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VARIABILITY IN *DIATRYPELLA FAVACEA* IN POLAND

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The results of a biometrical examination of Polish specimens of *Diatrypella favacea* on dead twigs of *Acer pseudoplatanus*, *Carpinus betulus*, *Corylus avellana*, *Betula* spp., *Alnus* spp., *Fagus sylvatica*, *Frangula alnus* and *Populus tremula* are presented. A new conidial anamorph associated with the perithecial stroma and *Diatrypella favacea* subsp. *nespiakii* subsp.nov. are described.

Both Tulasne (1863) and Nitschke (1867) described conidia (macroconidia) associated with perithecial stromata of *Diatrypella favacea* (Fr.) Ces. & de Not. To this conidial stage Traverso (1906) gave the name *Libertella favacea* Trav. The second type of conidium (microconidia) was described by Fuisting (1867). Croxall (1950) grew all British forms of *D. favacea* in culture and obtained two types of conidial fructification – conidiomata characteristic of the genus *Libertella* Desm. producing long, hyaline, filamentous, curved macroconidia and conidiomata characteristic of the genus *Naemospora* Pers. which produced short, slightly curved, hyaline microconidia. Croxall (1950) also observed conidial stromata associated with perithecial stromata in four collections: on *Betula*, *Corylus avellana* and two on *Fagus sylvatica*.

Conidial stromata associated with perithecial stromata on *Acer pseudoplatanus* and on *Carpinus betulus* have also been found. The purpose of this study is to try to explain the systematic position of these fungi.

MATERIALS AND METHODS

One hundred specimens from the herbarium of the author, and from W. Truszkowska, J. Kochman, the Institute of Botany, Polish Academy of Sciences, Kraków and the Herbarium of Warsaw University were examined.

Measurements were made from each specimen: the length of 10 necks, the height (excluding necks) of 10 perithecia, the length of the sporing part (p. sp.) of 20 asci, the length of 10 ascospores and, for the specimens where conidiomata were observed, the length of 100 conidia.

Statistical calculations for the measurements of asci included the following parameters: *m*, arithmetic mean; *sd*, standard deviaton; and *se*, standard error of the mean (Mayr, 1974). To demonstrate differences and establish similarities the calculations were presented by the Dice and Leraas method (Mayr, 1974). In Fig. 1 the character range, its mean, two standard deviations and two standard errors of the mean are provided. Coefficient differences (*cd*), were calculated for all populations as follows:

$$cd = \frac{m_b - m_a}{sd_a + sd_b},$$

where *m*-arithmetic mean, *sd* = standard deviation and a, b signify populations investigated (Mayr, 1974).

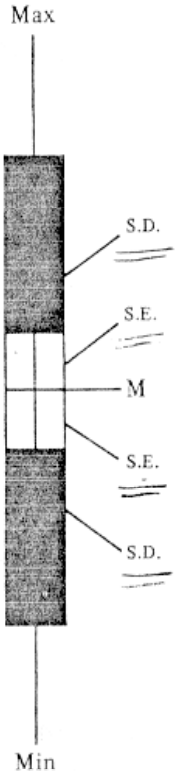


Fig. 1. M, mean; Min and Max, lowest and largest values for a character; S.D., standard deviation: S.E., standard error of the mean.

