Cytological Evidence for a Spontaneous Chromosome Translocation in the Domestic Fowl

Statistical evidence in the form of approximately 50% early embryonic mortality in a line of Single Comb White Leghorn fowl led to the hypothesis of a dominant lethal[1] as an explanation for this embryonic mortality. This dominant lethal was later postulated to be a chromosomal aberration, likely a reciprocal translocation[2]. Homozygous carriers of the aberration have twice been isolated from matings of heterozygous carriers.

Identification of genotype was achieved by mating a number of normal females, usually 4, to a given male through artificial insemination. The eggs were incubated for a period of 6–7 days and then broken out to determine whether the embryos were normal or aberrant. Only males identified as heterozygous after examination of at least 12 fertile eggs with a clear-cut ratio of 50% abnormal were investigated. Only male karyotypes were investigated because male birds are readily and certainly identified as heterozygous or homozygous, whether normal or aberrant.

Recent advances in cytological techniques[3,4] made it feasible to undertake a chromosomal study in order to find cytological proof for the suspected reciprocal translocation.

In studying the mitotic chromosomes leucocytes were cultured and slides prepared in a manner similar to that developed by MOORHEAD et al.[5] for studying human chromosomes.

Examination of karyograms from these slides showed that there was no demonstrable difference between normal birds and suspected translocation heterozygotes. This result suggested that if the lethal dominant was truly a reciprocal translocation, the portions exchanged were either of nearly the same length or involved only a very small part of each chromosome.

For examination of meiotic chromosomes fresh testicular tissue was obtained from birds suspected of being heterozygous for the translocation. This tissue was placed in a 0.7% sodium citrate solution at 37 °C. A cloudy suspension of cells was obtained by cutting the testis into minute cubes and aspirating these cubes with a Pasteur pipette and bulb. After filtering through cheese cloth the suspension was centrifuged at 60 rcf for 5 min. The harvested cells were then fixed in acetic-alcohol. After several washings with fresh fixative the cells were resuspended in 45% acetic acid and drops of the suspension air-dried onto cover slips. Staining was in 0.5% acetic-orcein followed by dehydration through an alcohol series without xylol. Permanent slides were prepared by mounting the cover slips in Euparal.

The normal meiotic complement of macrochromosomes, as seen on slides prepared in this manner, is shown in Figure 1. In good preparations it was possible to identify, consistently, 4 of the 6 large elements. Two bivalents, the fourth and fifth largest, are so similar in length that their positive identification was always difficult. However, while it was difficult to tell these 2 bivalents apart it was always possible to distinguish them from the other 4 large bivalents. Figure 2 which illustrates meiotic chromosomes from birds heterozygous for the translocation shows chromosomes 2 and 3 associated as a quadrivalent.

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[2] P. E. BERNIER, Poultry Sci. *39*, 1234 (1960).
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[5] P. S. MOORHEAD, P. C. NOWELL, W. J. MELLMAN, D. M. BATTIPS and D. A. HUNGERFORD, Expl Cell Res. *29*, 613 (1960).

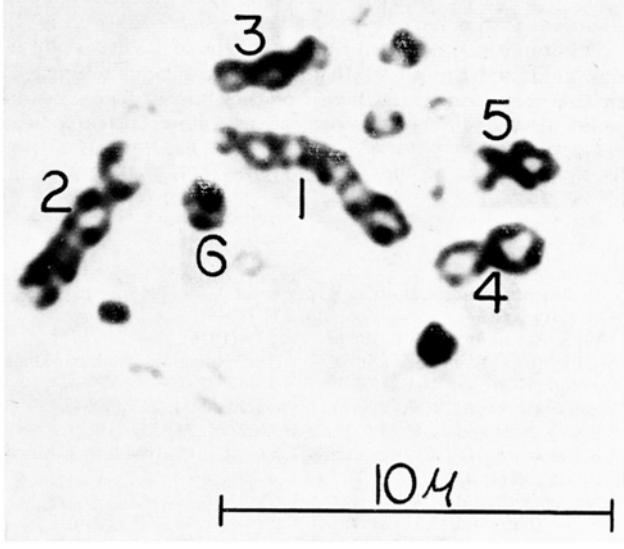

Fig. 1. Meiosis Prophase I chromosomes from normal male fowl.

