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Compatibility groups in *Ascobolus immersus* – an indication of speciation

F. Meinhardt, H. Koch and K. Esser

Lehrstuhl für Allgemeine Botanik, Ruhr-Universität, Postfach 102148, D-4630 Bochum 1, Federal Republic of Germany

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Summary. The mating pattern between 38 strains collected at various places in Europe and Southern India was determined. There are at least three compatibility groups: A (23 strains) and B (9 strains) comprising the European isolates, and C (6 strains), the Indian isolates. Within each compatibility group sexual reproduction is, as expected, controlled by a bipolar mechanism of homogenic incompatibility. No fertile offspring are obtained in any intergroup crossing showing that there is genetic separation by heterogenic incompatibility. However, the European group B seems to be closer related to the Indian (C) group in that sterile fruit bodies are produced between + and – mating types. An indication for a further subdivision is the occurrence of a barrage-like phenomenon between representatives of all three groups. The data thus indicate how the start of speciation may be occurring in *Ascobolus immersus* by means of both spatial and genetic isolation.

Key words: Compatibility groups – *Ascobolus* – Speciation – Barrage

Introduction

According to the investigations of Rizet (1939), the sexual propagation of the coprophilous ascomycete *Ascobolus immersus* is controlled by a bipolar mechanism of homogenic incompatibility. It has subsequently been observed that in combinations between + and – mating types of strains of different geographic origin there are deviations from the expected fertility ranging from aborted ascospores to complete failure of apothecia formation (Lissouba 1960; Rizet, personal communication).

In order to understand these phenomena it was necessary to study the mating reactions of a sample of isolates collected in different geographic areas. The data presented in this paper show the existence of a least three different compatibility groups which are incompatible with each other.

Material and methods

Strains

Thirty-eight strains of *Ascobolus immersus* were isolated from cow manure collected from several places in Europe (Austria 1, Denmark 2, England 1, France 1, Germany 26, Switzerland 1) and in India (States of Kerala and Tamil Nadu 6).

Media

Peptone agar medium was used for the germination of ascospores (Esser 1976) and yeast extract agar medium for mycelial growth and fruit body production (Esser 1976).

Culture conditions

Ascospores were isolated and incubated on germination medium for 24–48 h at 40 °C. Following germination the resulting mycelia were transferred to yeast extract slants for storage.

Matings were achieved by inoculating the two partners on yeast extract medium in petri dishes. Cultures were kept under continuous illumination at 25 °C until fruit bodies and ascospores were formed (12–18 days).

Results and discussion

For each of the 38 races under investigation, the two compatible mating types were determined. Thereafter, both + and – mating types were crossed in all possible combinations in order to find out the interstrain mating reactions. The results are summarized in Fig. 1.