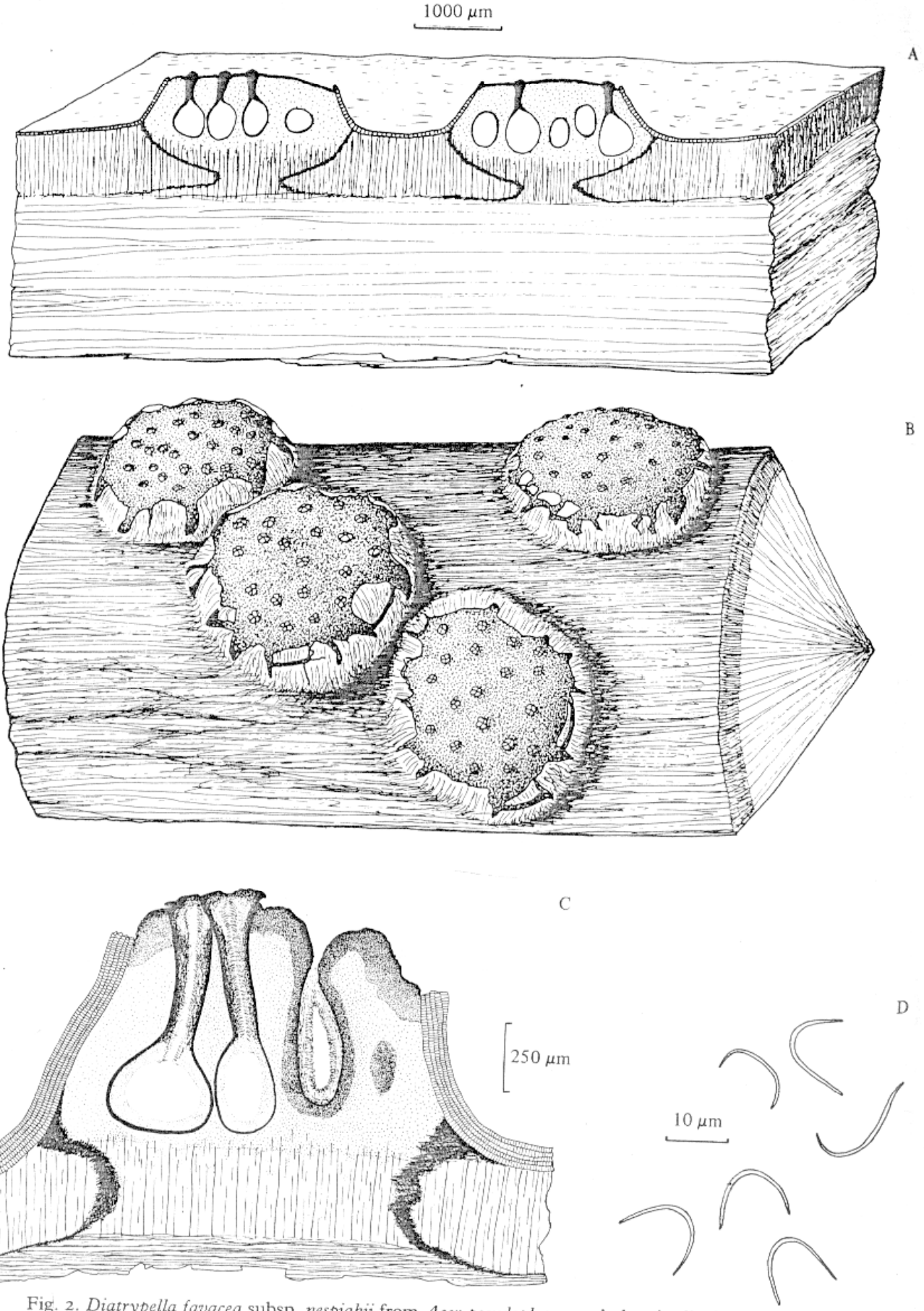


Fig. 2. *Diatrypella favacea* subsp. *nespiakii* from *Acer pseudoplatanus*. A, longitudinal section of perithecial stroma of sample 1; B, external appearance of the stroma of sample 1; C, longitudinal section of perithecial stroma of sample 2 showing conidiomata and young perithecia; D, macroconidia.

