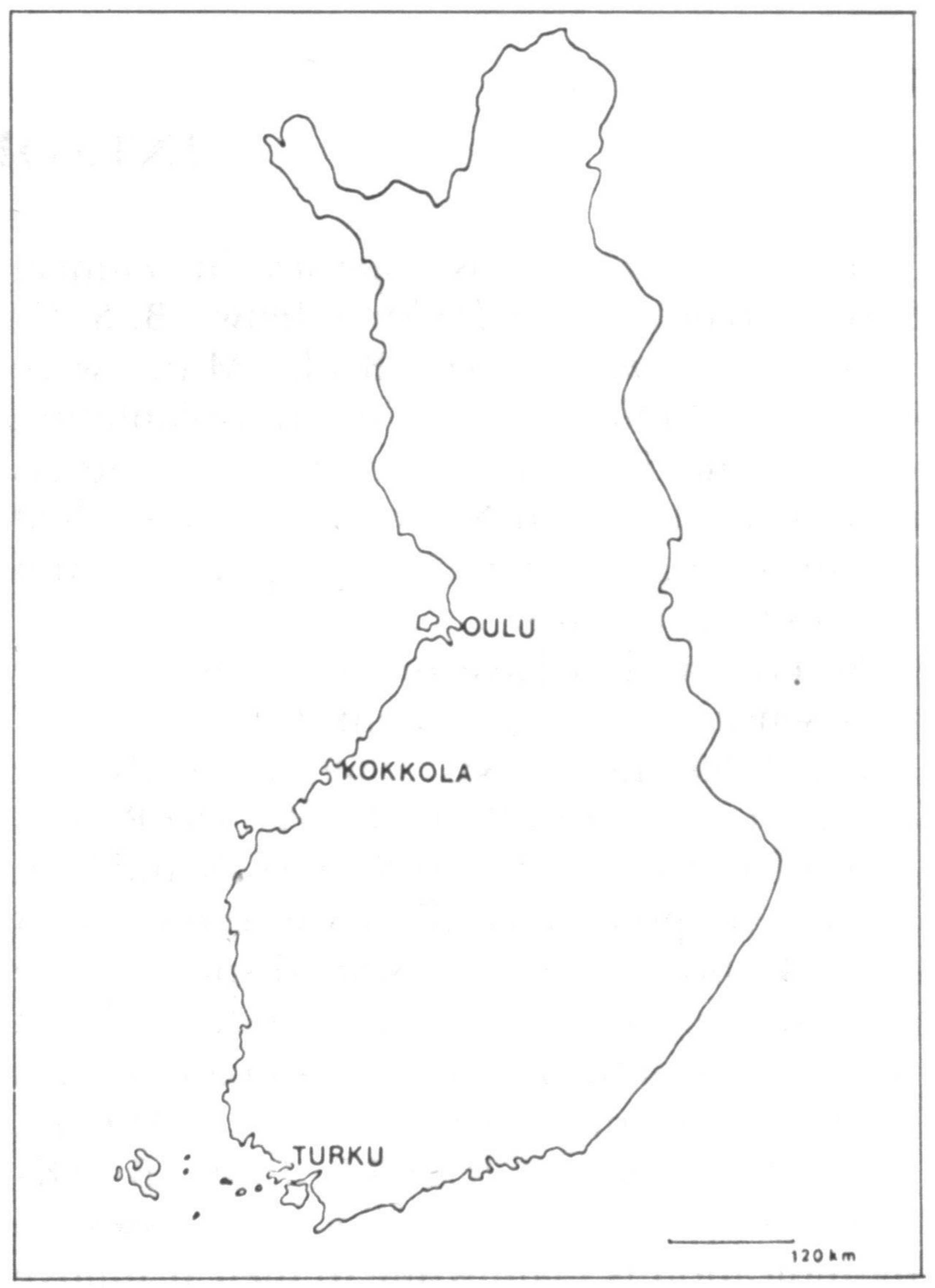


Fig. 2. Location of the moss transplantation areas of the nitrogenase activity studies (1978 and 1979), the towns of Oulu, Kokkola and Turku.



## RESULTS AND DISCUSSION

### Changes in moss surface structure (SEM)

The epidermal surface structure of natural growing *Hylocomium splendens* is characterized by a slightly wavy structure and small papilloid appendages (Fig. 3a). The additional small phylloid structure is destroyed and the wavy nature disturbed under air pollution conditions (Fig. 3b). *Pleurozium schreberi* also has a wavy, but twist-

