of pupation (Table 3). Both species pupated farther away from medium hydrated with *Aspergillus* extract, the response being much more extreme in *D. hydei*. (Tukey tests isolated fungus extract treatment for both *Drosophila* species; *D. mel.* $P < 0.05$, *D. hyd.* $P < 0.005$).

Discussion: Similar to the findings of Atkinson (1981) when investigating the effects of *Penicillium* on *D. immigrans* and *D. melanogaster*, we found that the inhibitory effect of *A. niger* on *Drosophila* was species-specific: survival of *D. hydei* larvae was reduced if *A. niger* or its extract were present on the resource whereas *D. melanogaster* larvae were unaffected. Many fungi, including species of *Aspergillus*, produced complexes of mycotoxins. This includes aflatoxin which is produced by *A. flavus* and has been shown to inhibit all parts of the *D. melanogaster* life-cycle (Matsumura and Knight, 1967). Other toxins produced by the *Aspergillus* group include ochratoxin A and sterigmatocystin and *A. niger* produces a number of bioactive substances, including enzymes such as amylases, invertases, pectinases and lipases (Jay, 1992). It appears that *A. niger* produces some water-soluble metabolite which significantly reduces the viability of *D. hydei* larvae. *A. niger* is used commercially to produce citric and oxalic acids (Collins *et al.*, 1989) and pH of the resource can affect *Drosophila* performance (Hodge *et al.*, 1996). However, the IDM buffered the pH of the *Aspergillus* extract from ~4.0 to ~6.0 so it is unlikely that pH *per se* produced the observed effects.

Differences in the life-history of the two *Drosophila* species may play a role in the specificity of the fungal effects. For example, the development time of *D. hydei* is longer than that of *D. melanogaster*, so *D. hydei* would tend to be exposed to toxins for a longer period. Also, *D. hydei* larvae tend to feed deeper in the medium than *D. melanogaster* which could influence the effects experienced if the distribution of the toxin(s) is not uniform.

In general, it is believed that dipteran larvae pupate further away from environments which may prove harmful to the developing pupae (Casares and Carracedo, 1987). In this experiment, the larvae of both species of *Drosophila* pupated at a greater distance from the medium if fungal extract was present. This suggests that they may be able to detect and respond to some potentially deleterious chemical in the resource.

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Joly, Dominique[1], **Christophe Bressac**[2] **and Daniel Lachaise.**[1] [1]Laboratoire Populations, Génétique et Evolution, C.N.R.S., 91198 Gif sur Yvette Cedex, France. [2] IBEAS, URA CNRS 1298, Faculté des Sciences et Techniques, Université F. Rabelais, Parc Grandmont, 37 200 Tours, France. Gradually Elongating Testis and 'Sperm Roller' in *Drosophila bifurca*.

The evolution of sperm of inordinate length has sporadically occurred in the arthropod phylogenetic tree: ostracods in crustaceans, *Scutigera* in millipedes, and waterbugs, ptiliid beetles, and fruitflies in insects (Sivinski, 1984). Within the Drosophilidae family, this trend has seemingly been magnified uniquely in the subgenus *Drosophila* (Joly *et al.*, 1989; Pitnick *et al.*, 1995a) and the most impressive lengths occur in the *hydei* species subgroup including *Drosophila hydei* and *D. bifurca*, of which sperm lengths are on average 16.9 (Joly and Bressac, 1994) and 58.37 mm (Joly *et al.*, 1995), respectively. The sperm of this last species was unambiguously determined both by direct measurements using a dissection technique described elsewhere (Pitnick *et al.*, 1995b) and by indirect measurements using the correlation curve between sperm and testis lengths established previously (Joly and Bressac, 1994).

During the pre-reproductive life, *D. bifurca* testes were shown to elongate gradually, growing two-fold every 5 days until the 20th day post-emergence (Figure 1). As a comparison, *D. melanogaster* testes reach their maximum size as soon as the first day after hatching. Testis and receptacle were measured from males or females at different ages from 24 hour until the age of sexual maturity in *D. melanogaster* Canton S and *D. bifurca* (from Bowling Green Stock Center, number 15085-1621.0). Flies were reared on a standard corn-meal medium at room temperature. In each case (that is 0, 1, 2 ,5, 10, 15 and 20 day-old flies), 25 testes and receptacles were dissected and spread out of the abdomen in a drop of saline solution on a microscope slide. Slides were then let dry at room temperature and organs from reproductive tracts were mounted in a drop of glycerol. The measurements were realized using a camera (Hitachi, model KP-C551) connected to a Macintosh 660 AV with the NIH-Image Program (written by W. Rasband at the U.S. National Institute of

