OCCURRENCE OF THE INTACT SPERMATOZOA IN SPONTANEOUS EJACULATIONS OF ISOLATED MALE GUINEA PIGS

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ABSTRACT.—Spermatozoa were observed in coagulated spontaneous ejaculates of isolated male guinea pigs. The spermatozoa were morphologically intact, mostly grouped in rouleaux and their acrosomes were PAS positive. Spontaneous ejaculation in a non-mating male may be a way of eliminating spermatozoa from the genital tract.

In the course of studies on mature male guinea pigs, coagulated ejaculates have been observed in the cage service pans. A review of the pertinent literature indicates that very little is known about the fate of spermatozoa which are not used in copulation (Bishop and Walton, 1960; Bishop, 1961; Mann, 1964). Although several theories have been suggested (Simeone and Young, 1931; Amann and Almquist, 1962), the processes and locations of spermatozoa breakdown and the methods of their elimination are unknown. Spontaneous ejaculations are not mentioned. Preliminary observations concerning this phenomenon are the subject of this report.

MATERIALS AND METHODS

Five adult male guinea pigs (6-12 months old) were caged individually and kept on a standard pellet diet, supplemented with hay, carrots and cabbage. For one week, service pans in their cages were paper-lined daily and inspected around 10 a.m. for coagulated ejaculates. Those found were fixed in 10% neutral formalin containing 1% cadmium chloride. Ejaculates were embedded in paraffin and sectioned at 8 μ. Feulgen or Azure B methods (Swift, 1955) were used to visualized deoxyribose nucleic acid in the nuclei of the spermatozoa. Acrosomes were localized by the PAS (periodic acid Schiff) reaction (Leblond and Clermont, 1952). The latter technique was also used in combination with hematoxylin, Azure B or Alcian blue methods (Mowry, 1956).

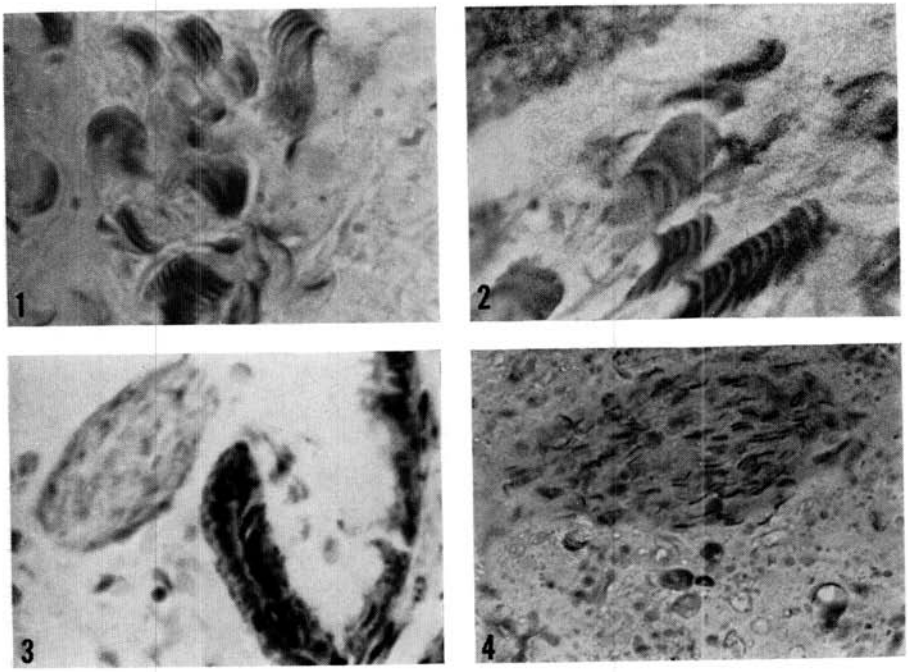
Epididymides and vasa deferentia from three adult guinea pig males (6-8 months old) were treated in the same manner as described for the ejaculates.

RESULTS

During the collecting week, 12 coagulated spontaneous ejaculates were found (range 2-3). All contained morphologically well preserved spermatozoa. The great majority of spermatozoa adhered to one another in tightly fitted groups with their bent heads in close contact (Fig. 1). Large crescent shaped acrosomes, characteristic of the guinea pig spermatozoa, were strongly positive with PAS technique (Fig. 1). In the all staining reactions performed, the spermatozoa in the spontaneous ejaculates did not differ from those inside the excurrent genital ducts.

In the cauda epididymis and the vas deferens most of the spermatozoa were also arranged in the orderly stacks described above (Fig. 2). The only exceptions were disordered, scattered spermatozoa in the spindle shaped masses occasionally observed in the lumina of the cauda epididymis and vas deferens (Fig. 3). The PAS method did not reveal any acrosomes in these spermatozoa, although their nuclei were still hematoxylin, Azure B or Feulgen posi-

FIGURE 1.—Spermatozoa forming rouleaux in the spontaneous ejaculate; PAS - Azure B. Reaction X 1,070. (2). Spermatozoa forming rouleaux in the cauda epididymis; PAS - Azure B reaction X 1,070. (3). Spindle shaped mass of disintegrating spermatozoa in the cauda epididymis; PAS - Azure B reaction X 430. (4). Spindle shaped mass of disintegrating spermatozoa in the spontaneous ejaculate; PAS - Hematoxylin X 430.



tive (Fig. 3). The whole spindle stained lightly with both Alcian blue and PAS techniques.

The identical masses, with the same staining reactions, were also observed in some of the spontaneous ejaculates (Fig. 4).

DISCUSSION

Experiments conducted by several investigators (Baker, 1931; Austin and Bishop, 1958) indicated that the acrosome in guinea pig spermatozoon is a highly vulnerable and very unstable structure which disintegrated and became detached from the head of spermatozoon soon after the cell died. Leblond and Clermont (1952) reported PAS technique to be specific for intact acrosomes of different spermatozoa. These researchers also noticed in the epididymis of the guinea pig that the successive acrosomes adhered to one another so that the sperm formed closely fitted groups, which the authors called rouleaux (Fig. 2). The spoon-shaped head of the guinea pig spermatozoon probably facilitates such formations (Baker, 1931). The PAS strongly reactive acrosomes and the presence of rouleaux in the spontaneous ejaculates indicate good morphological condition of the ejaculated spermatozoa (Fig. 1).

The spindle-shaped masses in the epididymis and vas deferens of the guinea pig were first reported by Simeone and Young (1931). They based their theory about the fate of unused spermatozoa on this phenomenon (Fig. 3). They proposed that spermatozoa not used in copulation dissolved in the vas deferens to form these masses. In my experiment the negative reaction of massed spermatozoa with PAS technique supported the view that the spermatozoa were dead for a longer time and their acrosomes were disintegrated. However,

both the size and the number of these masses were too small to account for elimination of vast numbers of spermatozoa by dissolution, which in the rat was shown to take four months (MacMillan, 1954). Also, the discovery of spindle-shaped masses in spontaneous ejaculates (Fig. 4) did not support the theory that spermatozoa disintegrate in the upper genital tract of the male.

The presence of morphologically intact spermatozoa in the spontaneous ejaculates of the male guinea pig indicated another way of eliminating surplus spermatozoa. Similar observations have been reported in the rat (Orbach, 1961; Martan and Risley, 1963).

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