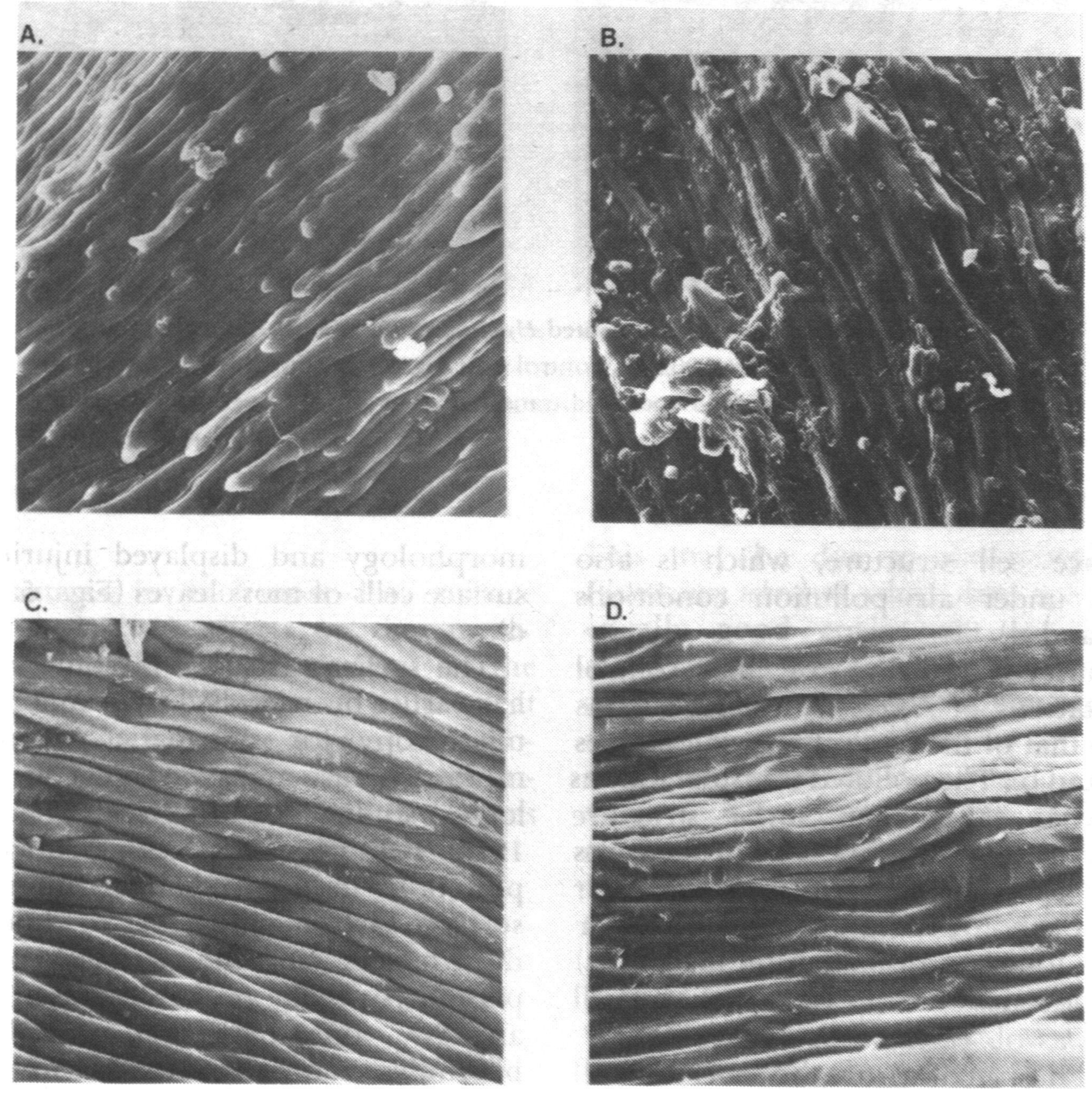


Fig. 3. The surface structure of natural growing mosses in normal and urban forest stands near the transplantation sites. a. *Hylocomium splendens* control area, b. *Hylocomium splendens* polluted area, c. *Pleurozium schreberi* control area, d. *Pleurozium schreberi* polluted area. 27. 9. 1978 (1000x).

