

Investigations on Post-fire Discomycetes: *Geopyxis rehmii* sp. nov.
and *G. carbonaria* (Alb. & Schw. ex Fr.) Sacc.

by

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With 7 figures and 2 plates

Abstract: A new species, *Geopyxis rehmii* Turnau, from burns on Stare Wierchy (The Gorce Range, Western Carpathians), is described and illustrated. It is compared with *G. carbonaria* (Alb. & Schw. ex Fr.) Sacc. which is also briefly characterized.

Introduction

In the period 1979-1981 investigations on the post-fire cup-fungi flora of Turbacz and Stare Wierchy in the Gorce Mts. (Turnau 1984a and b) were carried out. Particular attention was paid to species typical or the consecutive post-fire successional stage. *Geopyxis carbonaria* was one of the most common fungi in the region. Several hundred specimens of this species fruited in an area of 1 m². Regular quantitative and qualitative samplings made it possible to observe differences within populations of species known to occur and to distinguish a new taxon — *Geopyxis rehmii* sp. nov.

Methods of Investigations

In the investigation 10 burned-over areas situated in close proximity on the northern slopes of Stare Wierchy were chosen. They were visited fortnightly between 1979 and 1980. Occasional studies were also carried out in 1981. Observations for several years made it possible to establish the sequence in which fruit-bodies appeared in the first, second, and third years after burning. On each occasion the number of fruit-bodies of each species was recorded. Distance of fruit-bodies from the burn centre were recorded. Fruit-bodies were also measured and fragments for microscopic examination were collected.

Fruit-bodies were photographed using "Practica" camera. Spores were examined under both a light and scanning electron microscope. Spore preparation is described by Turnau (1984a).

To determine the heterogeneity of populations methods of statistical analysis and numerical classification were used. Sixteen fruit-bodies from both populations,

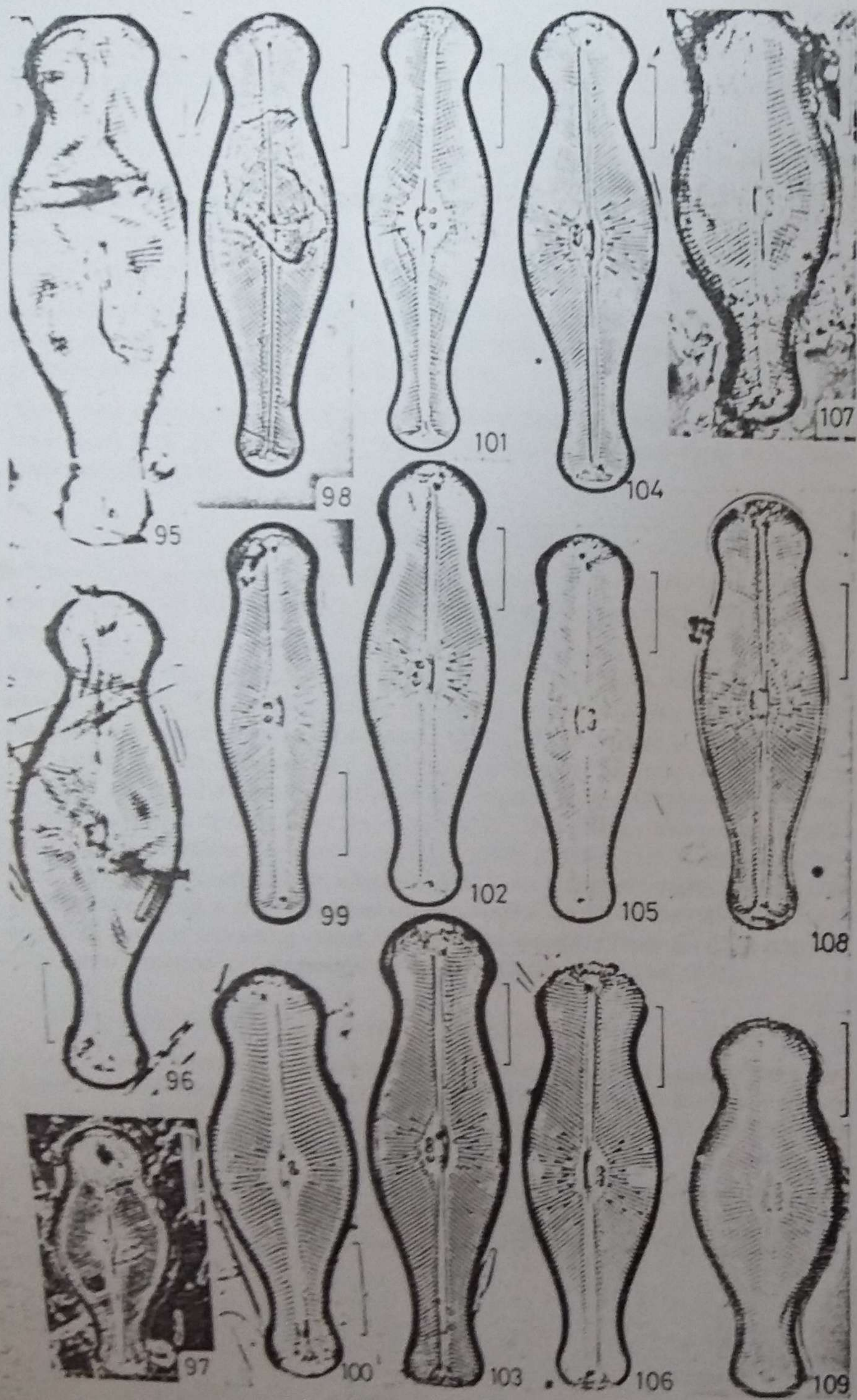


Plate VII. Figures 95-109. Figures 95-96. Other examples of valves from North Wales. Figures 97-109. Examples of valves from Mid Wales (River Wye, author's Collection). Scale 20 µm.

collected simultaneously from 3 burns on Stare Wierchy were analyzed. In the analysis a number of different qualitative and quantitative characters were initially chosen.

To determine which quantitative characters clearly distinguished the populations variational analysis was used. The results of this analysis were tested at the 5% significance level (Oktaba 1977). After numerous calculations finally 4 quantitative characters were decided upon. These include length of spores, length of asci, diameter of apothecia, height of apothecial cup and length of the stalk.

Six qualitative characters were selected: shape of apothecia, colour, presence of a lighter margin of the fruit-body, presence of a stalk, ornamentation of spores, and position of fruit-body relative to the burn centre.

These four quantitative and six qualitative characters were used in the numerical classification of the 32 specimens. Sums of differences between them were calculated according to the formula:

$$d_{ij} = \sum_{k=1}^n x_{ik} - x_{jk}$$

where n is the number of characters, x_{ik} the value of the k -qualitative or quantitative character for the specimen i , x_{jk} , and value of the k -character for the specimen j .

