Active liberation of reproductive units in terrestrial fungi

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The dispersive units of terrestrial fungi are often set free violently. On finding a body at the base of a vertical cliff, the vital question is asked: "did he jump or was he pushed?". Likewise, the mycologist considering the active liberation of reproductive units should distinguish between the *jumpers* and the *pushed*. For both, the term 'discharge' has commonly been used but it is clearly not appropriate for the jumpers.

In species of *Conidiobolus*, favourite toys of practical classes in mycology, the conidium is a jumper. The end of the conidiophore (e.g. in *C. adiaeretus* Drechsler) juts into the conidium as a columella restraining it in an unstable configuration (Fig 1,A) (Ingold, 1992a). Suddenly the restraint is overcome and the re-entrant part everts with the result that the turgid conidium springs into the air.

The same kind of jumping occurs in rust fungi when aeciospores, singly or in small groups, lean from the aecium. An account of this is given by Buller (1924) and the phenonemon had also been observed by earlier workers.

It now seems that the most widespread jumpers of the Fungal Kingdom are the ballistospores of hymenomycetes, rusts and mirror-image yeasts. How such spores are liberated has vexed mycologists for most of this century, but at long last the solution of this problem seems to have been found. It appears that they spring from their sterigmata; they are not discharged. Theories relating to their liberation have been ably summarized by Webster & Chien (1990). The ballistospore story is here illustrated in *Itersonilia perplexans* Derx (Fig 1,B). When fully grown the spore is walled off from its sterigma to which it then adheres so loosely that it is removable by the gentlest touch of a microdissection needle on its sticky surface. Buller's drop suddenly appears on the hilar appendix of the spore and grows to its full size in seconds. At precisely the same time another drop, or rather a blob, develops on the nearby surface of the ballistospore. Both drops seem to be formed by condensation on strictly limited areas of the spore-wall where a strongly hygroscopic substance is suddenly exuded or uncovered. Having reached a definite size, Buller's drop makes contact with the other one and they instantaneously flow together, advancing the centre of mass of the combined spore and drops, with the result that this unit springs into the air. In much the same way one jumps from the ground by split-second upward displacement of one's centre of mass using muscular energy. With the ballistospore, power is supplied by the surface energy of the droplets, of which there is more than enough (Ingold, 1939). It is important to note that the rigid tip of the sterigma, as a taking-off pad, plays a vital part in the process. One difficulty in accepting the validity of this remarkable mechanism is envisaging how it might have evolved.

The distance to which ballistospores spring is in the range 0.1-1.5mm. However, in any one species the distance apparently varies within extremely narrow limits. This allows the existence of hymenomycete basidiomes in which extensive vertical hymenial surfaces are closely opposed. An extreme example is to be seen in *Ganoderma australe* (Fr.) Pat. (Ingold, 1992b).

Water-cannon mechanisms of spore discharge occur in most ascomycetes and in a few zygomycetes. In these a turgid cell, with a stretched elastic wall, ultimately bursts in a pre-determined manner shooting the spore-projectile, within or on top of it, into the air.

Although the basic dynamics of the active ascus are the same throughout the ascomycetes, there are considerable variations in detail. Here the situation in *Ascobolus immersus* Pers.:Fr. is illustrated as a rare example where there is a precise picture of an individual ascus just before and immediately after discharge (Ingold, 1985) (Fig 1,C). This is a very big ascus which squirts its coherent mass of eight spores to up to 35 cm. However, many asci have a range of only a few mm. Further, where dehiscence is by an apical pore, instead of a relatively wide lid, and especially when the spores are elongated, discharge is successive with each spore temporarily plugging the contracting ascus.

In passing, it is illuminating to compare ballistospores with ascospores that are violently discharged. First, ballistospores are always unicellular when they spring from their sterigmata. Secondly, there is comparatively little variation in size and form among ballistospores, while in ascospores the variation is enormous and many are septate (Ingold, 1966). Thirdly, according to the species, the distance of ascospore discharge is up to 70 cm, while ballistospores never jump to more than 0.15 cm.

In zygomycetes the most familiar water-cannon is *Pilobolus*, the subject of much study in the past century. Buller (1934) devoted half of Vol. 6 of his *Researches on fungi* to *Pilobolus* in a beautiful and most readable account of his own and earlier work. The sporangiophore is a large, turgid cell beset by aqueous droplets which are probably the result of condensation. It eventually bursts along a subsporangial line of weakness in the cell-wall, shooting its sporangium towards the light to up to 2.5 m. By an ingenious method Page (1964) managed to photograph the sporangium with its issuing jet of sap at the instant of discharge when it had travelled only a tiny fraction of its path of flight (Fig 1,D) .