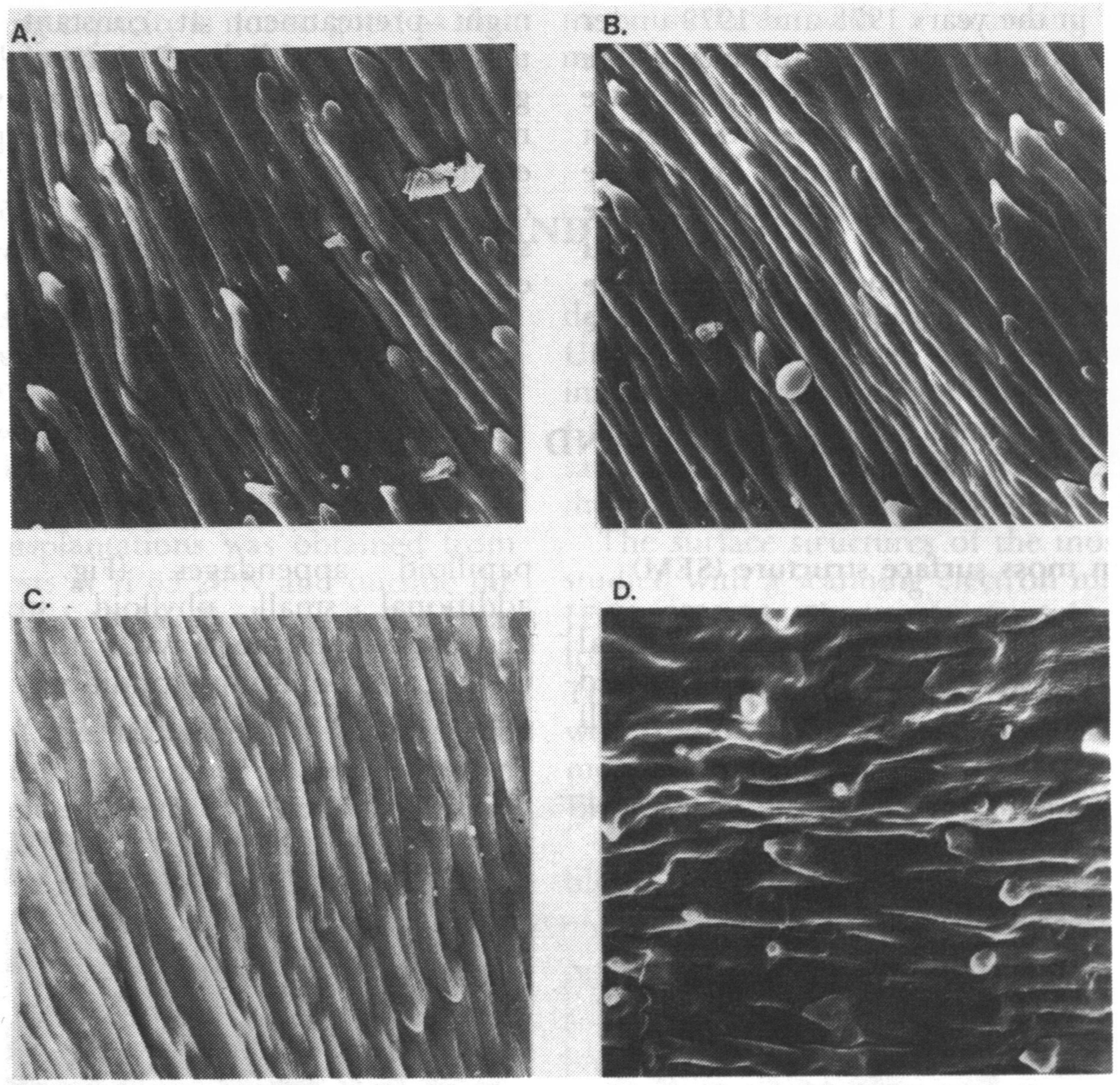

Fig. 4. The surface structure of transplanted *Hylocomium splendens* in control and polluted transplantations 27. 9. 1978 (1000x). a. control transplantation after 14 weeks, b. control transplantation after 62 weeks, c. polluted transplantation after 14 weeks and d. polluted transplantation after 62 weeks.

like surface cell structure, which is also disturbed under air pollution conditions (Figs. 3c and d).

The surface structure of the control transplantations of *Hylocomium splendens* was similar to that of the natural growing mosses (Figs. 4a and b). The polluted transplantations showed changes in their surfae structure after 14 weeks. The surface cells of the moss leaves had lost their turbidity and even their normal morphology (Figs. 4c and d). Later on, after a longer transplantation (62 weeks) period, destruction of the surface cell structure was observed.

Similar destruction of the surface cell structure was also observed in transplanted *Pleurozium schreberi*. The control transplantations were undamaged, but the polluted transplantations showed a destruction of cell morphology and displayed injuries of the surface cells of moss leaves (Figs. 5a, b, c and d).

The surface structure of some bryophytes has earlier been studied by scanning electron microscopy for systematic purposes. The method is easy and convenient for morphological studies (ROBINSON 1971, SARAFIS 1971). Because of the lacking cuticle, the air pollution induced changes in moss leaf surfaces are mainly due to injuries and disturbances in the surface cells. Cell plasmolysis is the probable cause. The $SO_3$– and $SO_4$–iones have been proved to cause plasmolysis on lichens, and acid substances destroy the protein part of the photosynthetic cells, thus causing irreversible changes (RAO and LeBLANC 1966). The changes also develop quite soon after the starting of the