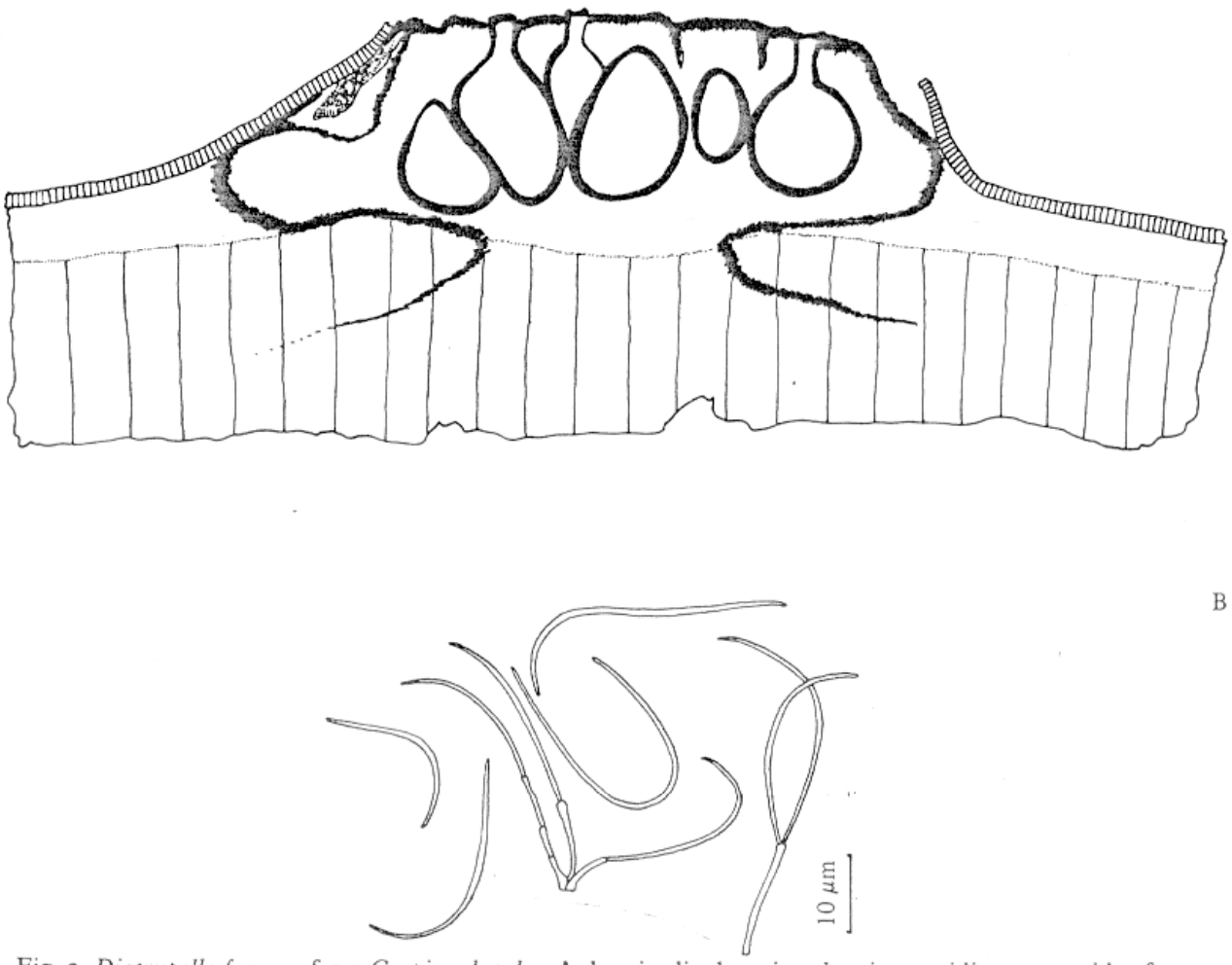


Fig. 3. *Diatrypella favacea* from *Carpinus betulus*. A, longitudinal section showing conidiomata on side of an old perithecial stroma of sample 100; B, macroconidia and conidiophores bearing macroconidia.

RESULTS

Anamorph stage

Separation of the stroma into two parts can be seen on the sections through the stroma (Fig. 2C). The dark, upper part or ectostroma forms a conidial stroma while the lower part of endostroma forms a perithecial stroma.

On Carpinus betulus (sample 100). Conidial stromata occur on the sides of old perithecial stromata and consist of a labyrinthiform cavity lined by a corrugated layer of conidiophores. The outer white hyphal layer is present below the conidiophores and is slightly darker in its lower part. As pointed out by Croxall (1950) this hyphal layer does not form a definite wall. The conidiomata contained strongly curved, hyaline macroconidia, 30–45 × 1·0–1·5 μm (Fig. 3B). The frequency–length curve for these conidia (Fig. 4) is similar to the curves obtained by Croxall (1950) for macroconidia of *D. favacea* on *Corylus*, *Betula*, *Alnus* and *Fagus*. Comparison of these curves and of the structure of conidiomata of *Libertella* supports the view that sample 100 from Białowieża National Park (leg. W. Truszkowska) may be included in *D. favacea*.