The primary conidium of *Entomophthora muscae* (Cohn)Fres., which attacks flies, is discharged in the same way. The inflated conidiophore, with its terminal conidium, projects from the abdomen of the dead insect and finally bursts discharging the conidium, together with a droplet of sap, to about 1 cm. It soon germinates to form a short conidiophore bearing a secondary conidium. However, this jumps off after the manner of that of a *Conidiobolus* and germinates to give a mycelium (Ingold & Plunkett, 1979).

The mechanism of discharge in *Basidiobolus ranarum* Eidam, which regularly develops on the excrement of frogs, is quite different from that in *Conidiobolus* spp. or in *Entomophthora muscae,* in spite of all belonging to the same family, for the conidium is propelled into the air by a rocket (Ingold, 1934). The conidiophore, whether derived from a cell of the mycelium or from a sporangium, is a single turgid cell remarkably like the sporangiophore of *Pilobolus*, but on a smaller scale. However, the line of dehiscence is towards the bottom, not the top, of the swelling below the reproductive body. When rupture occurs, this upper region instantly contracts, squirting sap backwards and flying, on the recoil, into the air with the conidium. In flight the two components separate, the conidium outdistancing the rocket itself (Fig 1,E).

The catapult of *Sphaerobolus stellatus* Pers. (Fig 1,F) is the most spectacular piece of artillery in fungi, projecting a reproductive body to a distance of 5m or more. Originally (and correctly) illustrated in action by Micheli in 1729, it proved irresistible to Buller (1933) who devoted a large part of Vol. 5 of his 'Researches' to it, relying heavily on a splendid paper by. Walker (1927). It was also the theme of my second Presidential Address to the BMS (Ingold, 1972).

Just before discharge the basidiome consists of two little cups with toothed margins one inside the other and connected only by the points of the teeth. Lying loosely within the inner cup, and submerged in an aqueous liquid, is the globose glebal mass containing basidiospores and unicellular, dikaryotic gemmae. It is the inner cup, the elastic of the catapult, which, in the relief of tissue tensions, suddenly turns inside out discharging the glebal mass.

Glomerobolus gelineus, recently described by Kohlmeyer & Volkmann-Kohlmeyer (1996) (Fig 1,G) on culms of *Juncus*, looks like a tiny *Sphaerobolus*. It does not form spores; reproduction depends on a globose body (*ca* 200 μm diam and composed solely of hyphae) which is violently discharged. However, the mechanism involved is quite different from that of *Sphaerobolus*. At maturity the projectile lies, with its base loosely attached, in a saucer-shaped structure gripped firmly by four or five of its turgid lobes which are straining to draw together. Finally the basal attachment gives way and the lobes spring together projecting the reproductive body, as an apple pip is expelled when squeezed between finger and thumb, to a distance of 30 cm or more (Ingold, 1997).

The types of active liberation of reproductive units so far considered involve mechanisms requiring reasonably damp conditions for their operation. There are, however, some microfungi in which drying is the trigger for spore discharge. In these cases discharge depends on

