Fine Structure of Centrioles of the Grasshoppers, *Acrida turrita* Linné and *Atractomorpha* *bedeli* Bolivar

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Many animal cells and some plant cells show centrioles under a light microscope as a dynamic cell center. Several investigators have observed the fine structure of this organella in leucocytes (Yamada 1956), lymphocytes (Amano 1957), interstitial cells of the ovary (Yamada 1956), embryo cells (Yamada 1958), as well as in some other material from mammals such as mice, rats and man. According to some authors (Amano, 1957, Fawcett 1961, Freeman 1964, Robertis *et al.* 1965), the structure of centrioles resembles that of the basal body of the cilium or flagellum. In some material, centrioles have satellite bodies (De Harven and Bernhard, according to Robertis *et al.* 1965), while in others, they are surrounded by the Golgi complex forming a crown (Amano 1957, Yamada 1956, Freeman 1964).

Previous information was mostly obtained from vertebrate cells. Those from studies on the grasshopper cells have remained unsatisfactorily. While working with the fine structure of spermatocytes of *Acrida turrita* and *Atractomorpha bedeli*, centrioles somewhat different in structure from those of higher animal cells were found as reported in this paper.

Materials and method

Testes of grasshoppers (*Acrida turrita* and *Atractomorpha bedeli*) were fixed for 1 hour with 5% glutaraldehyde adjusted to pH 7.40 with phosphate buffer and post-fixed with 1% OsO₄ adjusted to pH 8.04 with phosphate buffer. Materials were rapidly dehydrated step-wise with ethanol and embedded in epoxy resin. Thin sections were cut and stained with Pb-tartrate. Observations were made with an electron microscope, model JEM 7.

Observations

Cells in the first meiotic prophase (about the diplotene stage) of *Acrida turrita* are shown in Figs. 1 and 2.

Sections of two centrioles in diplosome are seen in the region near the nucleus (Fig. 1, n). One is a section of the centriole longitudinal to the centriole axis and the other is a transverse section. As seen in three dimensions, the centriole may be a cylinder, 750–800 m μ long and about 170 m μ wide. The wall of the centriole is electron opaque, while the interior is less electron opaque. The longitudinal section of the centrioles shows that the wall of the centriole consists of two equal parts: thick (about 50 m μ) and electron opaque proximal half (near the nucleus) and a distal half which is thinner (about 25 m μ) and less opaque. The distal half of the centriole is surrounded by a granular body limited by a membrane. The electron opaque granules in the body are about 40–55 m μ in diameter.

Parts of two cells close to each other are shown in Fig. 2. A somewhat oblique section of a centriole surrounded by a granular body is also seen. The "arrow" indicates part of the granular body close to the cell membrane. In the narrow space between these two cells, intercellular substance (Fig. 2, i) containing electron opaque granules (about 40–50m μ in diameter) is seen. These granules in the intercellular substance resemble the granules in the granular body in size and in electron opacity.

Cells of *Atractomorpha bedeli* in meiotic prophase are shown in Figs. 3 and 4.

In a cell thought to be in the pachytene stage (Fig. 3), two transverse (C_1 and C_3) and one longitudinal sections (C_2) of centrioles are seen. Of these three centrioles C_1 and C_2 belong to a diplosome. C_3 may be a member of the other diplosome. In the wall of centriole C_3 , 9 groups of tubules, each of which contains three units, are recognizable. The proximal half of centriole C_2 is surrounded by the granular body.

Intercellular substance containing many electron opaque granules (about 40–50m μ in diameter) is seen between two adjacent cells thought to be in the diplotene stage (Fig. 4, i). Two centrioles in a diplosome, which are at right angles to each other, are seen between the nucleus (Fig. 4, n) and the cell membrane (indicated by the double arrows in Fig. 4). The cell membrane near the centrioles is seen protruding into the cytoplasm to cover the proximal half of a centriole in the diplosome, while the cell membrane (indicated by arrows in Fig. 4) of the adjacent cell, which is far from the centrioles, is flat.

