DIVERSITY, ECOLOGY, AND SYSTEMATICS OF SMUT FUNGI

Meike Piepenbring
Department of Mycology, Institute for Ecology, Evolution and Diversity, University of Frankfurt am Main, Germany

Keywords: cultivated plants, Entylomatales, *Erratomyces*, evolution, Exobasidiales, false smuts, Microbotryales, phylogeny, phytopathology, plant parasitic fungi, *Sporisorium*, systematics, *Tilletia*, Tilletiales, Ustilaginales, *Ustilago maydis*, *Ustilago scitaminea*

Contents

Summary

Smut fungi are plant parasitic microfungi living on host plants belonging to grasses, sedges, and numerous other families of plants. Host plants are mostly herbaceous and grow with elevated numbers of individuals in open vegetation. Spores of smut fungi are found as more or less voluminous powdery masses in different organs of the plant, often visible to the naked eye in flowers, inflorescences, or leaves. Other species of smut fungi develop their teliospores embedded in host tissue causing spots on leaves. These so-called sori are often accompanied by abortion of plant organs or abnormal growth of host tissue (galls). Spores are exposed by rupture of envelopes formed by fungal or plant cells, dispersed by wind, water, or animals, and germinate forming basidia, sexual cells carrying small spores. These spores contribute to further dispersal and multiply as yeast. After sexual fusion of two compatible cells, the fungus is able to infect a new host plant. Important genera of smut fungi are presented with their morphological characteristics, host relationships, and important species. Short descriptions help to recognize smut fungi known on cultivated plants in the neotropics.

Smut fungi do not correspond to one systematic group, but to several evolutionary lineages more or less independent of each other. Anther smuts in the Microbotryales are more closely related to rust fungi than to other smut fungi. Plant parasitic fungi without teliospores belonging to the Exobasidiales as well as yeasts growing on human skin, the Malasseziales, correspond to lineages which originated in close relationship to smut fungi.

Smut fungi are fascinating organisms important for humans as edible species and as parasites of cultivated plants. Numerous smut fungi on wild plants contribute to the biodiversity and complexity of interactions in natural and anthropogenic ecosystems.

1. Introduction

1.1. What are Smut Fungi?

During evolution, species of many different systematic groups of fungi discovered plants as sources of sugars. Some fungi reward the plants, for example as mycorrhizal fungi which help the plant to obtain water and minerals from the soil. Other fungi exploit the plant without advantage for the latter, living as parasites. Plants try to defend themselves against the intruders, by thick cell walls, hairs, or repugnant physiological mechanisms. Many fungi, however, learned to overcome the defenses, which are specific for different plants, thereby becoming specific for certain host plants. Especially biotrophic plant parasitic fungi, which interact with living host cells, generally have restricted ranges of host plants. For the identification of the fungi, it is therefore important to know the host plant species.

Species of smut fungi are most common on species of grasses (Poaceae). More than 40 % of the known species of smut fungi infect species of Poaceae, 15 % infect species of sedges (Cyperaceae). Most other host plants are herbaceous, including species of many different host relationships of dicotyledons and monocotyledons. Smut fungi occur throughout the world in ecosystems including their host plants, in grasslands, savannahs, swamps, on roadsides and in other disturbed vegetation, while they are almost absent in primary forests.

With approximately 1 450 species in about 77 genera, smut fungi are the second largest group of plant parasitic Basidiomycota. The largest group of plant parasitic Basidiomycota are the rust fungi (Pucciniales, formerly Uredinales) with approximately 9 000 species. In addition to these two groups, plant parasitic Basidiomycota can be found among macrofungi like pore fungi or some gill fungi and in small groups of plant parasitic microfungi like Exobasidiales.

The number of known species of smut fungi is higher for regions with temperate climate than for tropical latitudes. This is probably mainly due to investigation efforts, which have been much more intense in regions with temperate climate. For Europe, for example, about 400 species of smut fungi are known, while for the neotropics, we only know about 227 species. For many other regions of the world, monographic treatments are lacking and many tropical regions have never been foraged for smut fungi.