On Acer pseudoplatanus (samples 2, 7, 8, 10). The conidial stromata contain 1–3 separate conidiomata formed as a result of appression of ectostroma towards entostroma (Fig. 2C). These conidiomata are always unilocular and either ovoid or lageniform, 300–450 × 100–270 μm, with a funnel-shaped orifice ±100 μm diam. The conidioma, with distinct, black walls 8–30 μm wide, is lined by an immediately abutting and simple layer of conidiophores, ±25 μm long, which bore filamentous, strongly curved macroconidia (Fig. 2D) 15–25 × 1 μm. Usually conidiomatal openings are visible in the peripheral parts of the disk. The range

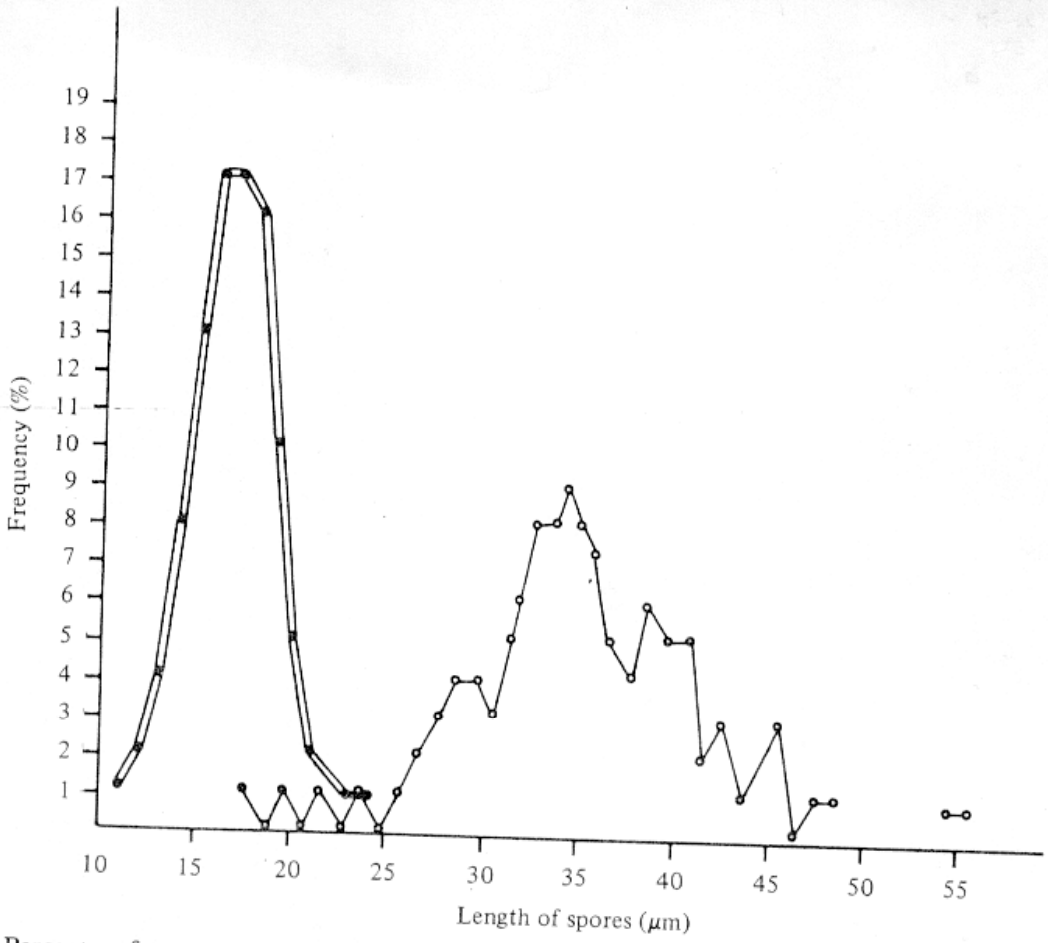


Fig. 4. Percentage frequency–spore length curves for macroconidia from herbarium specimens. =, *Diatrypella favacea* subsp. *nespiakii* from *Acer pseudoplatanus*; —, *Diatrypella favacea* from *Carpinus betulus*.

in macroconidial length partly corresponds with the range for *Libertella acerina* West. described by Westendorp (*Cinquième Not. Hypox. inèd.* p. 29 in Saccardo, 1884) from Belgium only as an anamorph. A comparison of the frequency-length curves of this stage from *Acer pseudoplatanus* with the corresponding curves obtained by Croxall (1950) indicates that they are quite distinct from the conidia of *D. favacea*.

Teleomorph stage

Length of ascus. Biometric analysis was based on the values of arithmetic mean together with its characteristics (Table 1) shown in the population–variability graph (Fig. 5). The differences in size range between populations on *Carpinus*, *Corylus*, *Alnus* and *Fagus* are not normally correlated with the substratum. This confirms the early observations and investigations of Croxall (1950). However, the size range of the sporing part of asci in populations on *Acer pseudoplatanus* and *Betula* sp. correlate with the substratum. Nitschke (1867) is probably correct in stating that the asci of *D. favacea* sensu stricto are consistently smaller than those of *D. verruciformis* and *D. nigro-annulata*. However, the differences between these asci are small.