For grouping, the unweighted pair group method using arithmetic averages was applied (Sneath, Sokal 1973). Before the numerical classification was performed, values of the quantitative characters were modified in such a way so that all characters would equally influence the calculated differences between individuals. In choosing a method of transformation it was important to recognize that with the simultaneous use of qualitative and quantitative characters, differences between values of quantitative characters would not differ greatly from numerical differences for qualitative characters, which are 0 or 1. As proposed by Gower (1971) the maximum value of a character after transformation equals 1, the minimum value equals 0, and the remaining values are fractional numbers from the (0,1) interval. This transformation is characterized by the formula (Sneath, Sokal 1973):

$$x = (x - x_{\min}) / (x'_{\max} - x'_{\min})$$

where x - is the character value after transformation, x'_{\max} and x'_{\min} are the maximum and minimum value of a given character before the transformation. It should be stressed, that the qualitative and quantitative characters, jointly considered can to a degree influence the result.

In the investigations of *Geopyxis*, chemotaxonomical methods were also applied. Dyes were analysed; fruit-bodies were put into acetone for 12 hours, the material was then homogenized, the dyes were absorbed in naphtol ether and the absorption spectra were examined.

Results

Regular observations of the flora of burns on Stare Wierchy has revealed the occurrence of two different species of *Geopyxis*. The first, *Geopyxis carbonaria* (Alb. & Schw. ex Fr.) Sacc., is one of the most common species of burns in the whole Europe, but the second, *G. rehmii* sp. nov., was formerly not distinguished because of co-occurrence with *G. carbonaria* and considerable resemblance to it. Detailed analysis, however, has revealed significant differences between these species; and it is now considered necessary to distinguish two taxa.

The differences are related to the following features:

- morphology, colour and size of fruit-bodies, asci and spores;
- phenology, ecological demands relative to the position of fruit-bodies in relation to the burn centre;
- geographical distribution in Poland.

a. Because fruit-bodies of both taxa occurred at the same time on the same burns, close investigations were carried out. To determine differences in microscopic features several hundred specimens of both species were collected and examined. Length and width of spores and asci, diameter and height of apothecium, as well as thickness and height of stalk were measured. For comparison, identical measurements were carried out on herbarium materials from a dozen more additional localities in Poland. Correspondence of measurements for both taxa on fresh material from Stare Wierchy, the use of fruit-bodies collected in the Gorce Mts. Both were recognized as fully representative and sufficient for further analysis namely variational analysis, the Duncan test and dendrogram.

On the basis of variational analysis two characters were selected for the investigations: length of spores (Fig. 1a) and length of asci (Fig. 1b). These pointed to the existence of two statistically different populations. In both cases the range of variability of asci and spore sizes is much broader for the fruit-bodies of *Geopyxis rehmii*. Diameter of apothecia and height without stalks, as well as stalk length were taken into account. In Fig. 2 the relationship between apothecium diameter and stalk length is presented. Fruit-bodies of *G. carbonaria* have consistently smaller apothecia and longer stalks, while those of *G. rehmii* are larger and the stalk is either underdeveloped or totally lacking.

The quantitative characters discussed above as well as six qualitative features were the basis for the numerical classification of fruit-bodies of both species. A dendrogram (Fig. 3), confirming the differences between the two populations, was obtained.

Since fruit-bodies of both groups differed in colour, their dyes were examined. Absorption spectra confirmed that colours originate from the same dye with a spectrum resembling that typical of lykopen — a carotene dye (Fig. 4). Colour differences probably result from different concentration of this dye. The main differences between the two species are contrasted (Tab. 1).