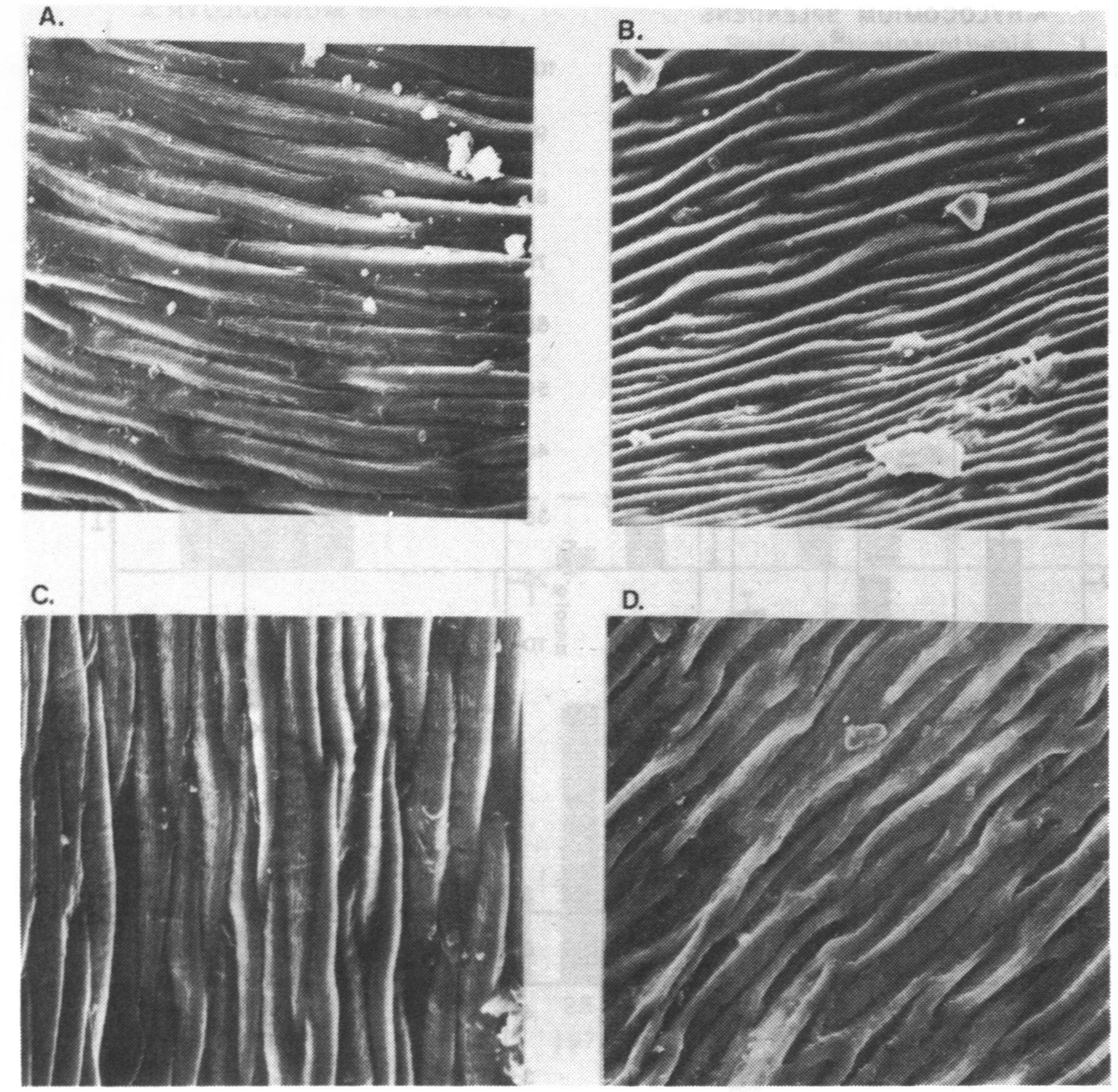

Fig. 5. The surface structure of transplanted *Pleurozium schreberi* in control and polluted transplantations 27. 9. 1978 (1000x). a. control transplantation after 14 weeks, b. control transplantation after 62 weeks, c. polluted transplantation after 14 weeks, d. polluted transplantation after 62 weeks.

influence.

The changes in the moss leaf surface structure must have effects on the microenvironment of the algal microflora on the moss surfaces. The destruction of the leaf structure may decrease the amount of blue-green algae due to unfavourable microenvironmental conditions. These conditions are probably correlated to microhumidity.

### Nitrogenase activity

The *Hylocomium* transplantations into polluted areas had significantly decreased nitrogenase activities as compared with the control transplantations (Fig. 6). The results for *Pleurozium schreberi* were more variable. This may be due to the morphology of *Pleurozium schreberi,* which does not provide an equally good enviroment for blue-green algae as does *Hylocomium splendens.* Generally, the biomass of blue-green algae *(Nostoc* and *Anabaena* -species mostly) on *Hylocomium* seems to be higher than that on *Pleurozium schreberi.* The total amount of blue-green algae on *Hylocomium* is also manifold in the Lappish forests compared with the southern ones (KALLIO and HAUKILAHTI, unpublished results).

In urban areas, combustion, heating, traffic and industry emit nitrogen compounds, which are washed down to plant surfaces by rain water. Other substances, e.g. sulphur compounds, may also have inhibitory effects on nitrogenase activity. The accumulation of sulphur compounds was

