Note on apomixis in ferns

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Abstract

In 1964, two new types of apogamy were described in ferns. The sporangia contain at the starting-point 16 spore mother cells and at the ending-point 32 diplospores. On the one hand, the meiosis is replaced by a mitosis, on the other hand, the first meiotic division aborts and the restitution nucleus divides mitotically. The author's observations imply that the second process is a variant of the first one.

INTRODUCTION

Manton (1950) has reviewed apogamy in *Pteridophyta*. In 1964, Braithwaite and Evans described independently a new type of apogamy in ferns; the sporangia contain at the starting-point 16 spore-mother-cells and at the ending-point 32 diplospores instead of 64 reduced spores. However, the cytological descriptions of the process by these authors differ greatly. Indeed Braithwaite (1964), working on *Asplenium aethiopicum* (Burm.) Bech complex describes: "a new type of apogamy in Ferns". Actually the latter author was rediscovering a process already described by Mehra and Singh (1957) and Bell (1960) in *Hymenophyllaceae*; but furthermore, he was demonstrating the apomictic nature of the phenomenon, a demonstration which is very difficult in *Hymenophyllaceae*; the culture of their gametophytes in artificial conditions is not yet possible and so, by-passing fertilisation cannot be controlled. Later, Braithwaite (1969, 1975) discovered the same phenomenon in *Gonocormus prolifer* (Bl.) Prantl (*Hymenophyllaceae*) collected in the Salomon Islands and in the New Hebrides. Mehra and Singh (1957) give the following cytological description: it was noted that the chromosomes paired and formed bivalents and trivalents at zygotene, though the pairing was limited, but at diplotene all the

previously formed spirals between the paired chromosomes regress, the chromosome pairs fall apart, and numerous univalents appear. The chromosomes are rather longish and "mitotic" in appearance. Soon, however, at diakinesis they undergo marked condensation, becoming ovoid. This is followed by a short period of interkinesis when the chromosomes become enveloped in a thin nuclear membrane. Commonly a single interkinetic nucleus is not able to include all the chromosomes, and hence two or three nuclei of different sizes may be organized. This stage is short lived, and soon afterwards chromosomes prepare to divide mitotically as the second meiotic division to form diads. The description of Bell (1960) and Braithwaite (1964, 1969, 1975) differs only in minor details. Independently, Evans (1964) claims that *Polypodium dispersum* Evans has a completely somatic alternation of generations; during sporogenesis the sporophyte exhibits 111 unpaired chromosomes, 16 spore mother cells and 32 diplospores per sporangium. This author considers that the meiosis is replaced by a mitosis but that the system is not entirely perfect; occasional imperfections reach back to the familiar obligate apogamous situation, such as rare spore mother cells that have shown chromosome doubling, and in many sporangia occasional aborted spores or spore fragments can be seen among the very distinct diplospores.

Our observations on African *Hymenophyllaceae* allow the assumption that the description and the interpretation of Evans (1964) are correct (16 spore mother cells giving mitotically 32 regular diplospores) and that abbreviated meiosis (aborted 1st division) like that described by Mehra and Singh, (1957), Bell (1960) and Braithwaite (1964, 1969, 1975) leads only to irregular spores by a random cleavage of the chromosomes in "diads, triads or tetrads", or by a restitution nucleus. This process is a variant of that described by Evans (1964) and reported above and in a sense an imperfect version of it.

OBSERVATIONS AND DISCUSSION

It should be noted that there exists an abnormal way for each type of sporogenesis (Fig. 1). For example, for type I there are irregular meiosis of hybrids. For type II and III, endomitosis is sometimes incomplete as signalled by Manton (1950), Bierhorst (1977) and as observed in *Microgonium chamaedrys* (Taton) Pic. Ser. (Tilquin, 1978a) with 10 spore mother cells of variable size. Type IV is the subject of the following discussion.

Fig. 1. Diagrammatic representations of the four types of sporogenesis in ferns. Type I: sexual sporogenesis with one $2N$ archesporial cell giving after 4 successive mitoses 16 $2N$ spore mother cells and a meiosis, 64 N spores; type II: meiotic apogamous sporogenesis, one $2N$ archesporial cell, 3 successive mitoses, one endomitosis leading to 8 $4N$ spore mother cells, meiosis, 32 $2N$ diplospores; type III: one $2N$ archesporial cell, 2 successive mitoses, 2 successive endomitoses, 4 $8N$ spore mother cells, meiosis, 16 $4N$ "tetraplospores"; type IV: ameiotic apogamous sporogenesis, one $2N$ archesporial cell giving by five successive mitoses 32 $2N$ diplospores.

1. Expression of apogamy in different populations (Table 1)

In the population n° 144, apogamy of type IV is well established. All the sporangia always contain 32 regular diplospores. All the spore mother cells divide mitotically.

In the population n° 149, among nearly meiotic (type II) or mitotic (type IV, Photo 1) plates are found occasionally sporangia containing meiotic prophase (Photo 2) with univalents, bivalents, trivalents, ..., hexavalents and multivalents as well as irregular spores may be found. But statistical analysis of the size of spores issued from one and the same sporophyte and from different sporophytes demonstrates that the variation of the size is acceptable ($V_1 = 9.3$ and $V_2 = 5.6$) and that the difference between the mean size of the two samples is not significant. Apogamy with 32 diplospores type II or IV is thus well established in the examined population. Irregular meiosis is strictly occasional.