In order to determine the degree of difference between populations on different substrate, the population on *Corylus avellana* was chosen as standard. The calculated coefficient (*cd*) between the standard population and residual populations is given in Table 3. For the population on *Acer*

Table 1. *Length (μm) of sporing part of asci*

		S.D.	S.E.
Acer pseudoplatanus	45·68	5·10	1·39
Carpinus betulus	86·84	11·48	3·30
Corylus avellana	91·70	9·89	2·91
Betula sp.	69·62	8·20	3·37
Alnus sp.	84·39	9·32	2·59
Fagus sylvatica	98·00	10·42	2·78
Populus tremula	73·01	11·51	2·57
Frangula alnus	70·46	5·24	1·65

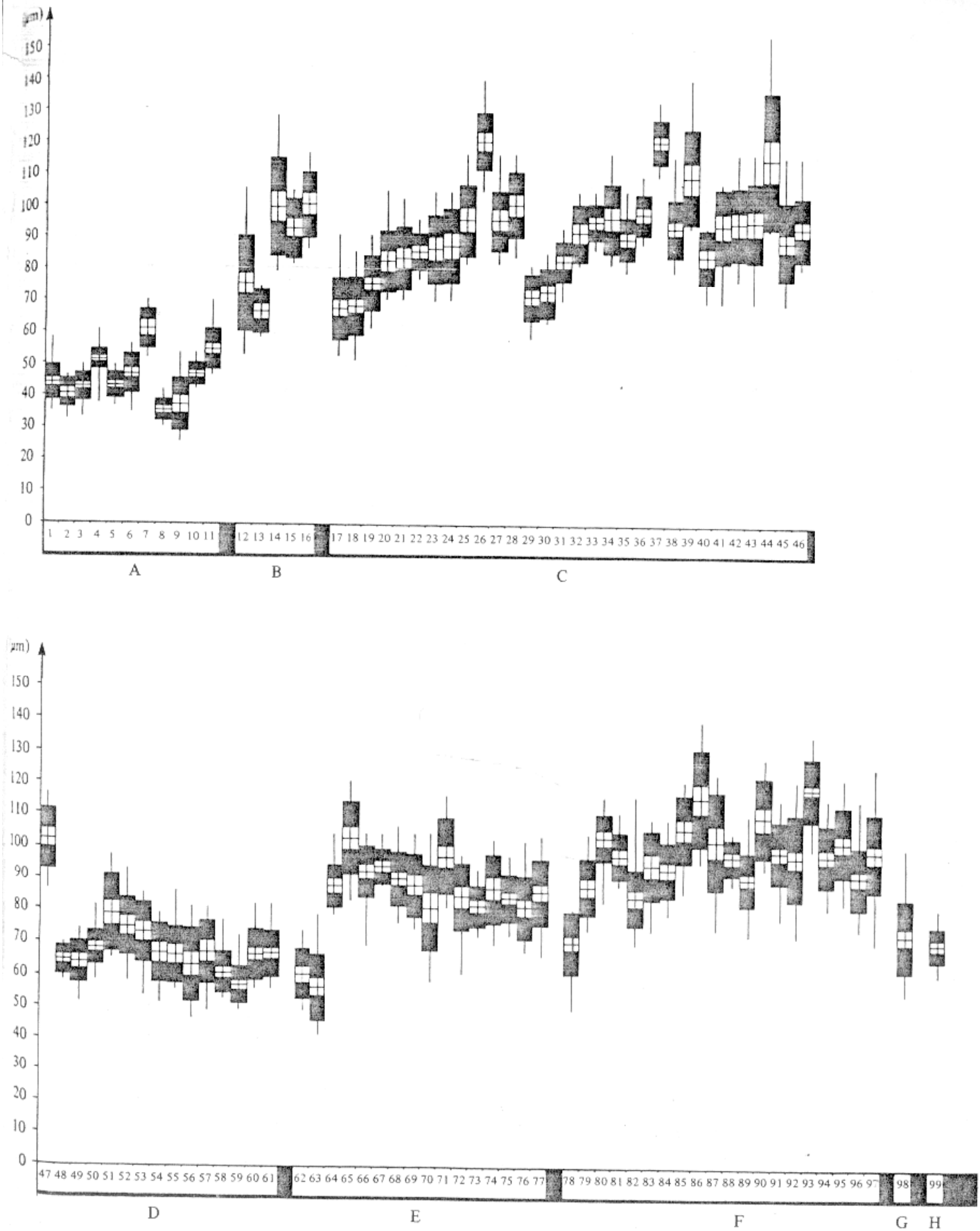


Fig. 5. Variability in ascus length. A, populaton of *Diatrypella* from *Acer pseudoplatanus*; B, population of *D. favacea* from *Carpinus betulus*; C, population of *D. favacea* from *Corylus avellana*; D, population of *D. favacea* from *Betula* sp.; E, population of *D. favacea* from *Alnus* sp.; F, population of *D. favacea* from *Fagus sylvatica*; G, population of *D. favacea* from *Populus tremula*; H, population of *D. favacea* from *Frangula alnus*.

Table 2. *Ascospore length (μm) of some populations of* Diatrypella

	Range	Mean
Acer pseudoplatanus	3·8–7·0	5·0
Corylus avellana	4·6–9·0	6·2
Carpinus betulus	5·2–8·0	6·3
Betula sp.	4·5–8·0	6·1
Alnus sp.	4·5–9·0	6·0
Fagus sylvatica	4·6–10·0	6·6
Frangula alnus	5·8–7·0	6·2
Populus tremula	5·7–9·0	6·8

Table 3. *Values of coefficient cd of some populations of* Diatrypella

	Total number of samples	*cd*
Acer pseudoplatanus	11	2·24
Carpinus betulus	5	0·17
Corylus avellana	30	0·00
Betula sp.	15	0·92
Alnus sp.	16	0·28
Fagus sylvatica	20	0·27

pseudoplatanus the value of the coefficient, $cd = 2{\cdot}24$, is considerably higher than the assumed subspecies difference level, where $cd = 1{\cdot}28$ (Mayr, 1974).