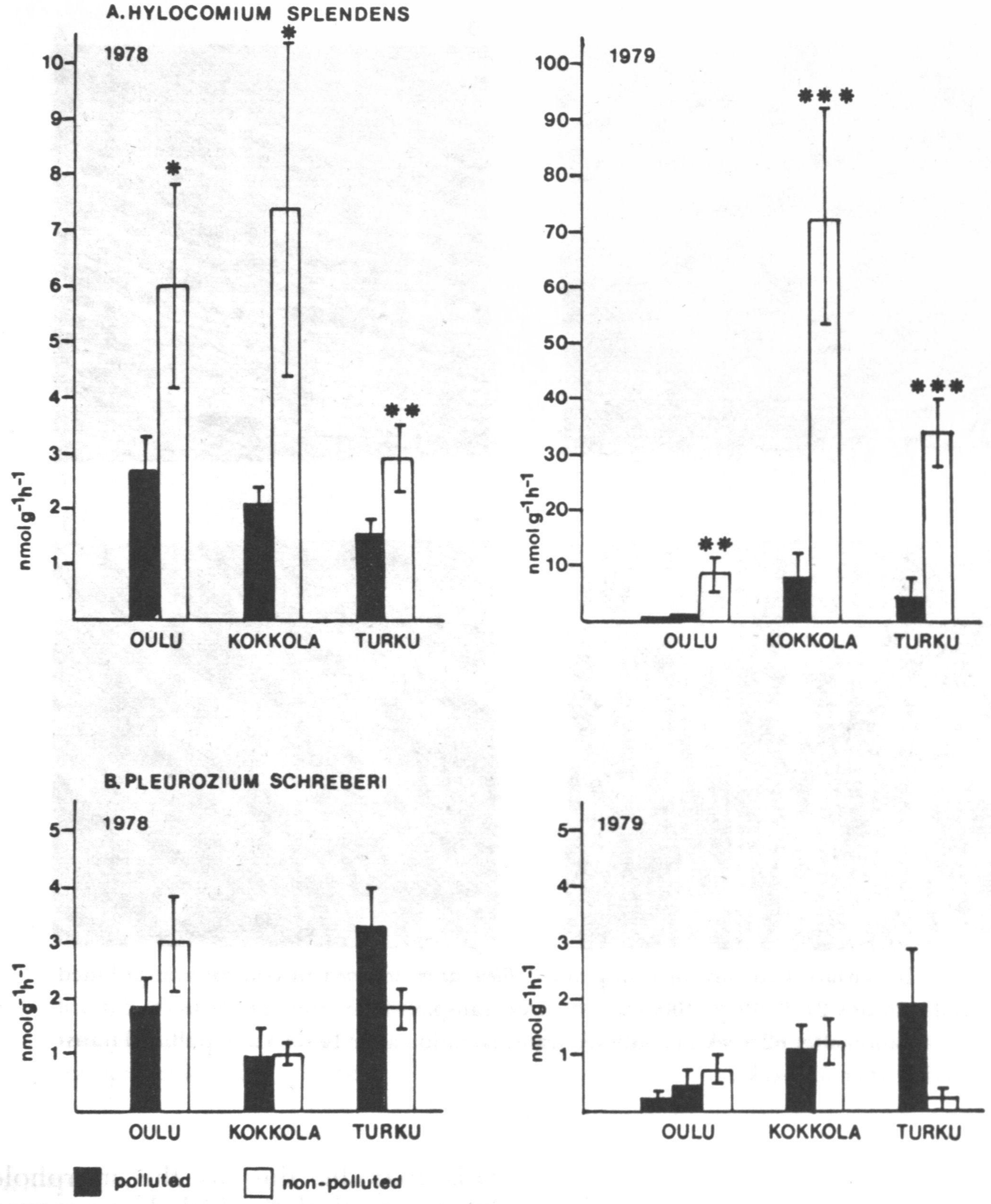


Fig. 6. The nitrogenase activity of transplanted mosses during the summers 1978 and 1979. The different moisture conditions during 1978 and 1979 caused the different nitrogenase activity levels. Each column represents the mean of 25–60 variables; the vertical bars show the standard errors of the means. The levels of significande of the differences between polluted and nonpolluted areas are shown above the columns.

found to be very intensive on moss surfaces in Oulu (HUTTUNEN et al. 1981). The nitrogen compounds in the air of polluted areas may, however, be more inhibitory on the nitrogenase activity of blue-green algae.

Generally, nitrogenase activity is reduced by the addition of combined nitrogen (i.e. STEWART et al. 1975, KALLIO 1976, SHERIDAN 1979). This is an ecological fact, because nitrogen fixing algae lose their capacity to use molecular nitrogen and disappear from the systems where combined nitrogen is supplied.

When reviewing the total level of nitrogenase activity in different years, the effect of transplantation must also be considered. In summer 1978 the transplantations were made with a smaller tussock than in summer 1979.



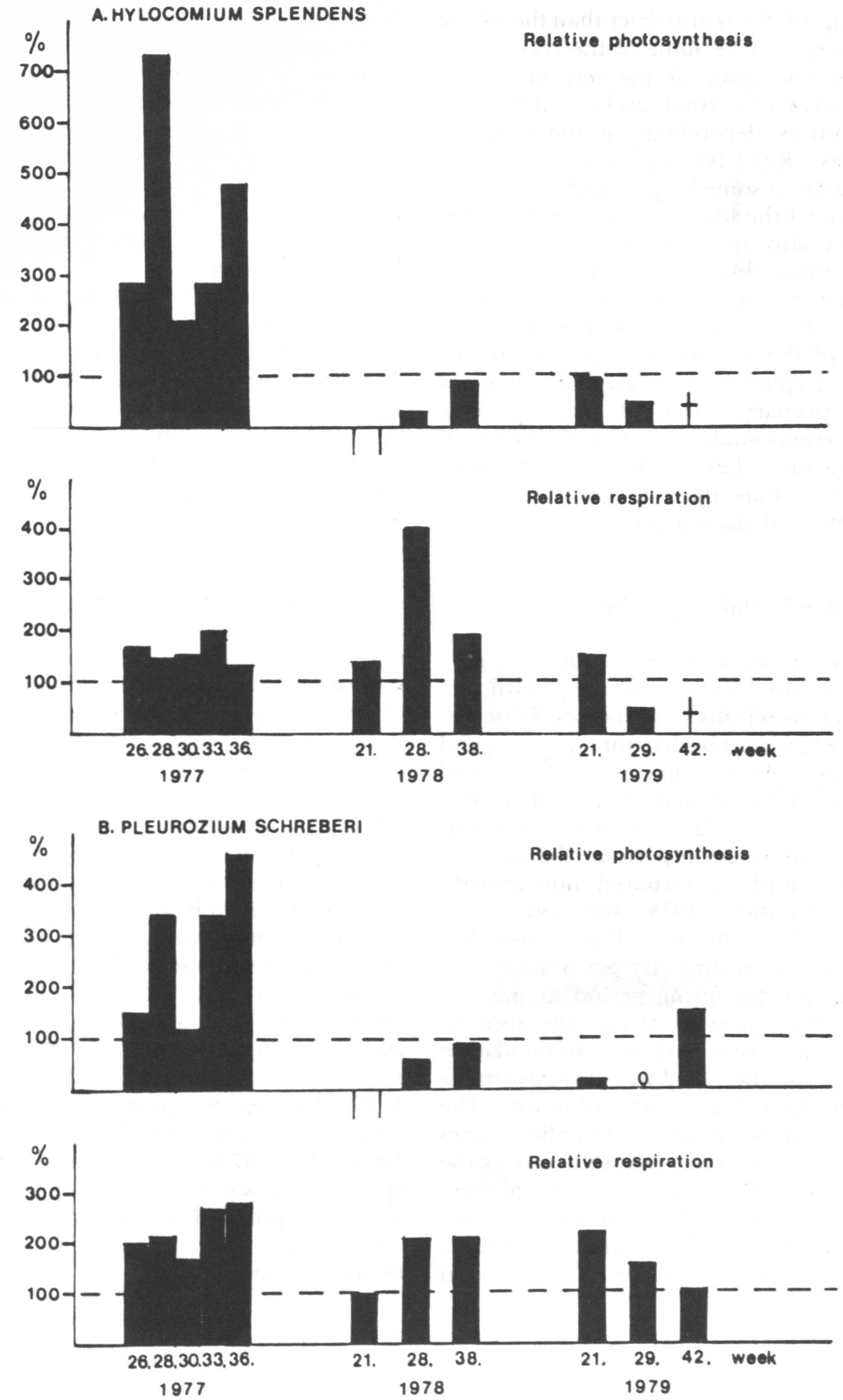


Fig. 7. The relative photosynthesis and respiration of natural growing mosses in 1977, 1978 and 1979.