Smut fungi are characterized by the presence of dark, dusty masses of thick-walled spores which germinate forming basidia. In contrast to rust fungi, their life cycle includes only one host plant and a yeast stage in most species of smut fungi. Fungi forming light colored spores germinating with basidia (e.g. *Entyloma* spp.) have also been included in this group. Based on molecular data, nowadays we know that even fungi without any thick-walled spores should be included, as well as some asexual fungi and yeasts.

A smut fungus lives in a host plant, mostly close to meristematic host tissue, as hyphae formed by dikaryotic cells, i.e. each cell contains two haploid nuclei. Depending on the different species of smut fungi, the hyphae are restricted to intercellular spaces or form hyphae or haustoria inside the cells. By interacting with living host cells, the fungus obtains nutrients for its development.

By the presence of smut fungi, many host plants show abnormal growth of certain organs or tissues, i.e. they develop galls. The entire plant can be stunted, tillering, or show giant growth. Specific organs can enlarge forming hypertrophic galls, others can be aborted or transformed. These developments of the plant can form part of the sorus, the area where the smut develops its teliospores, and contribute to exposal and dispersal of the spores. Host tissue can form peridia (envelopes) for the teliospore mass. Peridia can also be formed by sterile cells of the smut or a combination of both, host tissue and fungal cells. In some smut fungi (especially *Sporisorium* spp.), vascular strands formed by the plant in the center of the sori develop hypertrophically forming elongated strands called columellae. Many smut fungi develop in ovaries, flowers, or in entire inflorescences, because these organs obtain nutrients provided by the plant to nourish fruits and seeds. In healthy plants, these organs are usually exposed for pollination and seed dispersal. Likewise, the teliospores of the smut are in excellent position for being spread by the wind.

For the development of teliospores, dikaryotic fungal hyphae in the host tissue are septated and numerous portions of cytoplasm are transformed to teliospore initials. The teliospore initials are embedded in a gelatinous matrix providing humidity and space for the growing spores. Each teliospore initial first develops a gelatinous outer sheath. Inside this gelatinous sheath, ornaments of the spores, a dark outer spore wall layer (exosporium), and a light colored inner spore wall layer (endosporium) are differentiated while the spore becomes more and more voluminous. This is the typical way of teliospore development for species of Ustilaginales, the largest order of smut fungi. In members of Tilletiales, teliospore initials are differentiated at the tips of sporogenous hyphae or lateral ramifications of hyphae. In these species, the gelatinous matrix is not as evident as in Ustilaginales. In mature teliospores of species of Tilletiales, endosporium and exosporium are separated by a thin middle layer. In species of Microbotryales, sporogenous hyphae are completely transformed to chains of teliospore initials which can apparently multiply by budding. A gelatinous matrix is lacking, but young teliospore initials are covered by thick gelatinous sheaths.

During the maturation of the teliospores, the two haploid nuclei (dikaryon) of the teliospore initials fuse forming a single diploid nucleus in each cell. This is the first step for the development of a basidium - therefore, teliospores of smut fungi can be

considered probasidia. The gelatinous matrix and gelatinous sheaths of teliospore initials are mostly consumed or get lost during teliospore maturation. In some species, however, the gelatinous matrix persists for teliospore exposal in slimy strands (*Melanopsichium* spp.) or the gelatinous sheath can be observed in mature teliospores as hyaline outer spore layer (*Entyloma* spp.) or as hyaline appendages (*Ustanciosporium* spp.).

Teliospores can be mixed with sterile fungal cells, cells which are dead or multiply asexually. Teliospores are simple in most species of smut fungi. Other smut fungi develop teliospores in pairs, in more or less firmly united groups, or in balls, sometimes including sterile fungal cells.

Teliospores are exposed to vectors for dispersal, like wind or water, by rupturing peridia or host tissue. While teliospores of smut fungi in warmer climate usually germinate immediately, teliospores of smut fungi in regions with temperate climate sometimes need low temperatures during a winter season to germinate in the following year.