Length of ascospores. The shape of ascospores in all populations was uniform, invariably only slightly curved. The differences in the length of ascospores for different populations are small. For the population on *Acer pseudoplatanus* the mean length of ascospores is 1 μm smaller (Table 2).

Necks of perithecia. The variability range of the neck length was determined from measurements of 1000 necks. It was found to be similar in all populations and varied from 200 to 400 (500) μm. The calculated index H_p/L_n, where H_p is the height of perithecia and L_n is the neck length, indicates that the population on *Acer pseudoplatanus* is distinct. Necks of this population are more than half as long as the height of perithecia. In residual populations necks are usually less than half as long as the height of perithecia (Fig. 6).

Distribution. In Poland, populations of *D. favacea* sensu Croxall (1950) occur commonly almost throughout the country (Krupa, 1888; Schroeter, 1908; Namysłowski, 1909; Wróblewski, 1916; Truszkowska, 1959, 1960, 1963, 1965, 1967, 1974, 1976; Przebój-Pieniakówna, 1962; Domański *et al.*, 1960, 1963, 1967, 1970; Weber-Czerwińska, 1974; Truszkowska & Chlebicki, 1983*a*, *b*): the distribution of this species usually coincides with the distribution of the host plants. According to the terminology used by Uvardy (1978) this fungus is referred to as a eurytopic species aclimatically distributed in Poland. The occurrence of the population from *Acer pseudoplatanus* does not coincide with the distribution of the host plant. In the Sudety as well in the Carpathians (Fig. 7) it usually occurs scattered only in damp, wild forests of a lower mountain zone. Thus, this is a stenotopic population with a zonal distribution.