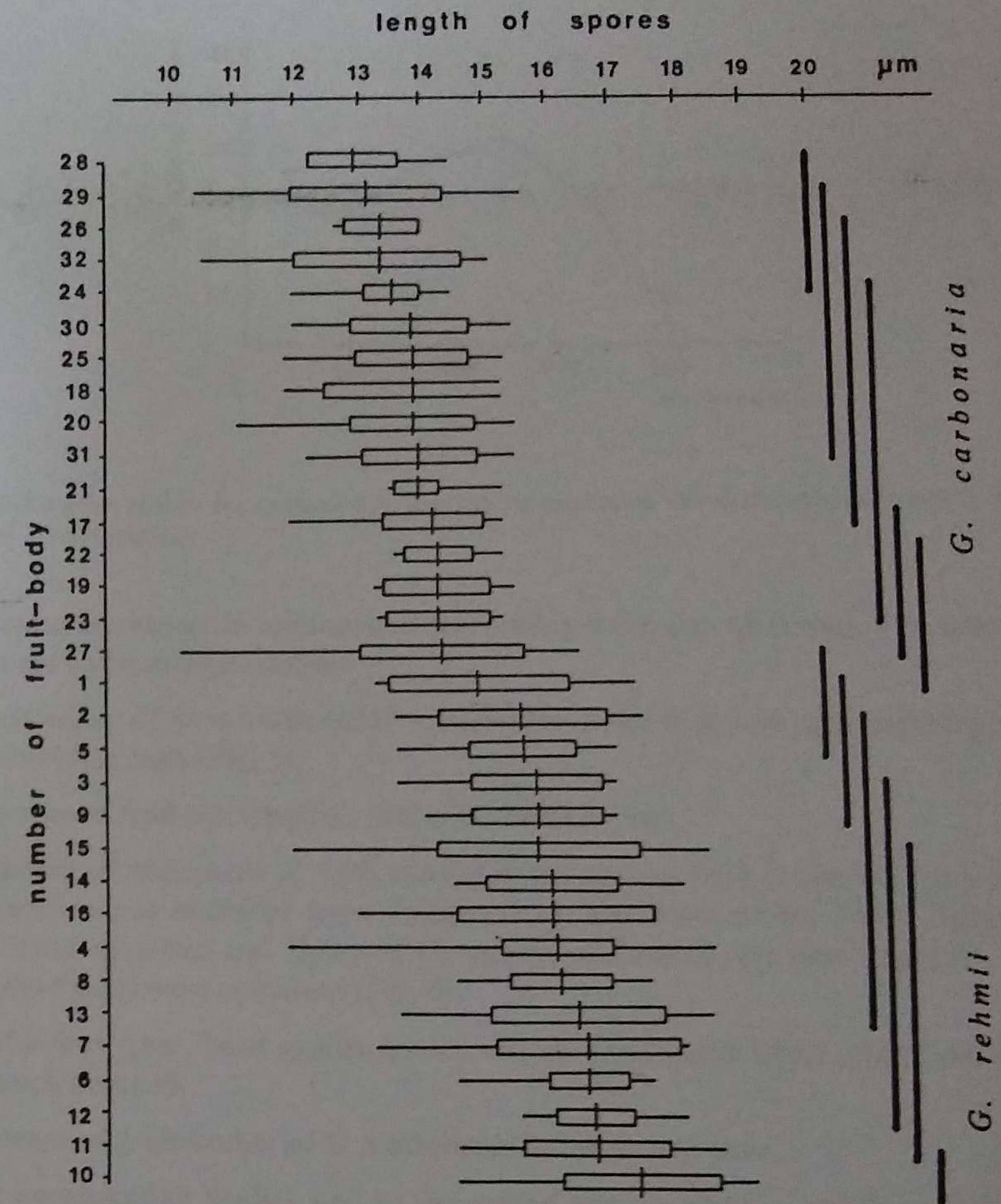
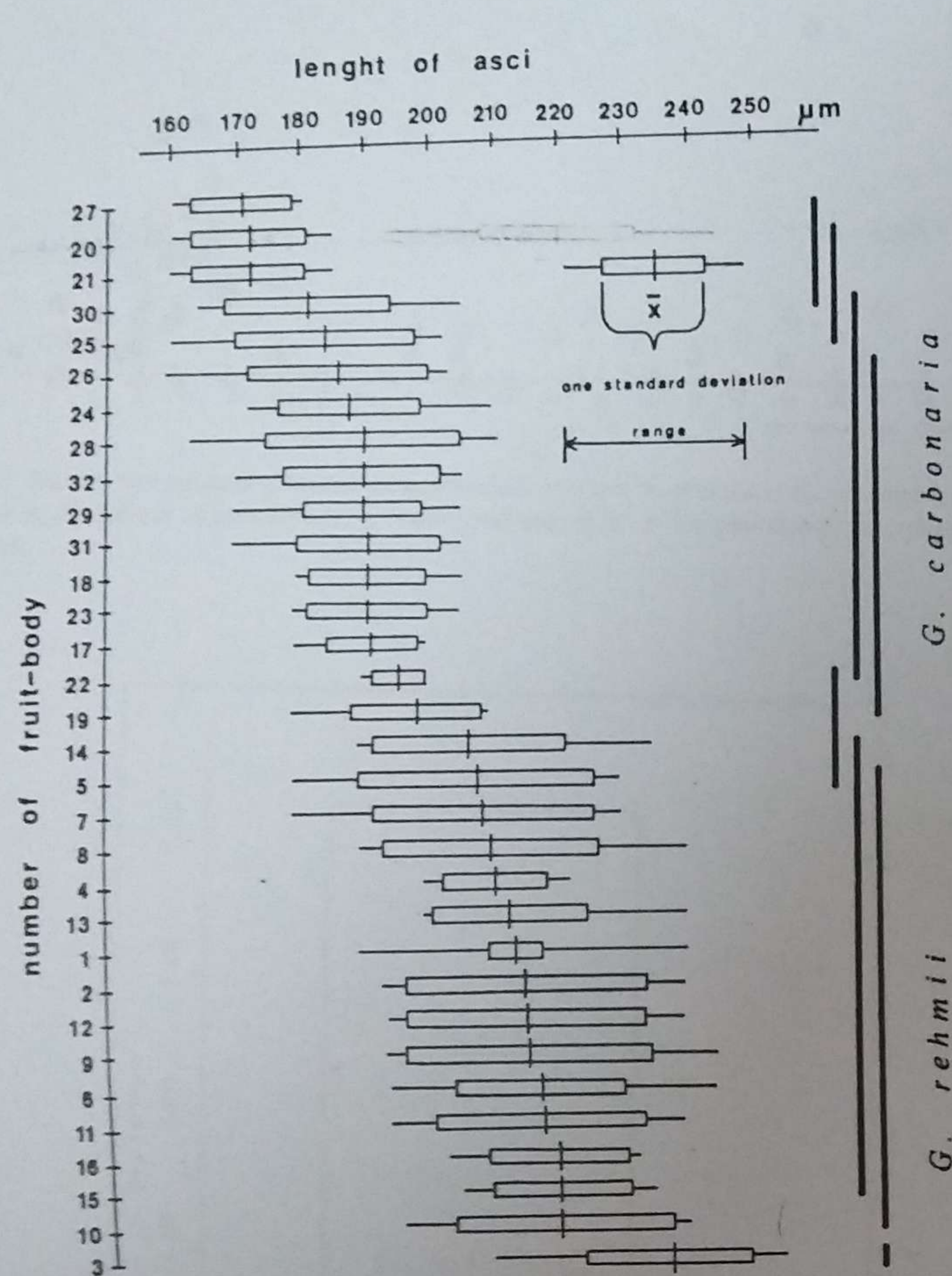


Fig. 1. Variational analysis and Duncan test: a - length of spores in *Geopyxis carbonaria* and *G. rehmsii*, b - length of asci in *G. carbonaria* and *G. rehmsii*.

Tab. 1. Differences between *G. carbonaria* and *G. rehmsii*.

character	<i>Geopyxis carbonaria</i>	<i>Geopyxis rehmsii</i>
diameter of apothecium	2-8 mm	2-30 mm
colour of apothecium	ochre-and brown or ochre-and-orange	orange or orange-and-red
margin of apothecium	visibly lighter, edge crenated	colour uniform, edge smooth
length of stalk	up to 15 mm	up to 1,5 mm
length of spores	10-16 μm	10-19 μm
length of asci	160-210 μm	180-250 μm
spore ornamentation (SEM)	smooth or slightly undulated (Pl. I)	visibly warded (Pl. II)

b. Fruit-bodies of *Geopyxis carbonaria* (Pl. I) occurred in mass on relatively dry but shaded burns, together with *Peziza praetervisa*, *Trichophaea hemisphaerioides*, *Marchantia polymorpha*, *Bryum argenteum*, *Funaria hygrometica*. *G. carbonaria*, fruiting in the company of these species strong predominated such was frequently recorded on wet burns, but there abundance of fruit-bodies was visibly less than that of *Scutellina scutellata* and *Ascobolus carbonarius*.

Fruit-bodies of *G. rehmsii* (Pl. II) grew in greatest numbers on dry burns, fully exposed to the sun, where the fruit-bodies of *G. carbonaria* were found much less frequently. This emphasizes the differences between the range of ecological tolerance for humidity.