The spring 1978 was also drier than the spring 1979. These may be some of the reasons for the poor condition of the test mosses in autumn 1978. The total level of nitrogenase activity varies depending on the moisture conditions (KALLIO and KALLIO 1975). The drought of some long periods in summer 1978 affected the life of the blue-green algae and hence also their nitrogenase activity.

The changes observed in the leaf surfaces of mosses are probably related to the observed decrease via the impaired moisture-holding capacity of the mosses. The decrease of the number of epiphytic blue-green algae might be the primary effect of anthropogenic nitrogen compounds. Secondarily, active ion phytotoxicants, heavy metals, and other emitted materials may destroy the micro-environment of these algae.

## Photosynthesis and respiration

Photosynthesis and respiration measurements were used to indicate the growth rate and pollutant sensitivity of mosses. Observations were made on both natural growing and transplanted mosses in air polluted and unpolluted sites around Oulu. Great differences in moss growth were observed during the monitoring period. The summer 1977 was humid and favoured moss growth, but the summers 1978 and 1979 (and especially the spring 1978) were less favourable due to long dry periods.

During the dry spring period in summer 1978, photorespiratory stress was seen in mosses. The heavy sulphur accumulation during the summer 1979 was also observable in the low photosynthetic rate of mosses. The last natural growing mosses on polluted sites exhibited distinct stress respiration during the summer 1978. The last *Hylocomium splendens* stands of the area died during the next summer 1979. The high sulphur accumulation indicates the greater air pollution during 1979 (see further HUTTUNEN et al. 1981).

At first, the relative photosynthetic rate of the transplanted mosses showed the effect of nitrogen compounds most clearly around a fertilizer plant. The longer transplantation over the winter had some inhibitory effects on photosynthesis, especially during the driest period. At the same time the stress respiration was also measured.

The most clearly observed effect of air pollution was the fertilizing effect of nitrogen compounds on both the natural growing and the transplanted mosses. The effect was greater on *Holocomium splendens* than on *Pleurozium*. The stress tolerance and adaptibility of the mosses seemed to be somewhat disturbed under air pollution conditions. The air polluted mosses suffered more drought stress than the control samples (Figs. 7 and 8).

The positive effect of nitrogen fertilization on moss growth has been proved experimentally by KALLIO and HEINONEN (1973). *Hylocomium splendens* has photosynthetic activity mainly in the last two years of growth. *Hylocomium* is very sensitive to environmental changes, because if the newest parts are damaged the whole moss is destroyed (TAMM 1964, CALLAGHAN et al. 1971). In *Pleurozium*, the annual growth cannot be divided, but photosynthesis is mainly concentrated to the top parts of the moss, about 2 cm from the tip (BATES 1979). The most important factors affecting the growth are temperature, light and air-borne nutrients (CALLAGHAN et al. 1978, BATES 1979, LONGTON and GREENE 1979). The extreme drought decreases the photosynthesis of both species (KELLOMÄKI and HARI 1976). The recovery after drought is delayed after longer dry periods (HINSHIRI and PROCTOR 1971). The extreme dry period in spring 1978 was observed to affect particularly the polluted moss samples and the recovery was slower in polluted samples than in control samples.