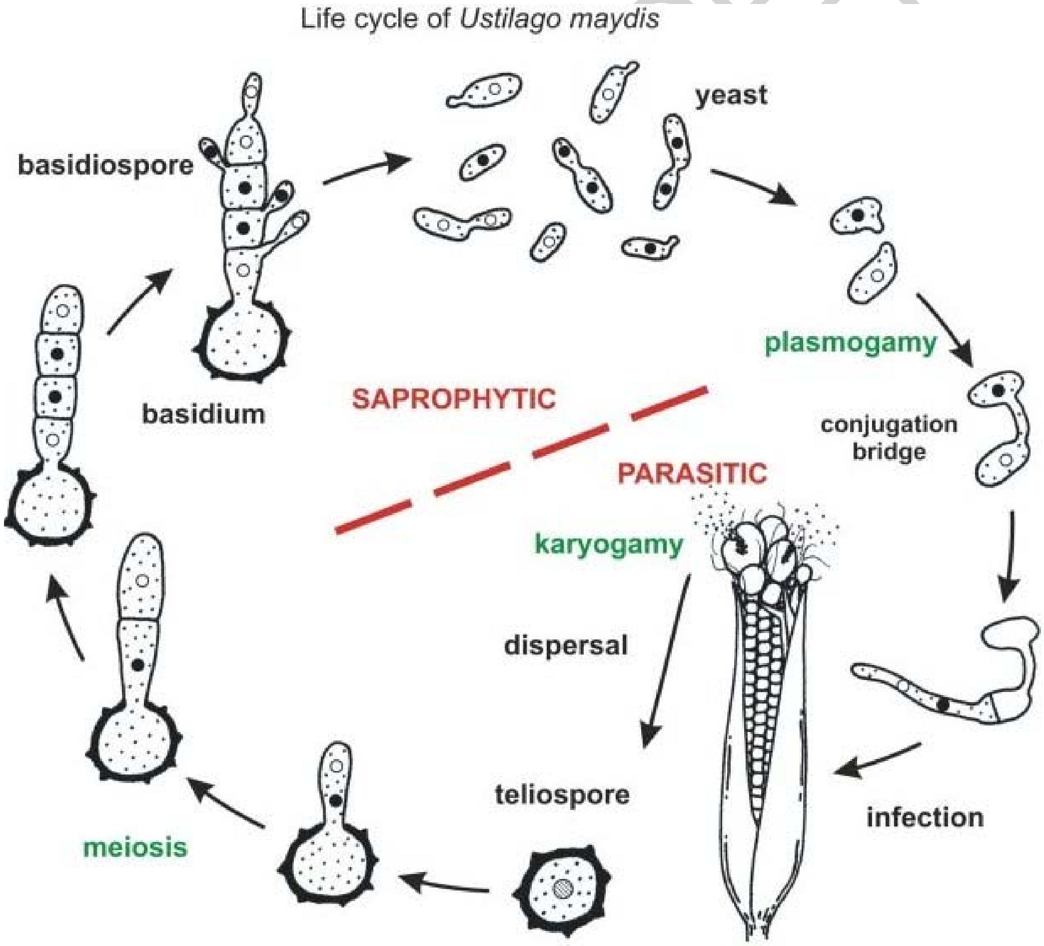

Figure 1. Life cycle of a typical smut fungus of the Ustilaginales, *Ustilago maydis*, on corn. Aspects of the nuclear and cellular cycle are written in green, ecological aspects in red.

When the teliospore is deposited in a humid place, the wall of the teliospore ruptures

irregularly, at germ areas, or at pores where the wall is thinner than in other parts, and develops a basidium. The hypha growing out of the teliospore forms one basidial cell (holobasidium) or mostly four basidial cells by septation (phragmobasidium). As a result of meiosis of the diploid nucleus, basidial cells contain four haploid nuclei. Holobasidia form four to numerous basidiospores at their apical parts, while phragmobasidia form basidiospores in terminal and lateral positions. Different types of basidia are characteristic for different genera of smut fungi.