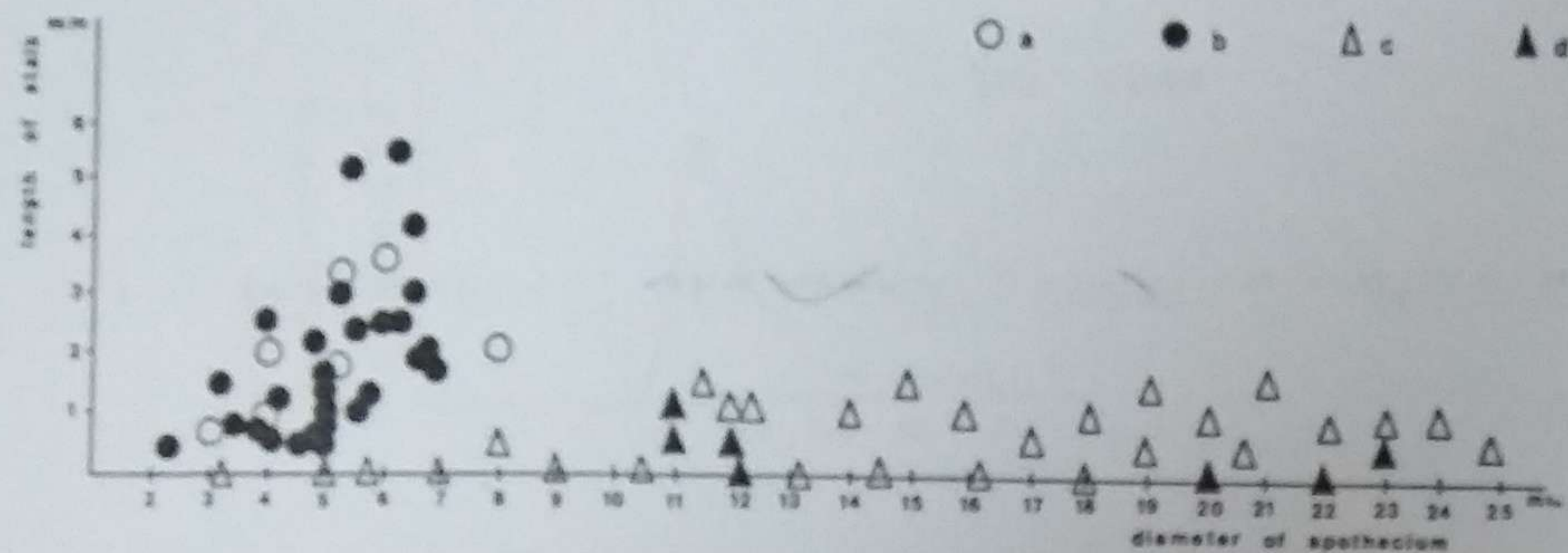


Fig. 2. Dependence between diameter of apothecium and length of stalk in *G. carbonaria* (a, b) and *G. rehmii* (c, d). Colour of apothecium: a - ochre and brown, b - ochre and orange, c - orange, d - orange and red.

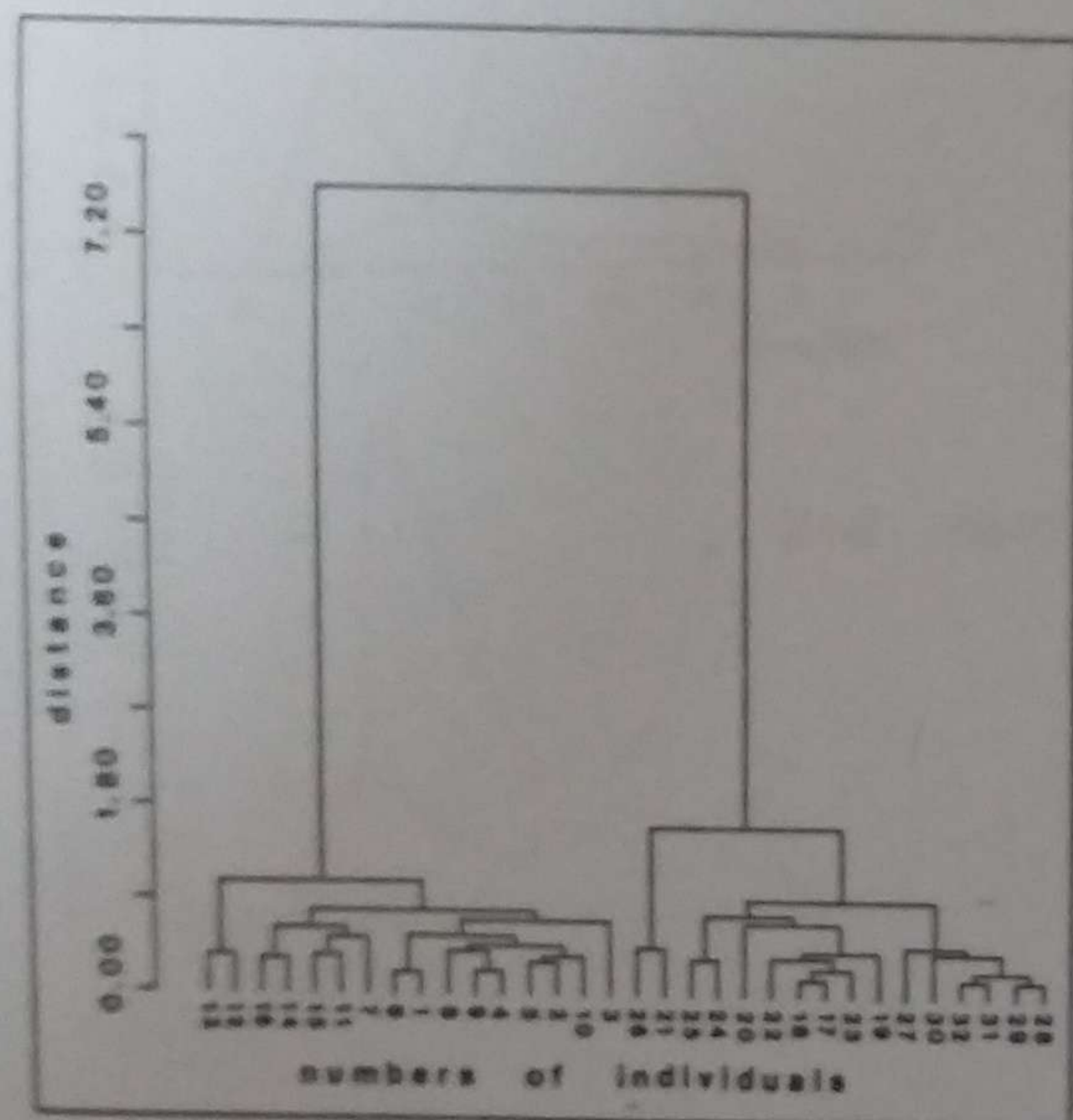


Fig. 3. Dendrogram showing the differences between populations of *G. carbonaria* (a) and *G. rehmii* (b) and similarity of fruit bodies within these populations.