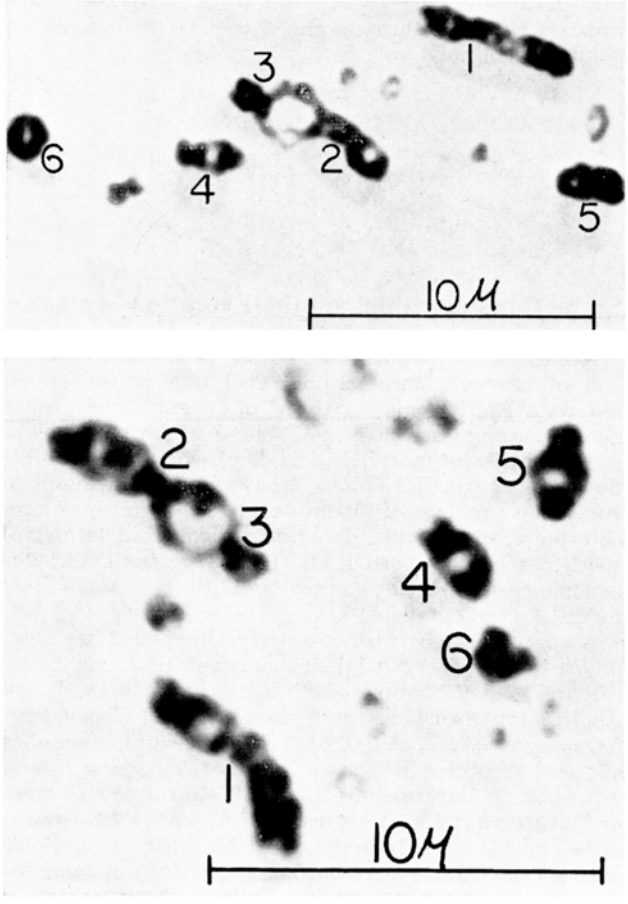

Fig. 2. Meiosis Prophase I chromosomes from male fowl heterozygous for translocation.

Because of the need to bring out the details of certain of the larger chromosome pairs, Figures 1 and 2 were printed in such a way as to emphasize the 6 or 8 largest bivalents. As a result, the smaller microchromosomes do not appear in these photographs. It was, however, possible to count 37–40 bivalents in these spreads when viewed through the microscope.

The opinion that chromosomes 2 and 3 are involved in this abnormal configuration was arrived at through careful examination of the quadrivalent as found in several birds and by comparison with the normal meiotic complement. The assumption was made that while some rearrangement of chromosomal material has occurred as a result of the translocation all macrochromosomes are still present in abnormal birds. With this assumption in mind, it was possible to identify 4 of the macrochromosomal bivalents (chromosomes 1, 4, 5 and 6). Chromosome 1 was identified on the basis of overall length and chiasmata frequency. Chromosomes 4 and 5 were identified by comparison with number 1 (each being less than half the length of the largest element), and by their appearance as either O- or figure-8-shapes. Chromosome 6 was identified by its size relative to chromosomes 4 and 5 and by its O-shaped appearance. Since chromosomes 1, 4, 5 and 6 were present in their normal bivalent condition it was concluded that chromosomes 2 and 3 were associated in the quadrivalent.

It has not been possible to make a precise determination of which arms of the 2 chromosomes are involved in the exchange nor of the extent of the translocation. However, based on study of the mitotic chromosomes, we believe that the short arm of chromosome 2 and the long arm of chromosome 3 have exchanged material. It also appears that the chromosomal segments exchanged are similar in length.

This is thought to be the first spontaneous chromosomal translocation to be demonstrated in the domestic fowl. By studying the effect of this translocation together with the X-ray induced translocation between chromosomes 1 and 2 described earlier[6] it should now be possible to establish a relationship between 3 of the 6 known linkage groups of the fowl and their residual chromosomes. If such relationships can be established the value of the domestic fowl as an experimental animal in genetic studies should be enhanced. Further, since the chromosomal aberration here described has been isolated in the homozygous condition it may be of some value in commercial breeding operations[7,8].

Résumé. La première translocation chromosomique spontanée observée chez le coq domestique implique un échange réciproque entre les chromosomes 2 et 3.

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Department of General Science and Department of Poultry Science, Oregon State University, Corvallis (Oregon 97331, USA), 29 January 1968.

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Some Considerations on the Evolution of the Karyotype of Microchiroptera

According to previous works of MATTHEY and BOVEY[1], and of BOVEY[2], chromosomal evolution in Chiroptera follows a Robertsonian pattern[3] of centric fusion. Since new techniques are now available[4,5] which can show a larger number of morphological details than was possible by means of gonadal squash, I have made an attempt to study the problem of evolution of karyotype in Microchiroptera, adding more data about the diploid number of species as yet unreported, and improving the knowledge in some species already known by addition of some more morphological details.

A comparison between the karyotypes of 3 Rhinolophidae[5–7] points out their morphological similarity (Figure 1). An example of centric fusion can be drawn from a comparison between the karyograms of *Rhinolophus ferrumequinum* and of *R. hipposideros*. The diploid number of these 2 species differs by 2 units (*R. ferrumequinum* $2n = 58$; *R. hipposideros* $2n = 56$), but a pair of large metacentric autosomes is present in *R. hipposideros* and is to be considered as brought about by centric fusion of 2 pairs of acrocentric large chromosomes. It is of some interest to underline that the peculiarly shaped chromosomes seen in *R. ferrumequinum* (2 pairs of small metacentric chromosomes and 1 pair of acrocentric ones with a heterochromatic zone) are present and morphologically identical in the karyotype of *R. hipposideros* (Figure 2).

A comparison between the karyotype of 4 Vespertilionidae indicated the possibility of centric fusions (Figure 3) in the evolutionary pathway of this family too. The diploid number in these 4 species is a follows: *Miniopterus schreibersii*[2,8] $2n = 46$; *Pipistrellus kuhli*[9] $2n = 44$; *P. savii*[10] $2n = 44$; *Barbastella barbastellus*[2,11]: $2n = 32$.

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