Discussion

Studies of the fine structure of centrioles have been carried out by various investigators (Amano 1957, Yamada 1956, 1958, Fawcett 1961, Freeman 1964). According to some authors (Amano 1957, Fawcett 1961, Freeman 1964, Robertis *et al.* 1965), the fine structure of the centrioles bears a resemblance to the structure of basal body of the cilium or flagellum. The basal body has an electron opaque wall enclosing 9 groups of tubules.

The wall of centrioles observed in spermatocytes of the grasshoppers, *Acrida turrita* and *Atractomorpha bedeli*, consist of two equal parts: a proximal half near the nucleus and distal half. The former is thick (about 50m μ) and electron opaque and the latter is thinner (about 25m μ) and less opaque than the former. The appearance of the centrioles resembles that of the basal part of the cilium or flagellum. The structure of the proximal half of the centrioles especially appears

Figs. 1 and 2. Part of *Acrida turrita* cell in first meiotic prophase in the diplotene stage. 1, sections of two centrioles in diplosome are seen, near the nucleus. One is a transverse section. The other is a longitudinal section in which the distal half is surrounded by a granular body (g). 2, A somewhat oblique section of a centriole is seen. The distal half of the centriole is also surrounded by a granular body. The arrow shows a part where the limiting membrane of the granular body is in proximity to the cell membrane. g: granular body, i: intercellular substance, n: nucleus. Scale, 1 μ .

correspond to that of the basal body of the cilium or flagellum. The distal half resembles the cilium or flagellum proper, although it has no central filament.

Robertis *et al.* (1965) state that De Harven and Bernhard observed spherical and electron opaque satellite bodies (masses of about 700Å in diameter) on the outside of the centriole wall in leucocytes. Centrioles have usually been observed under a light microscope in the region of the Golgi complex and recently it has been suggested from electron microscope studies that the latter may form a crown round the centriole (Amano, 1957, Yamada 1956, Freeman 1964, Robertis *et al.* 1965). In the present materials, the distal half of the centriole is surrounded by a granular body, which contains many electron opaque granules (Figs. 1-3,) but satellite bodies such as those described by De Harven and others, or a crown of Golgi complex was not observed.

The granular body appears to originate from intercellular substance, that is to say, part of the intercellular substance intrudes into the spermatocyte cell covering the distal half of the centriole and this part is pinched off pinocytotically to crown the distal end of the centrioles. This possibility is supported by the following facts: 1) Granules in the intercellular substance closely resemble those in the granular body in size and in electron opacity (Figs. 1, 2 and 4). 2) A spermatocyte cell membrane is seen protruding into the cytoplasm with the distal half of the centrioles in a diplosome (Fig. 4). 3) The limiting membrane of the granular body is seen close to the cell membrane (Fig. 2).

Further studies are required on the behavior of the granular body during cell division.

Summary

The fine structure of centrioles was studied in spermatocytes at prophase of two species grasshoppers, *Acrida turrita* and *Atractomorpha bedeli*. It was found that the centrioles consist of two equal parts: a proximal half (near the nucleus) as a thickness of 50mμ in width and an electron opaque wall, while a distal half as a thinner (25mμ in width) and less electron opaque wall. The distal half of the centrioles is surrounded by a granular body which appears to originate from intercellular substance.

gs. 3 and 4. Part of *Atractomorpha bedeli* cell in first meiotic prophase. 3, meiotic cell in the pachytene stage. Three sections (C₁, C₂ and C₃) of centrioles are seen. C₁ and C₃ may be transverse sections of proximal halves of centrioles and C₂ may be a longitudinal section. C₃, 9 groups of tubules are seen embedded within the wall of the centriole (indicated by the arrow). C₁ and C₂ may be a member of a diplosome. The distal half of centriole C₃ is seen surrounded by the granular body. 4, part of two adjacent cells in the diplotene stage. Centrioles in a diplosome are seen. Intercellular substance, thought to be intruding into the cytoplasm, surrounds the distal half of the centriole. The double arrows and single arrows show the cell membrane of the upper cell and that of the lower cell, respectively.

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