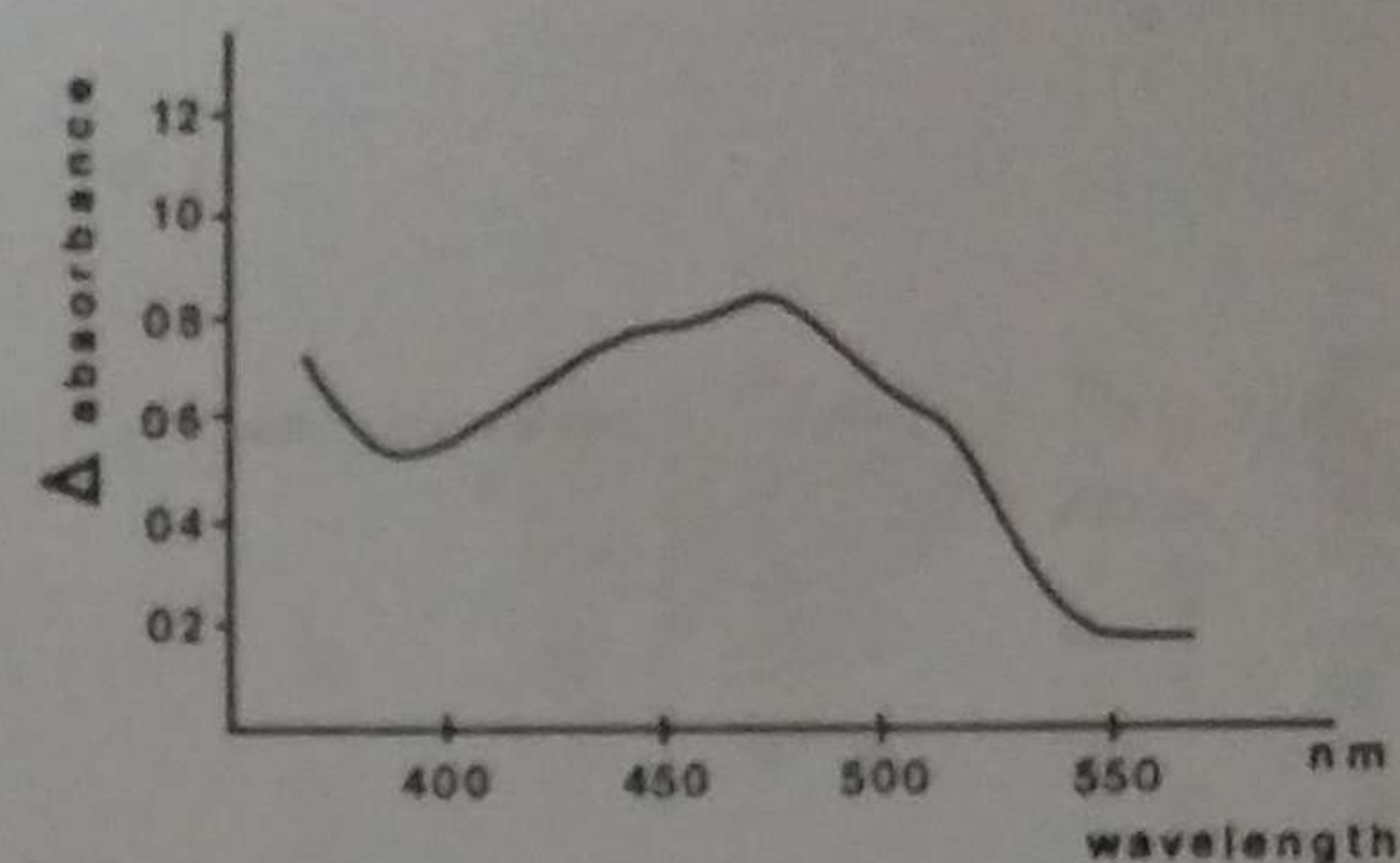


Fig. 4. Absorption spectra for carotene dyes dissolved in naphthol in *Geopyxis carbonaria* and *G. rehmii* (identical in both species).

Differences in respect to tolerance of soil acidity were also observed. The following factors are supporting evidence.

1. Appearance of fruit-bodies of *G. carbonaria* in the first year after burning, when the soil pH was high (Fig. 5).
2. Absence of fruit-bodies of *G. rehmii* in the first year.
3. Presence of specimens of both species in the second year, however, fruit-bodies of *G. carbonaria* occurred more frequently in the burn centre where higher pH values were recorded and those of *G. rehmii* exclusively on burn margins where lower pH-values were measured (Fig. 6).
4. In the third year, both species fruited on the whole burn when central pH values were much reduced.
5. Absence of fruit-bodies of *G. carbonaria* in the fourth year.

c. For comparative studies and to determine the distribution of these species in Poland herbarium specimens from the following Universities were examined: Institute of Botany, University of Warsaw (UW), Institute of Botany, University of Lublin (UMCS), Institute of Biology, University of Poznań (UAM), Institute of Botany, University of Wrocław (UW), Department of Pharmacy of the Medical Academy (collection of Prof. A. Nespiak), and Institute of Botany, Jagellonian University (UJ), Institute of Botany of the Polish Academy of Sciences.

All specimens in the above herbaria had been identified as *Geopyxis carbonaria* (Alb. & Schw. ex Fr.) Sacc., but after detailed examination, fruiting bodies from 5 localities were identified as *G. rehmii*. These included Babia Góra Mt., 815 m above sea-level, 13.05.1974, lg. A. Bujakiewicz; Świątokrzyskie Mts. National Park, 30.09.1966, lg. M. Lisiewska; Ojców National Park, 13.07.1961, lg. W. Wojewoda; Bukowina Obidowska, Western Carpathians, 14.07.1966, lg. W. Wojewoda; Tatras Mts., Zakopane, 20.05.1909, lg. K. Rouppert. Localities of occurrence of *G. carbonaria* are found throughout Poland, whereas those of *G. rehmii* are visibly restricted to mountain areas (fig. 7).