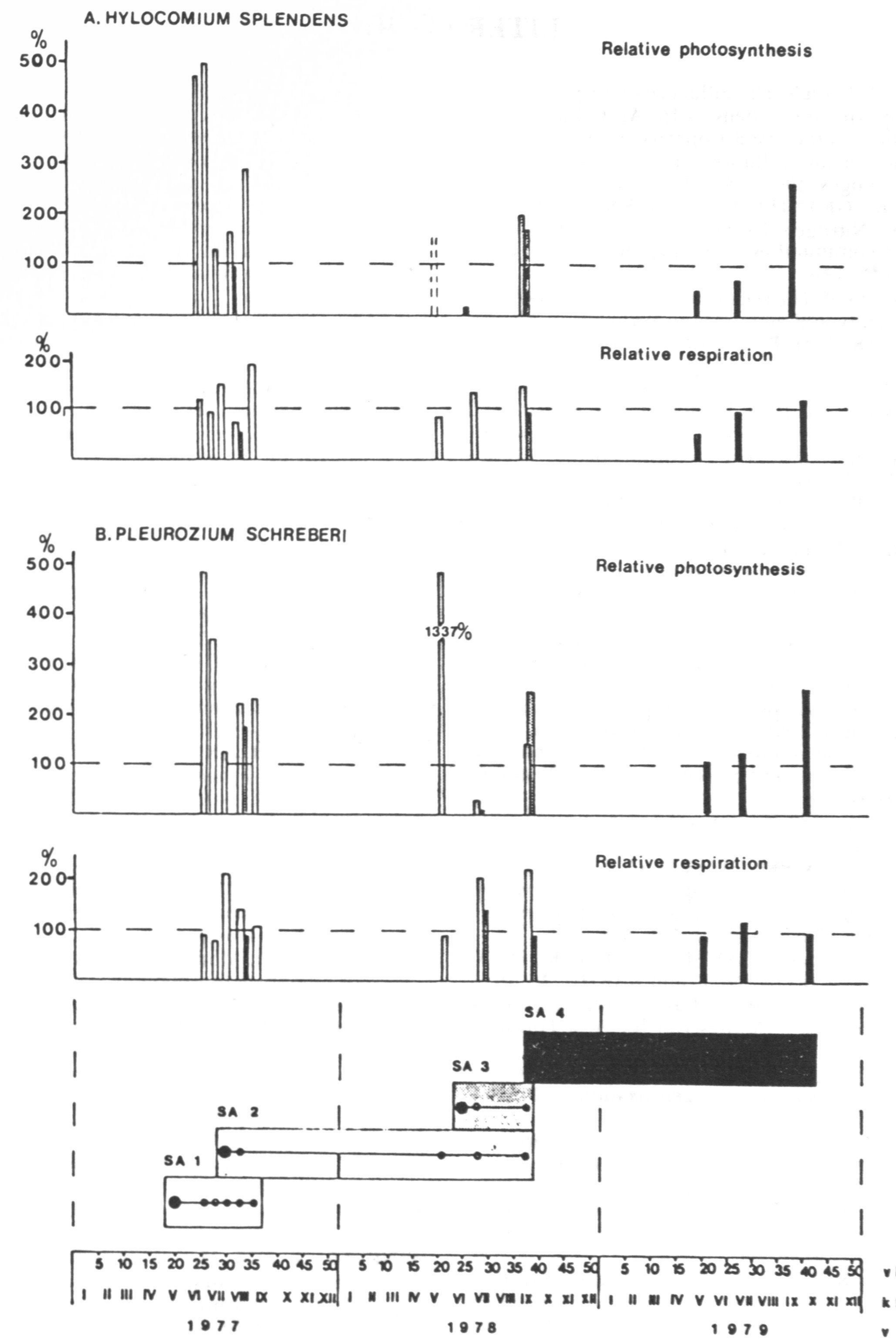


Fig. 8. The relative photosynthesis and respiration of transplanted mosses in 1977, 1978 and 1979. The transplantation time is presented at the horizontal blocks (SA=-transplantation time).

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**LUUKKANEN, OLAVI** O.D.C. 232.311.3 : 181.521
*Pinus sylvestris*

1981. Effects of gibberellins $GA_4$ and $GA_7$ on flowering in Scots pine grafts. Seloste: Havaintoja gibberelliinien $GA_4$ ja $GA_7$ vaikutuksesta männynvartteiden kukintaan. – **Silva Fennica** Vol. 15 (4):359–365. Helsinki.

Ethanolic sprays of $GA_4$ or $GA_7$ on 9-year-old Scots pine grafts, repeated four times during the shoot elongation period, resulted in a statistically significant increase in female flowering one year after the treatment. Of the two compounds, $GA_4$ seemed to be somewhat more efficient, yielding 47 female strobili/100 shoots *vs.* 36 and 6 strobili/100 shoots in $GA_7$ and control treatments respectively. The mixture $GA_{4/7}$ was also applied and seemed to have an effect intermediary to those of the purified compounds. However, due to the limited amount of material, none of the differences between the gibberellins could be statistically confirmed. Male flowering frequencies were also too low to allow any firm conclusions, but the numerical results suggested that the purified gibberellins may promote male and female flowering in different ways.

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**JALKANEN,[1] R., HUTTUNEN,[2] S. & VÄISÄNEN,[2] S.** O.D.C. 443.3:164.5

1981. The wax structure of the developing needles of *Pinus sylvestris* progenies infected by *Lophodermella sulcigena*. Seloste: Nuorten neulasten vaharakenne harmaakaristeisissa mäntyjälkeläistöissä. – **Silva Fennica** Vol. 15 (4):377–381. Helsinki.

The development of the stomatal area wax structure of pine needles was studied in flushing needles with a scanning electron microscope. The needles were obtained from eleven Finnish plus pine *(Pinus sylvestris L.)* progenies. The needles were taken from nearly unifected and heavily infected trees. No differencs in the early developmental stages of stomatal wax structure were observed between the southern Finnish, central Finnish and northern Finnish progenies. The general structure differed in the stomatal cavity chamber size. The stomatal openings were larger in heavily infected trees than in healthy trees. This might have an influence on the mechanical penetration of the fungal hyphae.

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**SAARILAHTI, MARTTI** O.D.C. 383.2

1981. Mikroaaltotutka jään paksuuden mittaukseen. Summary: Radar measurement of ice thickness on ice roads and landings. – **Silva Fennica** Vol. 15 (4): 366–376. Helsinki.

Light and economic FM-CW radar has been developed for controlling the bearing capacity of ice roads and landings. It was tested on natural ice and ice roads of 3 lakes and 1 sea. Ice thickness varied from 0 to 100 cm and the total number of observations was 166. Correlation coefficient between auger and radar measured ice thickness was $r = 0.99$ and absolute error varied from $-10$ to $+7$ cm. The quality of ice had no influence, but by improving the interpretation of results also the quality factors might be detectable.

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**HUTTUNEN, SATU (ed.)** O.D.C. 424

1981. Proceedings of the symposium: "Air pollutants as additional stress factors under northern conditions". November 11–13, 1980, Oulu, Finland. Ilman epäpuhtaudet kasvien stressitekijöinä pohjoisessa. Oulussa 11–13. 11. 1980 pidetyn symposiumin esitelmät. – **Silva Fennica** Vol. 15 (4):382–504. Helsinki.

The symposium of "Air pollutants as additional stress factors under northern conditions" was held in Oulu during November 11–13, 1980. The symposium had four sessions, one on the ecophysiology of air pollution effects and long-term risks of air pollution effects, one on bioindication in forest ecosystems, one on particle and metal problems, and a general poster session mainly related to current research and the results of the research programme on the dispersion and effects of air pollutants in forest environments financed by the Academy of Finland. Most of the posters and papers presented at the symposium have been included in the proceedings. The rest of the results presented are reviewed in research report of the programme and in related scientific papers.