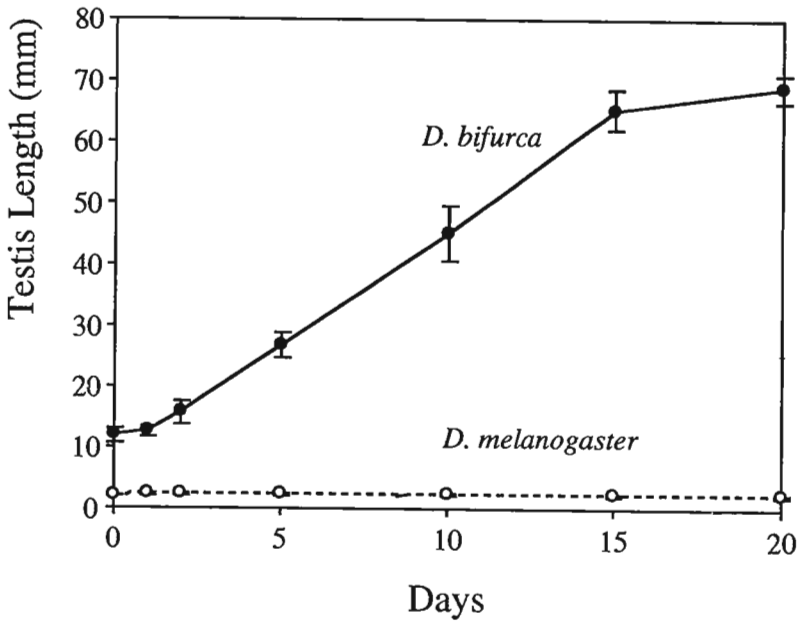

Figure 1. Enhancement of the testis length as a function of time in *D. bifurca* (giant sperm of more than 58 mm) and *D. melanogaster* (mid-sized sperm of less than 2 mm). Means are indicated with standard deviation for both species, but it is not visible in the case of *D. melanogaster* because of the Y scale.

Health) which can be obtained through anonymous ftp from zippy.nimh.nih.gov. One single testis was measured for each male. It is worth noting that sexual maturity in males (estimated by the occurrence of sperm in seminal vesicles) is reached when the testis elongation is nearly finished (e.g., 17 day-old in *D. bifurca*). This is consistent with Pitnick *et al.*,'s (1995a) data suggesting a relationship between the duration of the adult male nonreproductive phase and the sperm length in *Drosophila*.

Moreover, the most intriguing observation is that uniquely in *D. bifurca* the male tractus exhibits huge testes connected to the seminal vesicle via a special twisted and coiled structure, the 'sperm roller', absent from any other species previously described in the literature (Figure 2). This structure is seen as soon as the fly hatches and elongates and coils gradually during the testis development. The role of this peculiar structure seems to consist primarily in disentangling sperm issued from the same cyst, separating and rolling up them individually as a ball in the seminal vesicle. In the latter, the sperm are arranged in a rosary of huge separated monospermatic pellets of 80 μm wide (Figure 3) which are offered to females one after another at the rate of about 25 per mating (Joly *et al.*, 1995; Figure 4). The evolutionary significance of such unique reproductive tractus pattern and way of transferring sperm is controversial. A possibility, given as a working hypothesis, is that it could have some relevance with the multiple-mating behavior prevailing in both *D. hydei* and *D. bifurca*. It could for instance be a way to subdivide the input of synchronously-produced sperm in the seminal vesicles and thereby allow to 'control' the amount of sperm offered to each female. However, the fate of such giant sperm in females is still unclear (Bressac *et al.*, 1994; Joly *et al.*, 1995; Pitnick *et al.*, 1995b).

Figure 2. The 'sperm roller' structure localized between the testis (in the left part) and the seminal vesicle (not visible here).

How sperm pellets are manufactured in the reproductive tractus of *D. bifurca* males is documented in a forthcoming paper.

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Sperm in competition. Pp. 85-115 *in* R. L. Smith, ed. Sperm Competition and the Evolution of Animal Mating Systems. Academic Press, New York.

Figure 3. Giant sperm pellets arranged in a single file in the seminal vesicle.

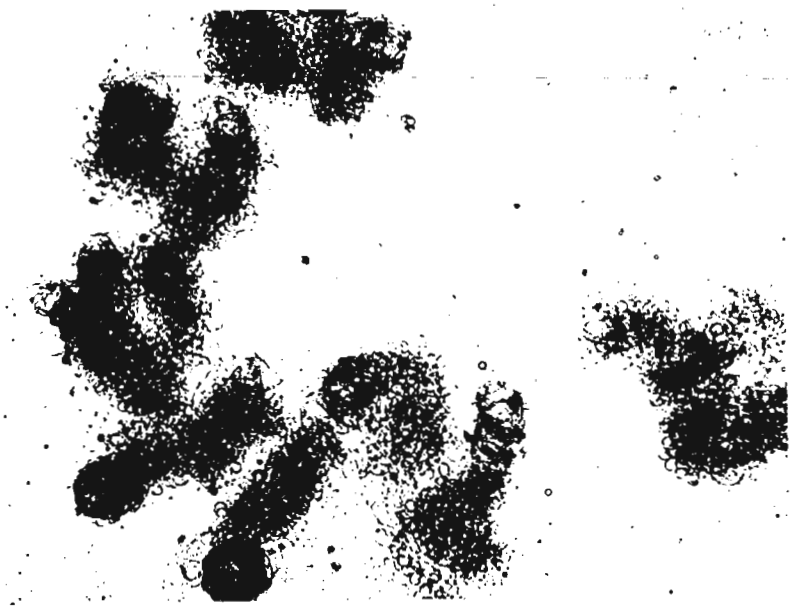

Figure 4. Giant sperm pellets released one after another into the female tractus.