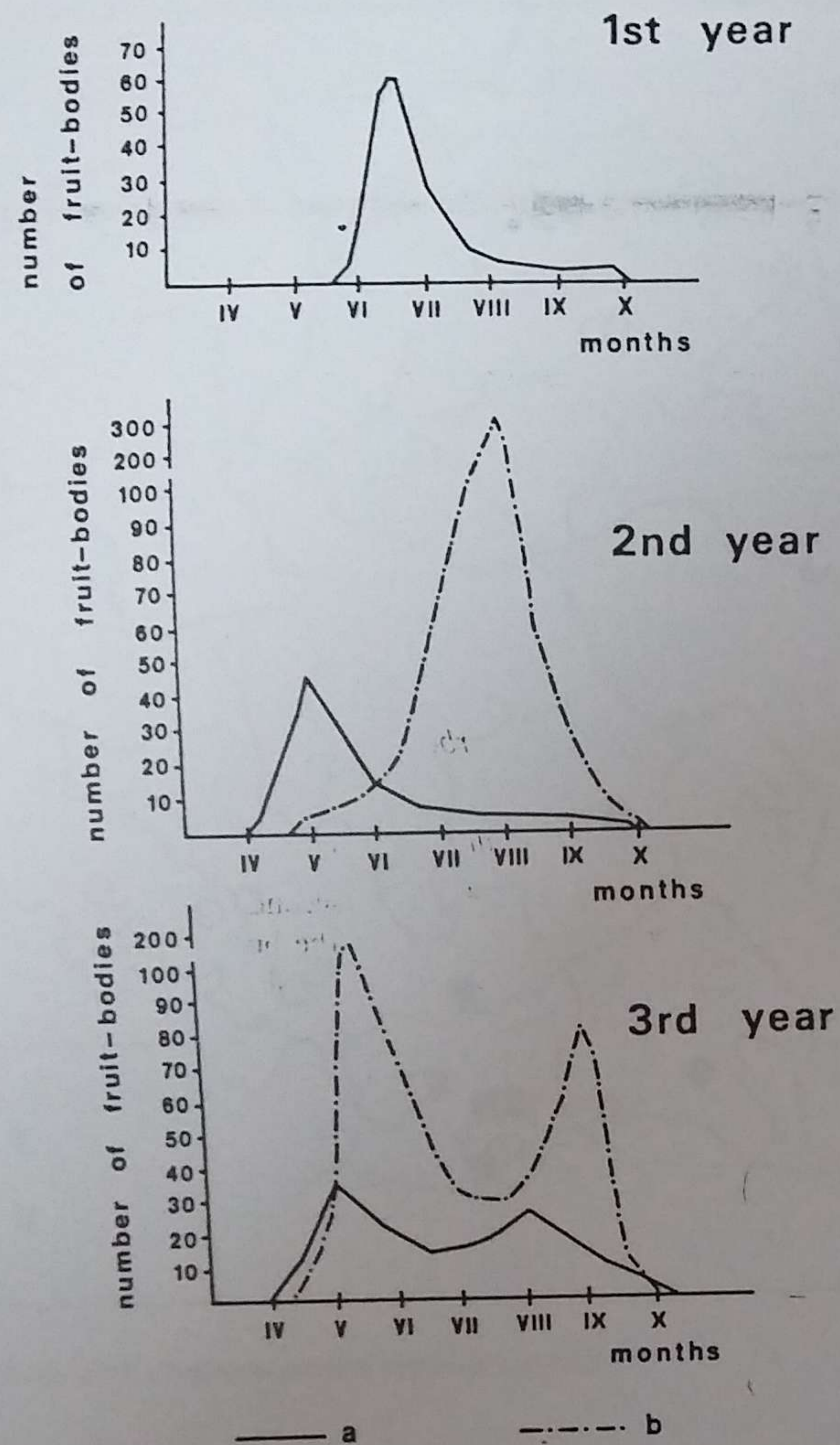


Fig. 5. Periodicity of fructification of *G. carbonaria* (a) and *G. rehmii* (b) on burns on Stare Wierchy for three consecutive years after the burning.

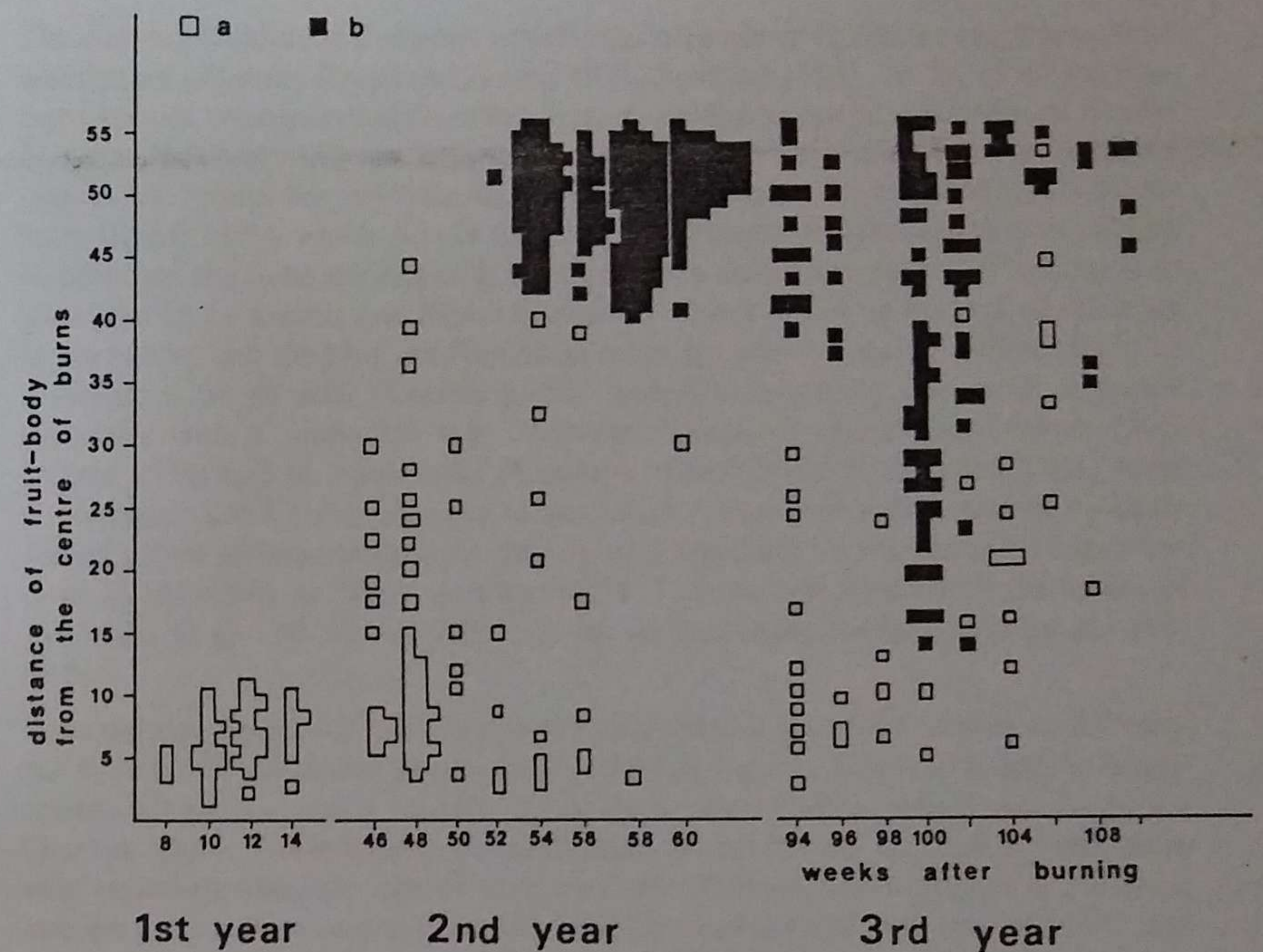


Fig. 6. Distribution of fruiting bodies of *G. carbonaria* (a) and *G. rehmii* (b). Data from 3 burns recorded at two-week-intervals for 3 years.

The Diagnosis

In the literature references to the great variability of fruit-bodies of *Geopyxis carbonaria* can be found, Moser (1949) described fruiting bodies which differed considerably in size from those of typical specimens, with diameters exceeding 3 cm. Rehm (1896) distinguished a form "major", characterized by orange fruit-bodies, much larger than those of the typical form. It is not possible to resolve the question of whether the major form described by Rehm corresponds to those specimens now distinguished as *G. rehmii*. The author was unable to locate Rehm's specimens from the Tatra Mts., and the description of the form itself is insufficient to acknowledge synonymy with *G. rehmii*.

