ON THE STRUCTURE OF THE XY BIVALENT
IN MUS MUSCULUS L.*

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With 4 Figures in the Text

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There has been a long-standing controversy concerning the identification, structure and mode of pairing of the XY bivalent in the house-mouse.

On the one hand Creuz and Koller (1932) and Koller and Darlington (1934) developed a theory that in mice as well as in some other mammals X and Y have one pairing section in common and differ in a non-pairing differential section. On the other hand Painter (1927), followed by Oguma (1935), Makino (1941) and Matthey (1949, 1953) supposed that X and Y pair only by connection between the ends opposite to the centromeres. The peculiar behavior of the sex bivalent begins already in meiotic prophase; in pachytene the X is enclosed within the sex vesicle, called by Makino (1941) “amphinucleolus”. End-to-end connection of X and Y begins in diplotene. According to the first theory the first meiotic division can be reductional or equational depending on the position of chiasmata in relation to the differential segment and the centromere. According to the second theory, the first meiotic division is always reductional, X and Y passing to opposite poles.

Recently spermatogenesis in mice was carefully reinvestigated by Ohno, Kaplan and Kinoshita (1957, 1959a, b). In general their findings are in good accordance with the views of Oguma, Makino and Matthey. At the start of meiotic prophase X and Y are positively heteropyenotic, the autosomes negatively heteropyenotic. X and Y are two separate bodies differing considerably in size. In zygotene and pachytene they are embedded in the sex vesicle, which is rich in RNA. Digestion of pachytene nuclei with ribonuclease reveals both X and Y clearly as bipartite structures and with end-to-end connection already established. The sex vesicle disintegrates before diakinesis. In first metaphase X is large and rod-shaped and connected end-to-end with a very small Y. In anaphase X and Y always pass to opposite poles, their separation being prereductional. The size difference of X and Y is apparent also in somatic mitosis (Stich and Hsu 1960).

With respect to the behavior of the sex bivalent in meiosis, Ohno et al. believe that X and Y are connected by the ends of the extremely short arms proximal to the centromere, which they assume to be homologous and where they suggest

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a chiasma may be formed. Oguma, Makino and Matthey on the other hand believe the connection not to involve a chiasma and to involve the ends opposite to the centromere. According to them X and Y do not have any homologous segment. Recently however Ohno and Weiler (1961) have also considered this possibility on the basis of sex bivalent behavior in *Mesocricetus auratus* and common features of this bivalent in rodents, oppossum and man.

In striking discordance with the above findings are descriptions of spermatogenesis in mice published by Slizynski (1949, 1955a and b, 1960) and Griffen (1955, 1960), especially with respect to the structure of the XY bivalent in pachytene and first meiotic metaphase. Both authors have published chromosome maps of the mouse based on the study of male pachytene. Slizynski (1955a) has given also a diagramatic scheme of the structure and behavior of the sex bivalent. According to Slizynski and to Griffen X and Y in pachytene are paired side-by-side through their entire length. Both authors considered the sex vesicle to be a nucleolus attached terminally to the sex bivalent. In their pachytene maps 20 bivalents are shown: 19 autosomes and the paired XY of equal size within the limits of the long autosomes, and alike in chromonomic "banding". Slizynski is aware that this is in disagreement with commonly accepted standards for the dimensions of X and Y. He suggests that Y is not one of the smallest chromosomes but of medium size as is the X.

Slizynski and Griffen used a technique of preparing slides which differed from that of the other authors cited in that it permitted complete separation of pachytene chromosomes, following the destruction of the cell and rupture of the nuclear membrane.

The observations of Slizynski and Griffen differ so sharply from all others on the XY bivalent of the mouse that it seems of interest to report some new my observations on mouse chromosomes prepared according to the method described by Slizynski.

In the course of the work directed toward the cytogenetic analysis of mutants at the locus *T* in the ninth linkage group (cf. Dunn, Bennett and Beasley 1962), 63 young adult males of various *t*-genotypes *T/t, T/+, +/−* etc. were studied.

Testes were removed immediately after killing the animals, cut in half and transferred into vials with distilled water. The seminiferous tubules were removed and, after 5 minutes of pretreatment with distilled water, stained with acetocarmine for from 48 hours to 5 days. Afterwards single tubules were chosen, transferred to albuminized slides, cut in pieces about 1−2 mm in length and covered with cellophane squares. The squashes were made by rolling over a test tube, then the cellophane squares were removed by placing the slides in tap water. The slides were hydrolyzed in NHC1 at 56° C for 5 minutes and stained with lukewarm basic fuchsin for 20 minutes, bleached in a weak solution of *H2SO4*, washed in tap water, dehydrated and mounted in diaphane. Some additional slides were fixed with Sudan black B method as described by Griffen (1955), but the results obtained were not good, so this method was abandoned. Slides were studied with Zeiss phase-contrast microscope, and photos taken with 100× immersion Ph. objective, periplan Leitz ocular 10× and a Leitz camera.

The sex bivalent was often observed in pachytene, diakinesis and first meiotic metaphase. In pachytene the XY bivalent is enclosed within the sex vesicle and not infrequently it can be seen quite distinctly