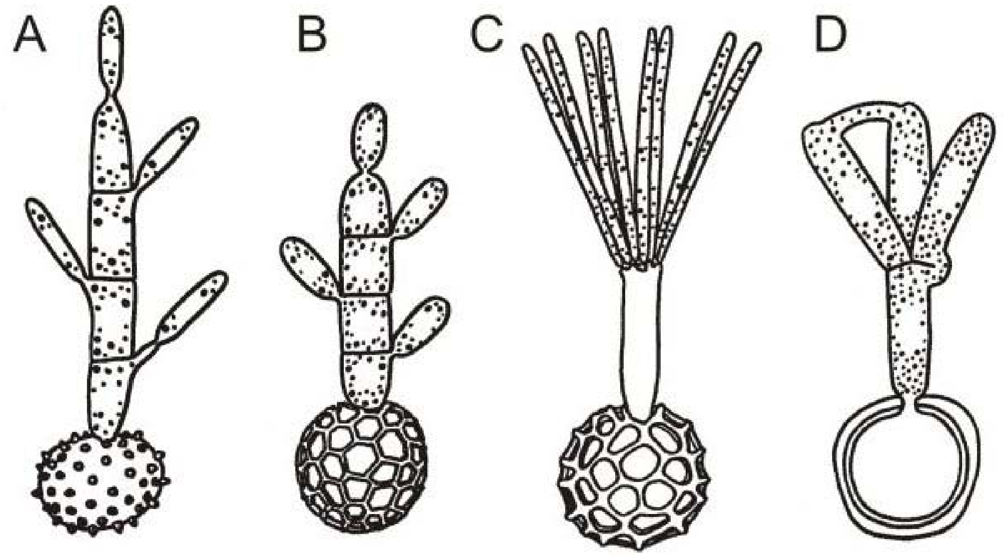

Figure 2. Different types of basidia of smut fungi. A. *Ustilago*. B. *Microbotryum*. C. *Tilletia*. D. *Entyloma*.

The haploid nucleus in a basidial cell of a phragmobasidium divides by mitosis before one nucleus migrates into the developing basidiospore. If the basidium finds enough nutrients in its close vicinity, one basidial cell is able to produce several basidiospores. In this case, the basidial cell buds like the basidiospores which are liberated. Many smut fungi grow saprophytically as yeast in the following developmental stage; others grow vegetatively forming hyphae and conidia which can correspond to ballistospores, i.e. spores which are actively discharged.

In order to infect a new host individuum and to switch to the plant parasitic way of life again, two compatible cells of two genetically different clones of the same smut species have to fuse by plasmogamy. The nuclei of the two cells, however, do not fuse immediately but are associated with each other forming a dikaryon. Dikaryotic cells form hyphae able to penetrate the tissue of susceptible host plants for infection. Some smut fungi cause local infections, i.e. the sorus develops where the infective hyphae succeeded in colonizing the plant. Smut fungi which infect plants systemically can spread over the entire plant individual and form sori in all the organs available for soral development as long as the host plant is alive. Systemic smuts with sori in flowers, for example, are present in all the flowers of the infected plant. The hyphae of the fungus finally develop sori and transform into teliospores as described above.

-
-
-

Bibliography

Bauer R., Oberwinkler F. and Vánky K. (1997). Ultrastructural markers and systematics in smut fungi and allied taxa. *Canadian Journal of Botany* **75**, 1273-1314. [One of the key contributions for the new system of smut fungi, dealing with character states of septal pores and host parasite interaction]

Bauer R., Begerow D., Oberwinkler F., Piepenbring M. and Berbee M.L. (2001). Ustilaginomycetes. *The Mycota*, VII Part B (eds. D.J. McLaughlin, E. McLaughlin and P.A. Lemke), 57-83. Berlin, Heidelberg: Springer-Verlag. [In this chapter of a book the new system of smut fungi is presented for the first time in detail. Publications in scientific journals forming the basis for the new system are cited therein.]