Below the Latin diagnosis for the species is presented. It is named to honour of the German natural historian Dr. Heinrich Rehm (1829-1916).

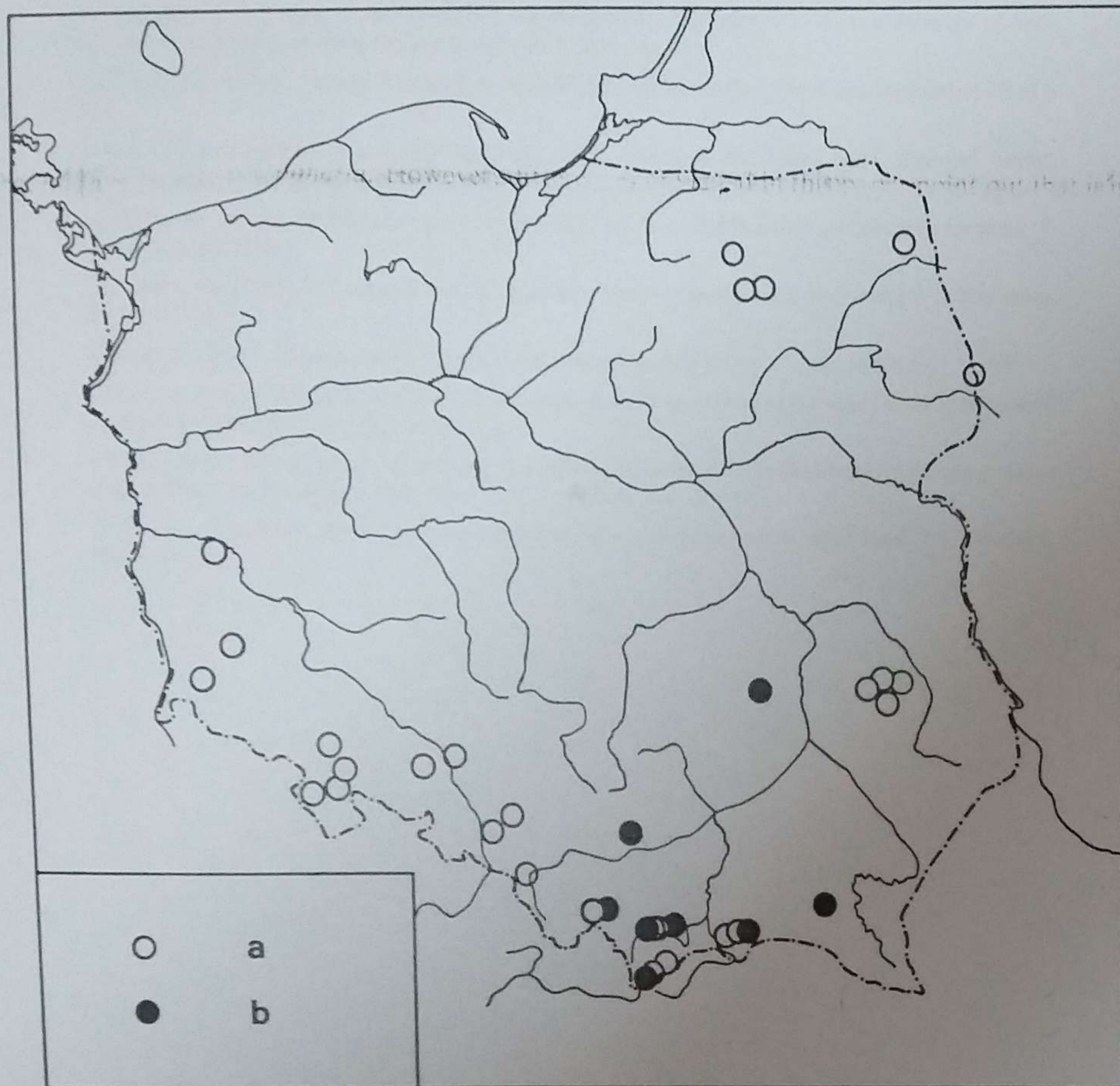


Fig. 7. Distribution of *G. carbonaria* (a) and *G. rehmii* (b) in Poland.

***Geopyxis rehmii* Turnau, sp. nov.**

Diagnosis: Apothecia 2-30 mm diam., aurantiaca vel aurantiacorubra, extus paulo pallidiora et sublaevia, margine subintegra, non stipitata vel stipite parum distincto. Asci 187-250 × 8-12 μm, cylindrici, octospori. Sporae 10-19 × 9-10 μm, elipsoideae, verrucosae, eguttulate. Paraphyses ascos superantes, apice 6-8 μm crassae, apice aurantiacae. AG. *carbonaria* (Alb. & Schw. ex Fr.) Sacc., cui proxima, apotheciis majoribus, laetius coloratis, margine albida destitutis, haud stipitatis, nec non sporis verrucosis, differt.

Habitat: locis exustis, ubi lignum Coniferatum urebatur.

Holotypus: Carpati Occid., montes Gorce, Stare Wierchy (prope montem Turbacz), altitud. 960 m s.m., 5.IX.1979. Ig. Katarzyna Turnau — in herbario Universitatis Jagellonicae Cracoviae (KRA) asservatur.

Conclusions

The characterization of *Geopyxis rehmii* presented above fulfils the requirements of description of genus *Geopyxis* (Dennis 1978, Eckblad 1968). As far as all the analyzed species are concerned *G. rehmii* is most similar to sessile specimens of *G. carbonaria*. However, differences presented in this paper point out that it is a separate species. *G. rehmii* has not been described so far, except for mentioning the major form (Rehm 1896), which did not fulfil the requirement of the diagnosis. In relation to other genera *Geopyxis rehmii* is very similar in shape and colour of apothecia to some species of *Aleuria* and *Peziza* (Dennis 1978) but differs in the lack of oil drops in the spores and the blue reaction of its ascus tip with iodine. *G. rehmii* might be confused with *Tarzetta* (Cooke) Lamb. and *Rhodotarzetta* Dissing & Sivertsen especially with *T. cupularis* sub. *Pustularia cupularis* (Maas Geesteranus 1967, Dennis 1978) and *R. rosea* sub. *Pustularia rosea* (Dissing, Sivertsen 1983, Maas Geesteranus 1967) cited from the same biotop. These two genera should be easily distinguished by large oil drops in their spores. Besides *R. rosea* can be distinguished from *G. rehmii* by its bright pink apothecia. *T. cupularis* is similar in the colour of apothecia to *G. rehmii* but differs as far as microscopic characteristics are concerned.

