THE SPERMATOZOON OF ARTHROPODA

VIII. The 9 + 3 Flagellum of Spider Sperm Cells

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In recent years a high number of exceptions to the prevailing 9 + 2 pattern of cilia and flagella has been found. Some cases are evident aberations in normal populations (2). Others appear characteristic of species or taxonomic groups, such as the well-known 9 + 9 + 2 of vertebrate and insect sperm, as well as the 9 + 0 of some fishes and the 9 + 1 of flatworms. Recently Baccetti, Dallai, and Giusti (4) and Phillips (11) have shown that 9 + 9 + 0 is peculiar to an insect order, Ephemeroptera; Baccetti, Dallai, and Rosati (5) have found 18 + 4 to be peculiar to Thysanoptera. Breland et al. (8) have found 9 + 9 + 1 in some mosquitoes; Phillips (11) has found 9 + 9 + 0 in *Psocus* and 9 + 7 in two Trichoptera. The sperm tail of Araneida was hitherto unstudied; it is now known to bear a quite particular and absolutely new pattern: 9 + 3.

MATERIALS AND METHODS

Testes of three species of araneids belonging to three different familier (*Pholcus phalangioides*, *Agelena labyrinthica*, and *Pardosa vittata*) were dissected in 2.5% Hoyle's-buffered, pH 7.2, glutaraldehyde for 20 min, postfixed for 5 min in osmium tetroxide in Hoyle's buffer, pH 7.2, dehydrated, and embedded in Epon. The sections were stained with 1.5% uranyl acetate followed by lead citrate, and were examined in a Siemens Elmiskop 1A 125 Kw.

RESULTS

The spermatozoon of spiders has a very long tail (~30 μ), which consists of the axial filament complex only because of the disappearance of the mitochondria during spermiogenesis. While the sperm is contained in the male gonads, it is rolled up and is enclosed by a thick wall, forming a cyst. The head is normally twice rolled, the tail is four times rolled. Throughout almost its whole length, the axial filament complex possesses nine peripheral doublets, each of which has short arms and a subfiber A which is smaller and more opaque than subfiber B, and three single central tubules of normal diameter (200 A) that are empty, arranged in a triangle, and are closely similar to subfiber B. As in the sperms of the lowest arthropods, rays, central sheath, and secondary fibrils are not evident. The centriole region is normal, with nine triple tubules in the periphery and no central units. The caudal part of the tail is devoid of doublets; only the three central tubules, still arranged in a triangle, remain.

DISCUSSION

The presence of a 9 + 3 sperm tail in a spermatozoon is a new cytological feature and apparently is characteristic of spiders. Afzelius (1) and Gibbons (10) demonstrated the presence of a third central element in the cilia of some Ctenophora and Mollusca. But the additional element was linear and not tubular like the other two elements. Baccetti, Dallai, and Rosati (6) found a similar small central fibril in Trichoptera sperms, which is obviously different from the two central tubules. None of these cases appears to be a true 9 + 3, but rather a normal 9 + 2 with a supernumerary filament and two central tubules lying, in cross-section, on the diameter of the tubule. Instead, the three central tubules of spiders are arranged in an equilateral triangle and are equidistant from the center of the cross-sectioned tail. Thus, the spider spermatozoon occupies a particular position in Arachnida; the lowest Arachnida (scorpions,

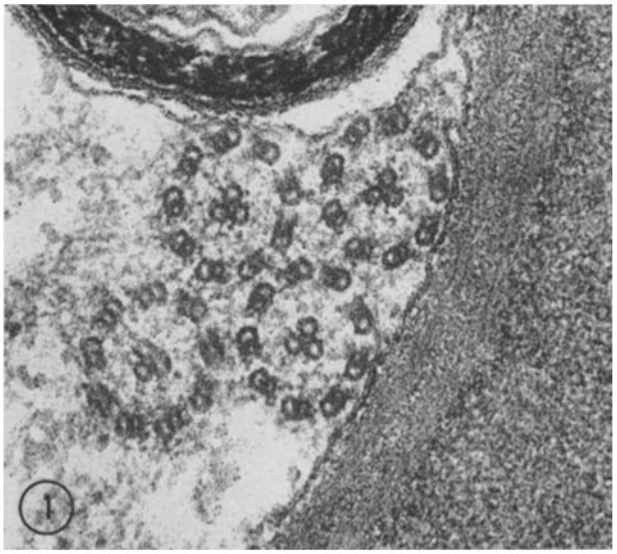

FIGURE 1 Cross-section of the tail region in a spermatozoon of *Pholcus phalangioides*. The tail is rolled four times on itself within the cell membrane. × 90,000.

pseudoscorpions) have a normal 9 + 2 spermatozoon (3, 7), whereas the highest (Acarina) have the well-known nonflagellate sperm (9, 12).

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REFERENCES

1. AFZELIUS, B. 1961. The fine structure of the cilia from Ctenophore swimming plates. *J. Biophys. Biochem. Cytol.* **9**:383.
2. AFZELIUS, B. 1963. Cilia and flagella that do not conform to the 9 + 2 pattern. I. Aberrant members within normal populations. *J. Ultrastruct. Res.* **9**:381.
3. ANDRÉ, J. 1963. Some aspects of specialization in sperm. *In* General Physiology of Cell Specialization. D. Mazia and A. Tyler, editors. McGraw-Hill Book Company, New York. 91.
4. BACCETTI, B., R. DALLAI, and F. GIUSTI. 1969. The spermatozoon of Arthropoda. VI. Ephemeroptera. *J. Ultrastruct. Res.* **29**:343.
5. BACCETTI, B., R. DALLAI, and F. ROSATI. 1969. The spermatozoon of Arthropoda. IV. Corrodentia, Mallophaga and Thysanoptera. *J. Microsc.* **8**:249.
6. BACCETTI, B., R. DALLAI, and F. ROSATI. 1970. The spermatozoon of Arthropoda. VII. Plecoptera and Trichoptera. *J. Ultrastruct. Res.* In press.
7. BOISSIN, L., and J. MANIER. 1966. Spermatogenése d'*Hysterochelifer meridianus* (L. Koch) (Arachnide, Pseudoscorpion, Cheliceridae). II. Étude de l'évolution du chondriome. *Bull. Soc. Zool. Fr.* **91**:697.
8. BRELAND, O. P., G. GASSNER, R. W. RIESS, and J. J. BIESELE. 1966. Certain aspects of the centriole adjunct, spermiogenesis and the mature sperm of insects. *Can. J. Genet. Cytol.* **8**:759.
9. BREUCKER, H., and E. HORSTMANN. 1968. Die Spermatozoen der Zecke *Ornithodorus moubata* (Murr). *Z. Zellforsch. Mikrosk. Anat.* **88**:1.
10. GIBBONS, I. R. 1961. The relationship between the fine structure and direction of beat in gill cilia of a Lamellibranch Mollusc. *J. Biophys. Biochem. Cytol.* **11**:179.
11. PHILLIPS, D. 1969. Exceptions to the prevailing pattern of tubules (9 + 9 + 2) in the sperm flagella of certain insect species. *J. Cell Biol.* **40**:28.
12. REGER, J. 1963. Spermiogenesis in the tick, *Amblyomma dissimili*, as revealed by electron microscopy. *J. Ultrastruct. Res.* **8**:607.