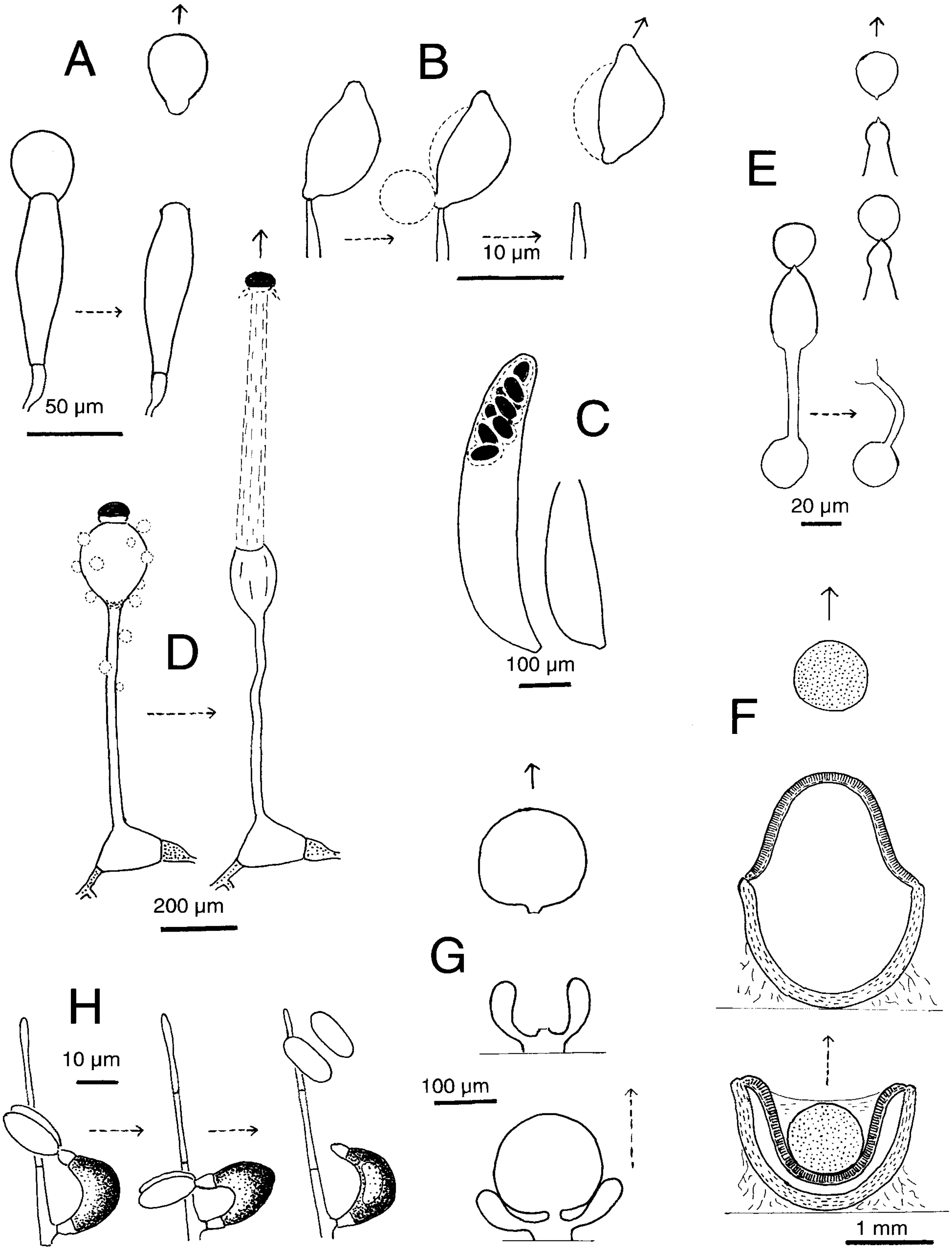

Fig 1 Active liberation of fungal reproductive units. A, *Conidiobolus adaeretus*; B, *Itersonilia perplexans*; 30 sec separate the first and third stages; C, *Ascobolus immersus*, ascus just before and just after discharge (traced from photograph); D, *Pilobolus* sp., intact sporangiophore and the same at the moment of discharge (issuing jet based on Page, 1964); E, *Basidiobolus ranarum*, mature conidiophore and the same on discharge (shown in L.S.);F, *Sphaerobolus stellatus*, open basidiome and at discharge (realistic diagram of L.S.); G, *Glomerobolus gelineus*, diagram of the situation immediately before and at discharge; H, *Zygosporium oscheoides*, stages in discharge, gas phase outlined in final stage (after Meredith). Solid arrows indicate projectiles still in flight. Dotted arrows indicate sequence of events.

the rupture of water under tension in certain cells in which the wall is thickened in a strikingly unequal manner. The fungi involved are dematiaceous hyphomycetes. An elegant example is *Zygosporium oscheoides* Mont. (Meredith, 1962) in which the conidiophore includes a curved, dark cell or 'falx' having a wall heavily thickened on the convex side, but thin on the concave one. The falx bears a hyaline cell capped by two oval spores. Drying leads to reduction in the water content of the falx, which accommodates by increased curvature involving the development of internal tension. When this reaches its limit, the tensile water breaks and the strain within the falx is released so that it returns instantly to its original form, slinging the two spores into the air. At this stage the contents of the falx consist mainly of gas, presumably water vapour (Fig 1,H). Another example of this mechanism is to be seen in *Deightoniella torulosa* (Syd.) M. B. Ellis (Meredith, 1961). However, it seems unlikely that active spore liberation of this kind is of much significance in the biology of these hyphomycetes. In passing, it may be noted that spore discharge involving the breaking of stretched water is the normal one in most ferns and leafy liverworts (Ingold, 1965).

Some of the fungi in the genera considered above, although beautifully organized in relation to discharge, diverge conspicuously in this respect from their immediate relatives. They seem remarkably isolated taxonomically. *Pilobolus*, *Basidiobolus*, *Sphaerobolus* and *Glomerobolus* are in this group. However, most fungi exhibiting active spore liberation are ascomycetes or basidiomycetes belonging to hundreds of genera. Finally it might be noted that, if the current view of ballistospore lift-off is right, the difference between the nature of spore liberation from ascus and basidium is profound.

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