Considerable changes in the physico-chemical conditions of burns periode differential habitats for particular species both in time and space. This is reflected in the succession of species, which usually appear on a burns over a certain specific period (Turnau 1984). Considerable differentiation of conditions on such a small areas over relatively short periods of time may have been of much greater importance, and may have been responsible for the differentiation of new species. Such may have been true of *Geopyxis carbonaria* and *G. rehmii*. These species differ greatly in respect to their range of tolerance for conditions that may prevail on a given burn. *G. carbonaria* fruits in sites where soil pH-values are high and appears on burns in the first year after burning, but in dry usually shaded places or in wet places where, because of a stalk, it is better adapted to the dissemination of spores. *G. rehmii* fruits later, when soil pH-values are lower. The maximum fructification in this fungus was recorded on dry burns, fully exposed to the sun. This species, therefore seems better adapted to retain moisture, a serious deficiency affecting other organisms found in this type of habitat.

I express sincere thanks to Assistant Professor Dr. Barbara Gumińska for her valuable instructions concerning the present paper, to Dr. Zbigniew Dzwonko for access to programmes for statistical analysis and to Dr. Tadeusz Tacik for assistance with the Latin translation of the diagnosis.

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Pl. I *Geopyxis carbonaria*. A - apothecia (full-size), B-D - spores (SEM, $\times 1500$).

Survival of *Botryodiplodia theobromae* in Yam Tissues

by

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With 2 plates and 1 figure

Abstract: The continued persistence of *Botryodiplodia theobromae* Pat., a yam-rot pathogen, from one storage period to another was investigated in the leaves, stem fragments and in the tuber of the host. The fungus survived in the stem for six months in the tubers for over eight months, and in the leaves for only three months. It was also viable for at least ten months in sterile soil samples inoculated with infected yam tuber pieces which served as food bases.

The fungus survived in yam tubers as modified hyphae in form of chlamydo spores.

Introduction

Although a considerable interest has been shown in *Botryodiplodia theobromae* as an important plant pathogen (Wardlaw, 1931; Meredith, 1961; Shaw, 1963, Strivastava, 1964; Olofinboba, 1967; Strivastava & Tandon, 1961 and Giha, 1977), there are not enough literature, if any, to show its continued existence after harvest until next cropping season. The fungus as a yam storage-rot pathogen as recorded by earlier workers (Dade & Wright, 1931; Okafor, 1966; Adeniji, 1970; and Ogundana et al., 1970) has not been further investigated as to how it survived in the host tissues from one storage period to another. This study examines the continued presence of the yam pathogen from one yam cropping season to another in the various tissues of the host (*Dioscorea* spp.) including the leaf, stem and tuber.

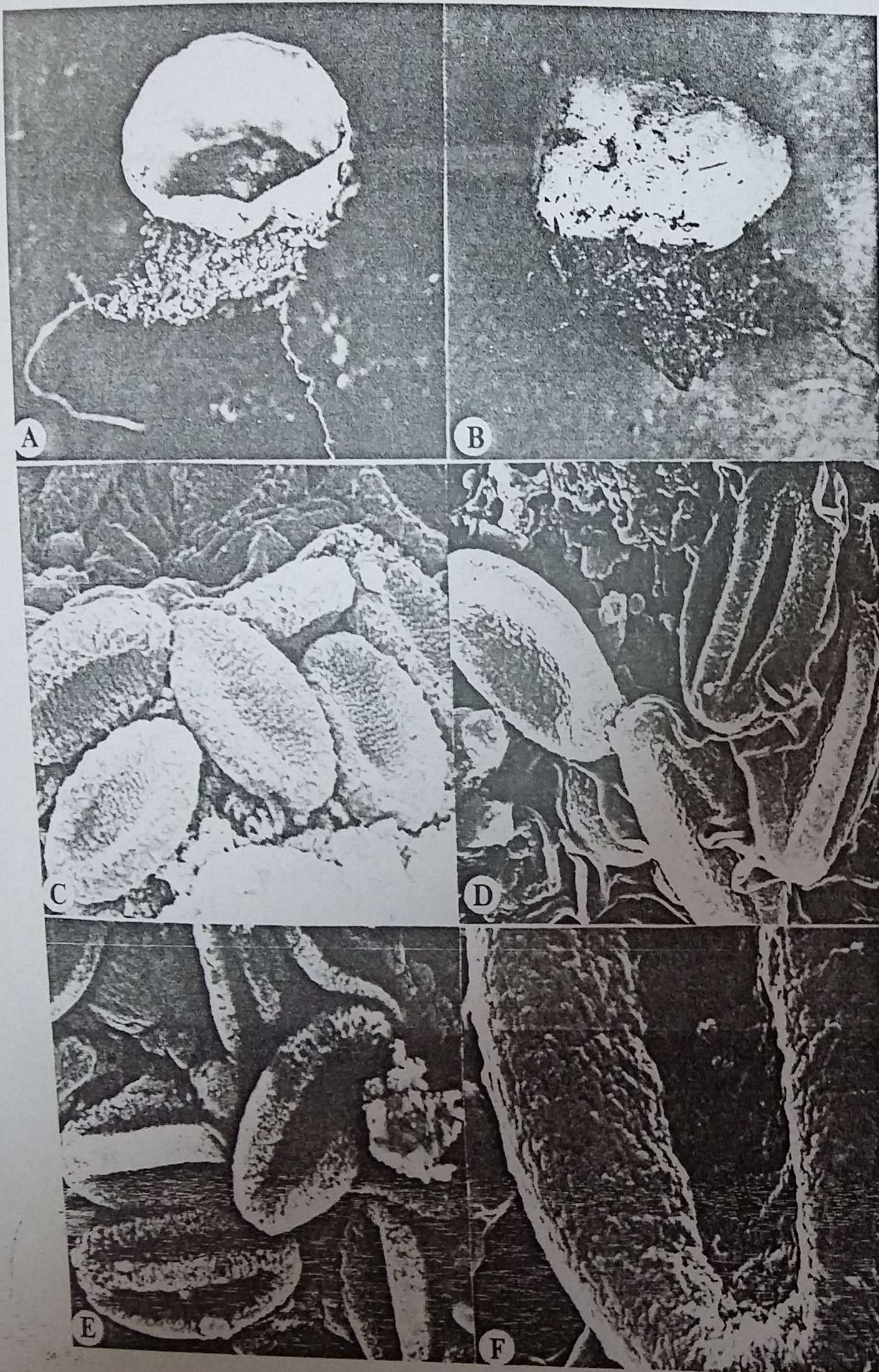
Materials and Methods

Preparation of Spore Suspension

Botryodiplodia theobromae was grown on potato dextrose agar (PDA) in aseptic conditions for twelve days in sterile Petri dishes at 25°C under continuous light (Ekundayo and Haskins, 1969).

Pycnidia from the twelve-day-old culture of *B. theobromae* were picked with a pair of forceps and broken with sterile spatula in a known volume of sterile distilled water. The suspension was later filtered through two thin layers of non-absorbent cotton wool and washed with sterile distilled water to a known concentration of the spore suspension.

The filtrate was transferred to infest the surface-sterilized host tissues which were stored in sterile conditions until completely rotten.



Pl. II. *Geopyxis rehmii*. A-B - apothecia ($\times 2$), C-F - spores (SEM, C-E - $\times 1500$, F - $\times 5500$).