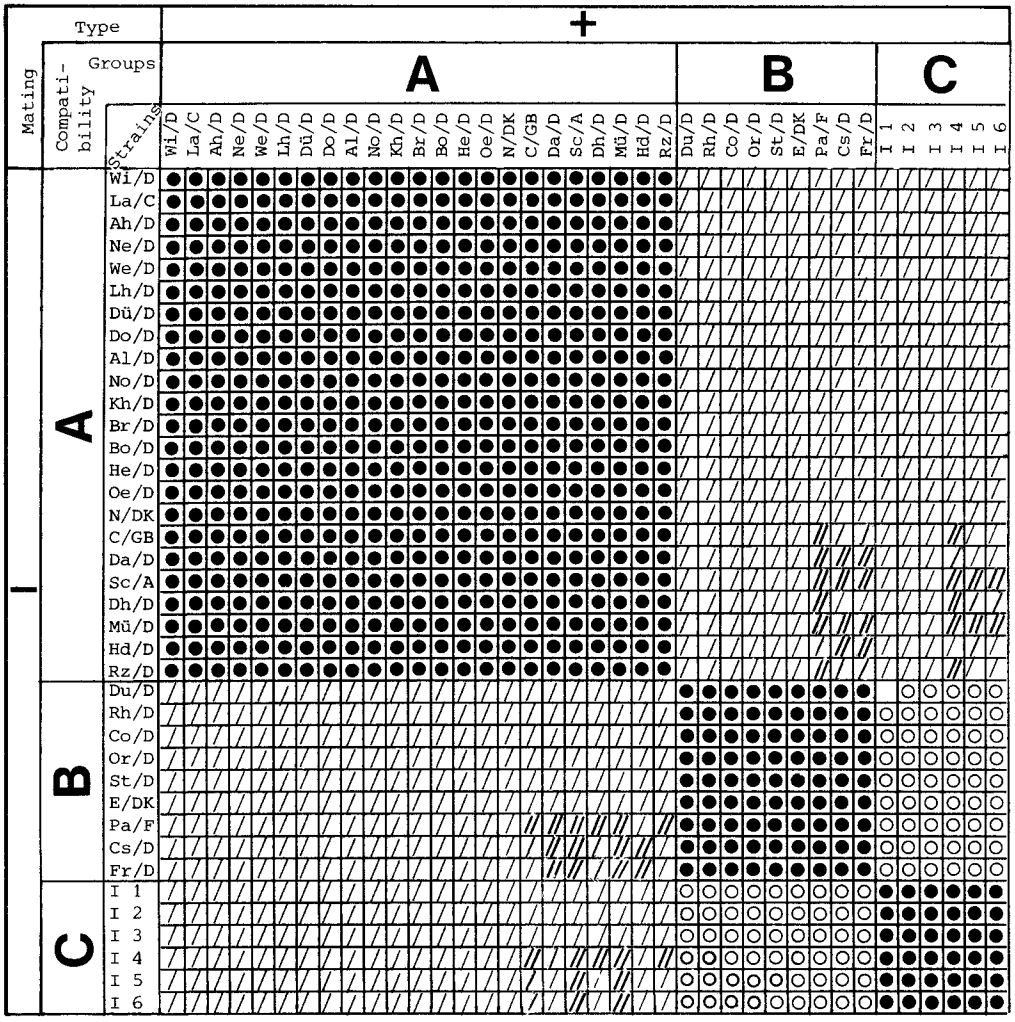

Fig. 1. *Ascobolus immersus.* Mating reactions between 38 strains isolated from different places in Europe and Southern India. The strains are designated according to their origin, that is the place (the two first letters) and country (last letters). A comprehensive list of all strains is available upon request. Meaning of the symbols: ● = compatibility, fertile fruit bodies; ○ = compatibility, sterile fruit bodies; / = incompatibility; // = incompatibility with strong pigment formation and cellular death in the line of contact

The data from Fig. 1 allow the following conclusions:

1. It is clear that the 38 strains can be subdivided into three compatibility groups. Within the strains originating from Europe there are two groups (A and B) and the strains collected in India have to be assigned to a third group, C. Within each group any combination of + and − mating types yields fruiting bodies with viable ascospores. This confirms the existence of the bipolar mechanism of homogenic incompatibility.

2. There is a complete lack of fruiting between the two European groups A and B. The same holds true for group A with the Indian group C. However, in all combinations between B and C sterile fruiting bodies are formed between opposite mating types.

3. In addition to the failure of apothecia formation between different mating types there are strong mycelial interactions in some combinations between A/B and A/C strains. These become manifest as a macroscopically visible black pigmentation in the zone of mycelial contact which is preceded by cellular death of the intermingling hyphae. This phenomenon is reminiscent of the border line formation between different species of the basidiomycete *Polyporus* (Hoffmann and Esser 1978) and the well known interspecific barrage formation in *Podospora* (Rizet 1952; Esser and Meinhardt 1984).

An interpretation of these experimental data may contribute to a better understanding of speciation which requires an initial separation and then independent

development of isolated populations (Mayr 1967; Stebbins 1968; Clémençon 1977).

Apart from spacial separation there are genetic isolation mechanisms known, such as heterogenic incompatibility (Esser 1971). In the material under investigation both of these mechanisms are present.

1. The existence of two compatibility groups within the European races seems to be caused by heterogenic incompatibility as there is no inhibition of fruiting within each group. This genetic barrier is not correlated with geographical dispersion. From this it follows that there are at least two genetically independent races of *Ascobolus immersus* within Europe. Albeit there is not yet any recognisable morphological differentiation. This phenomenon can be considered as the start of speciation. Because of the lack of genetic exchange between these two races, they will go their independent way in evolution.

2. The geographical distance between the European and the Indian races is also linked with genetic diversity. It is surprising that despite this diversity this group can interact differently with compatibility group A and B. The latter seems to be closer related to group C in that formation of fruiting bodies is still possible. This however does not abolish the de facto separation of groups B and C because of the sterility barrier.

3. An indication of a further subdivision of each incompatibility group is the occurrence of a barrage-like phenomenon showing clearly the expression of heterogenic incompatibility at the vegetative level. Due to hyphal death in the contact zone there is no heterokaryosis. This also inhibits any exchange of genetic material via the parasexual cycle.

In conclusion, the data presented exemplifies for *Ascobolus immersus* the existence of the two main isolation mechanisms (spacial separation, genetic isolation) which will eventually lead to further speciation in this taxon.

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