Table 1

Species and voucher specimens showing apomixis

Species	Voucher (1)	Chromosome number	Number of spores per sporangium	Type of sporogenesis
<i>Microgonium erosum</i> (Willd.) Presl	n°144	2N=204	32	IV
	n°149	2N=197	32	IV
		"N"=197	32	II
<i>M. chamaedrys</i> (Taton) Pic. Ser.	n°178	N=68	64	I (very exceptional)
		³⁹ II+ ¹⁰ I	variable	variant of type I (very exceptional)
		"N"=209	32	II
		"N"=479	16	III
		2N=166	variable	variant of type IV
<i>Selenodesmium guineense</i> (Afz. ex Sw.) Pic. Ser.	n°103	2N=168	variable	variant of type IV
		⁶ I+ ⁶ II+ ²⁷ III	variable	variant of type I
		¹³ I+ ³⁵ II+ ³ III	variable	variant of type I
<i>Vandenboschia chevalieri</i> (C. Chr.) Kunkel	n°150	+ ¹ IV	32	IV
		2N=102-104	32	IV
		"N"=36	32	II
		²⁷ II+ ² I	variable	variant of type I
		n=9	64	type I (exceptional)

(1) Voucher specimens have been placed in the Bruxelles Herbarium (Br), in the Herbarium of the National University of Zaire, Kinshasa Campus. The voucher specimens have been determined by Prof. R. E. G. Pichi-Sermolli.

However, among the population n° 178, statistical analysis is not necessary to show the significance of the variation of the size of the spores: the size varies between 10 and 85 microns with the model class around 37.5 microns, the size of the diplospores, suggesting a different origin of the spore production.

After this first survey, we studied the frequency of the different types of sporangia (Table 2). It is important to point out the similarity between the two samples. This analysis demonstrates again that within one population and even within one individual all the known cytological behaviours for apomictic ferns may exist. This situation has been recorded by Manton (1950) in one particular sorus of *Cyrtomium fortunei* J. Sm. in which sixteen-celled (type I), eight-celled (type II) and four-celled (type III) sporangia were found. Bell (1960) and Evans (1964)

Photo 1. Late mitotic prophase with the nucleolus (view in interferential contrast "Nomarski") — population n° 149

Photo 2. Meiotic prophase, phase contrast, populatino n° 149

Photo 3. Typical figure of "dispersion" metaphase, population n° 178 4. A restitution nucleus of population n° 178

Scale line equals 10 μ m.

Photos 1 and 2 are reprinted from *Caryologia* 31: 23-42, 1978.

Photos 5 and 6. A restitution nucleus of population n* 178 (continued)

Photo 7. Tetrad with micronuclei

Photo 8. Diad — population n° 144

Scale line equals 10 μ m, except Photo 8 — 1/3.

observed such a situation and speak of occasional imperfections. It is clear that a diversity of this degree between the developmental histories of different sporangia in an individual is a matter of great genetical interest: it is now evident that apogamy is due to one or more mutations. One population, one individual should be able to accumulate all the mutations which govern the different types of apogamy and the external conditions must determine one way or another.

Table 2

Relative frequency of the different types of sporangia in *M. chamaedrys* population n° 178

Number of spores	16	32	From 32 to 64
Sori issued from different sporophytes	14	63	30
Sori issued from the same sporophyte	5	20	9
	19	83	39

The total number of sporangia is 141.

2. Sporangia containing from 32 to 64 spores

In examining the relative frequency of the different types of sporangia, we see that the number of sporangia with from 32 to 64 spores is important (39/141). We have rarely observed hybrid meiosis, but frequently characteristic figures like in Photos 3, 4 and 5 giving by a dispersion or a random cleavage of the chromosomes irregular diads, triads, tetrads or a restitution nucleus instead of regular diads (Photos 6, 7, 8). This process is an aborted meiosis and has led into error Mehra and Singh (1957), Bell (1960) and Braithwaite (1964, 1969, 1975), masking the exact origin of the regular diads. It may be result of an imperfect genetic control of the replacement of meiosis by mitosis. Indeed, the residual tendency to meiosis 1) is not present in population n° 144; 2) is present but not significantly in population n° 149; 3) is very important in population n° 179.

Furthermore, it is difficult to accept that Photo 1 is not a late mitotic prophase because we can see the nucleolus, when in the meiotic prophase (Photo 2), the nucleolus has disappeared. It is yet less probable that Photo 1 is prophase II with a nucleolus after a short interkinesis.

We must come back to one assertion of Braithwaite: "Counts of spores per sporangia indicate that the theoretical maximum of thirty-two spores in each is not always attained, good sporangia yielding anything from twenty-three to thirty-two spores and occasionally less. This may be explained by a residual tendency for the spore mother cells to cleave into triads or tetrads at the end of the division". Some-

times, he observed the abortion in all the sporangia and attributes to the physiological state of the plant. It is clear that a tendency for the 16-spore mother cells to cleave into triads or tetrads leads to sporangia containing between 32 to 64 spores and not less than 32 as he says. If sporangium yields less than 32 spores, it can only be formed by spore mother cells one of which at least gives a restitution nucleus instead of a diad.

Generally we consider that irregular spores are not viable. But in the case of *Microgonium chamaedrys*, we have seen that irregular spores germinate. Such a peculiarity may be explained by the high degree of polyploidy attained by this fern (Tilquin, 1978a,b) and may explain the aneuploidy encountered (Tilquin, 1979).

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