Begerow D., Bauer R. and Oberwinkler F. (1997). Phylogenetic studies on nuclear large subunit ribosomal DNA sequences of smut fungi and related taxa. *Canadian Journal of Botany* **75**, 2045-2056. [One of the key contributions for the new system of smut fungi, dealing with sequence data]

Begerow D., Bauer R. and Oberwinkler F. (2000) Phylogenetic placements of ustilaginomycetous anamorphs as deduced from nuclear LSU rDNA sequences. *Mycological Research* **104**, 53-60. [Systematic analysis of asexual stages of Ustilaginomycotina]

Begerow D., Bauer R., and Oberwinkler F. (2002) The Exobasidiales: an evolutionary hypothesis. *Mycological Progress* **1**(2), 187-199. [Systematic revision of four families included in the Exobasidiales]

Hendrichs M., Bauer R. and Oberwinkler F. (2003). The Cryptobasidiaceae of tropical Central and South America. *Sydowia* **55**(1), 33-64. [A revision of neotropical species of Cryptobasidiaceae]

Piepenbring M. (2000). The species of *Cintractia* s. l. (Ustilaginales, Basidiomycota). *Nova Hedwigia* **70**, 289-372. [A revision of the species of the *Cintractia* group on Cyperaceae]

Piepenbring M. (2003). Smut fungi (Ustilaginomycetes p.p. and Microbotryales, Basidiomycota). Flora Neotropica Monograph 86. New York Botanical Garden Press, New York. [Monographic treatment of 227 species of smut fungi known from the neotropics including keys, descriptions, illustrations, data on distribution, and host plants]

Piepenbring M., Bauer R. and Oberwinkler F. (1998). Teliospores of smut fungi: General aspects of teliospores and sporogenesis. *Protoplasma* **204**, 155-169. [Introduction to two following publications, one of them is cited here]

Piepenbring M., Bauer R. and Oberwinkler F. (1998). Teliospores of smut fungi: Teliospore connections, appendages, and germ pores studied by electron microscopy; phylogenetic discussion of characteristics of teliospores. *Protoplasma* **204**, 202-218. [The new system of smut fungi as reflected by characteristics of teliospore walls]

Piepenbring M. and Bauer R. (1997). *Erratomyces*, a new genus of Tilletiales with species on Leguminosae. *Mycologia* **89**, 924-936. [Presentation of a poorly known agent of leaf spots of beans in the Tilletiales relationship]

Piepenbring M., Stoll M. and Oberwinkler F. (2002). The generic position of *Ustilago maydis*, *Ustilago scitaminea*, and *Ustilago esculenta* (Ustilaginales). *Mycological Progress* **1**, 71-80. [Aspects of the *Ustilago – Sporisorium* group discussed based on morphological, molecular, and host characteristics]

Piepenbring M., Begerow D. and Oberwinkler F. (1999). Molecular sequence data assess the value of

morphological characteristics for a phylogenetic classification of species of *Cintractia*. *Mycologia* **91**, 485-498. [Results published here are used by Piepenbring 2000]

Vánky K. (1994). *European smut fungi*. Gustav Fischer, Stuttgart. [Monographic treatment of 400 species of smut fungi known from Europe including keys, descriptions, illustrations, data on distribution, and host plants]

Vánky K. (2002). *Illustrated genera of smut fungi, second edition*, 238 pp. St. Paul, Minnesota, USA: APS press. [A comprehensive treatment of all genera known at present].

Biographical Sketch

Meike Piepenbring occupies the professorship of Mycology within the Institute for Ecology, Evolution, and Diversity of the University of Frankfurt am Main, Germany, since 2001. She studied biology at the universities of Cologne, Germany, and Clermont-Ferrand, France, obtaining a Licence des Sciences Naturelles in 1990 and a Master in Biology in 1991. For her Ph.D. and habilitation (post-doc) she worked in the group of Prof. Dr. Franz Oberwinkler at the University of Tübingen 1991-2001. Since 1992 she travels to the neotropics for research and teaching. Her main research interests are neotropical plant parasitic microfungi, their ecology, morphology, systematics, and host plants. Teaching activities cover mycology, tropical mycology, anatomy and morphology of vascular plants, diversity and systematics of cryptogams and vascular plants, as well as tropical botany.