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# KIRJOITUSTEN LAATIMISOHJEET

Silva Fennica-sarjassa julkaistaan lyhyitä metsätieteellisiä tutkimuksia ja kirjoituksia kotimaisilla kielillä tai jollakin suurella tieteellisellä kielellä. Julkaistavaksi tarkoitettu käsikirjoitus on jätettävä Seuran sihteerille painatuskelpoisessa asussa. Seuran hallitus ratkaisee asiantuntijoita kuultuaan, hyväksytäänkö kirjoitus painettavaksi.

Kirjoitusten laadinnassa noudatetaan Silva Fennican numerossa Vol. 4, 1970, N:o 3 painettuja kansainvälisiä ohjeita. Suureissa, yksiköissä sekä symbolien ja kaavojen merkinnöissä noudatetaan ohjeita, jotka ovat suomalaisissa standardeissa SFS 2300, 3100 ja 3101. Oikoluvussa noudatetaan standardia SFS 2324.

Kirjoituksen alkuun tulee julkaisun kielellä lyhyt yhdistelmä tutkimuksen tuloksista. Samoin laaditaan tutkimuksen yhteyteen lyhyt englanninkielinen tiivistelmä, jonka lisäksi kunkin Silvan numeron loppuun painetaan irti leikattavan kortin muotoon kustakin tutkimuksesta englanninkielinen esittely. Sisällysluetteloa ei käytetä. Mahdolliset kiitokset esitetään lyhyesti johdannon lopussa ja merkitään painettavaksi petiitillä.

Kuvien ja piirrosten viivapaksuudet ja tekstikoko on valittava siten, että ne sallivat painatuksen vaatiman pienennyksen. Kuvien ja piirrosten painatuskoosta on syytä neuvotella etukäteen toimittajan kanssa, sillä tarpeettomia kustannuksia aiheuttavaa painatuskokoa ei sallita. Valokuvien tulee olla teknisesti moitteettomia ja kiiltävälle valkealle paperille suurennettuja. Värikuvia ei yleensä hyväksytä painettavaksi. Kuvat ja taulukot numeroidaan kummatkin erikseen juoksevasti, ja niiden otsikoista laaditaan erillinen luettelo kirjapainoa varten.

Jos vieraskielisessä lyhennelmässä viitataan tiettyihin kuviin ja taulukoihin, on nämä varustettava vieraskielisin otsikoin ja selityksin. Muut kuvat ja taulukot voivat olla yksikielisiä.

Lähdeviittauksissa tekijännimet sijapäätteineen kirjoitetaan isoin kirjaimin mikäli tekijännimen vartalo on muuttunut. Muutoin taivutuspääte kirjoitetaan pienaakkosin. Esimerkkejä: KOSKISEN (1972) tutkimus . . ., YLI-VAKKURIn (1972) tutkimus . . . Milloin tekijöitä on kolme tai useampia, mainitaan tekstissä vain ensimmäinen (esim. HEIKURAINEN ym. 1961). Vieraskielisessä tekstissä ym. korvataan merkinnällä et al. Jos julkaisulla on kaksi tekijää viitteessä, pannaan tekijöiden nimien väliin ja-sana painatuskielellä. Esimerkki: KELTIKANGAS ja SEPPÄLÄ (1973, s. 222) osoittivat . . .

Viitekirjallisuus luetteloidaan tekijännimien (kirjoitetaan isoin kirjaimin) mukaisessa aakkosjärjestyksessä. Jos tekijöitä on useampia, nimet erotetaan pilkulla, paitsi kaksi viimeistä jotka erotetaan &-merkillä. Tekijän etunimistä suositellaan käytettäväksi vain alkukirjaimia. Tutkimusten nimet kirjoitetaan lyhentämättä. Julkaisusarjoista käytetään niitä lyhenteitä, jotka on painettu Silva Fennican numerossa Vol. 5, 1971, N:o 2. Täydellisempi luettelo on nähtävissä Seuran toimistossa. Kirjoituksen löytämisen helpottamiseksi mainitaan aikakauslehdistä myös sivunumerot. Suomenkielisistä tutkimuksista otetaan mukaan vieraskielisen lyhennelmän nimi. Volyymi merkitään julkaisusarjan nimen jälkeen. Jos kyseessä on aikakauslehti tai vastaava, numero merkitään volyymin jälkeen suluissa. Sivunumerot erotetaan kaksoispisteellä volyymistä tai suluissa olevasta numerosta. Jos samalla kertaa ilmestynyt volyymi sisältää useita tutkimuksia, merkinnässä sovelletaan ko. julkaisussa noudatettua tapaa. Esimerkkejä:

ILVESSALO. Y. 1952. Metsikön kasvun ja poistuman välisestä suhteesta. Summary: On the relation between growth and removal in forest stands. – Commun. Inst. For. Fenn. 40.1.

WILCOX, W. W., PONG, W. Y. & PARMETER, J. R. 1972. Effects of mistletoe and other defects on lumber guality in white fir. Wood & Fiber 4 (4): 272–277.

Englanninkielisen lyhennelmän ja mahdollisten kuva- ja taulukkotekstien kääntämisestä ja pätevän kieliasiantuntijan tekemästä tarkastamisesta huolehtii kirjoittaja. Seura voi maksaa kustannukset valtiovarainministeriön antamien ohjeiden mukaan. Jos kääntäjän lasku on ohjeiden edellyttämää tasoa korkeampi, kirjoittaja vastaa ylittävästä osuudesta.

Lähempiä tietoja antaa Seuran julkaisujen toimittaja.

# KANNATTAJAJÄSENET – UNDERSTÖDANDE MEDLEMMAR

CENTRALSKOGSNÄMNDEN SKOGSKULTUR
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OY KAUKAS AB
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KYMI KYMMENE
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MAATALOUSTUOTTAJAIN KESKUSLIITTO
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