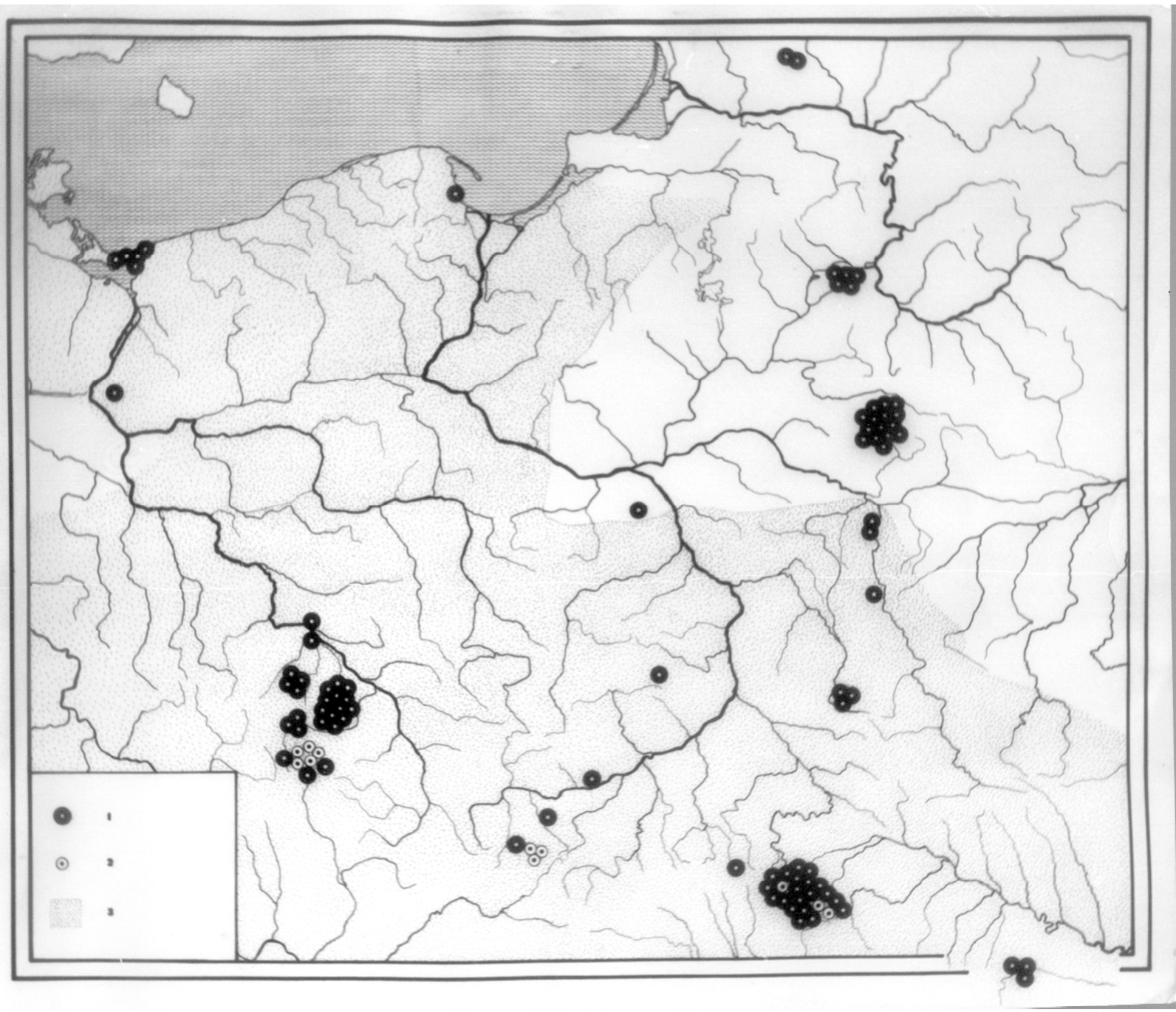
DISCUSSION

As pointed out by Croxall (1950) the five species so far described (*D. verruciformis*, *D. favacea*, *D. nigro-annulata*, *D. tocciaena* and *D. aspera*) (Nitschke, 1867; Saccardo, 1882; Winter, 1887; Traverso, 1906; Schroeter, 1908; Munk, 1957; Schrantz, 1960; Truszkowska, 1974; Smyk, 1980) are only different forms of one species, *D. favacea*. Croxall suggested that 'the differences in external form of the stroma of *D. favacea* in different collections are due not only to differences in the physical and chemical constitution of the substratum species but also to variations in the external environment and the rate of development of the fungus.' The results obtained confirm Croxall's opinion. The differences in the variability range of several morphological and anatomical characters for these species are small. The difference degree for the investigated forms of *D. favacea* is expressed by the values of *cd* (Table 3). Only the population from sycamore is distinctly different here. It is different not only in the size range of the sporing part of the ascus but also in the length of ascospores and macroconidia, in the form of conidial stroma, in the values of H_p/L_n index and in its distribution. In particular the form of the conidial stroma deserves attention and cannot be compared with those described by Tulasne (1863), Nitschke (1867), Traverso (1906), Grove (1937) and Croxall (1950). It is described as follows.

Diatrypella favacea (Fr.) Ces. & de Not. subsp. **nespiakii** subsp.n.

Etym. in honour of the late Andrzej Nespiak

Stroma peritheciigerum minutum, e basi suborbiculari, conico-attenuatum 0·5–3 mm diam, erumpens, vertice planiusculo vel convexo nigricans intus albidum. Perithecia in singulo stromate 4–25, monosticha, minuta, ovoidea vel sphaeroidea, 200–500 μm diam, in collum longiusculum long. 300–500 μm, ostiolis ± radiatim sulcatis, rugosis, exsertis exasperato, crass. 220–250 μm. Asci minimi, anguste clavati, longe pedicellati, polyspori, parte sporifera 37–60 × 5–7 μm. Paraphysibus filiformibus, sporidiis leviter curvatis, dilute fuscidulis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4–6 × 1·0–1·5 μm. Stroma conidiiferum in forma pustulae plerumque rotundato-ampulliformis, membrana pustulae distincta, crass. 8–30 μm, nigricans, ostiolum ± 100 μm diam. Conidiis filiformibus, curvatis 15–25 × 1 μm. A typo differt membrana pustulifera distincta haud plicata, ascis et conidiis minimis.

In ramis *Aceris pseudoplatani* L. in Bielice (Wałbrzych voivodeship) Bialskie Mts (E. Sudety Mts), 23 Apr. 1983, leg. A. Chlebicki, WA 27826, holotypus (Herb. Inst. Bot., Warsaw University, Ujazdowskie 4, Warszawa, Poland).

Specimens examined: Abbreviations: FPE, *Fungi Polonici Exsiccati* (W. Truszkowska), Laboratory of Phytopathology, Academy of Agriculture, Cybulskiego 32, Wrocław; MP, *Mycotheca Polonica* (J. Kochman), Institute of Phytopathology, Warsaw Agricultureal University, Rakowiecka 8, Warszawa; KRAM, Herbarium of Institute of Botany, Polish Academy of Science, Lubicz 46, Kraków; WA, Herbarium of Institute of Botany, Warsaw University, Ujazdowskie 4, Warszawa; HCH, author's herbarium; W.T., W. Truszkowska; A.C., A. Chlebicki, *Diatrypella favacea* subsp. *nespiakii* on *Acer pseudoplatanus*, 1 (WA-27826), Bialskie Mts (Sudety), 23 Apr. 1983, leg. et det. A.C.; 2 (HCH-7), Bialskie Mts (Sudety), Oct. 1979, leg. et det. A.C.; 3 (HCH-505), Kletno, Śnieżnik Mt (Sudety), 21 Oct. 1984, leg. et det. A.C.; 4 (HCH-89a), Babia Góra Mts, 22 July 1983, leg. et det. A.C.; 5 (HCH-409), Babia Góra Mts, 16 Sept. 1984, leg. et det. A.C.; 6 (HCH-410), Babia Góra Mts, 16 Sept. 1984, leg. et det. A.C.; 7 (FPE-18), Bialskie Mts (Sudety), 20 June 1965, leg. W.T.; 8 (FPE-18), Bialskie Mts (Sudety), Sept. 1973, leg. W.T.; 9 (FPE-18), Ustrzyki Górne, Bieszczady Mts, Aug. 1960, leg. W.T.; 10 (FPE-18), Łubne, Bieszczady Mts, Sept. 1962, leg. W.T.; 11 (FPE-18), Ustrzyki Górne, Bieszczady Mts, Aug. 1960, leg. W.T. (Earlier Truszkowska determined samples 7, 8, 9 and 11 as *D. verruciformis* and 10 as *D. aspera*.) *Diatrypella favacea* on *Carpinus betulus*, KRAM, 1; FPE, 5; on *Corylus avellana*, KRAM, 2; FPE, 22; HCH, 5; MP, 1; on *Betula* sp., FPE, 5; HCH, 10; on *Alnus* sp., KRAM, 1; FPE, 9; HCH, 6; on *Fagus sylvatica*, KRAM, 1; FPE, 10; HCH, 8; MP, 1; on *Populus tremula*, KRAM, 1; on *Frangula alnus*, FPE, 1.

Through the kindness of Professor dr W. Truszkowska I have been able to examine the specimens of her herbarium. I am grateful to the Senior Editor, B. C. Sutton for his kind help. Professor dr A. Skirgiełło and Docent dr W. Wojewoda kindly read an early version of the manuscript. I am greatly indebted to Professor dr A. Parguey for library facilities and Doctor T. Tacik for preparation of the Latin diagnosis.

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