The table shows that the average SCE value per cell was extremely similar regardless of the diploid number. It has been known that the nuclear DNA content of mammalian cells is very similar. By measuring the total area of metaphase chromosomes of 8 mammalian species (diploid number from 18 to 78), Ohno et al., found similar values in all of these. Some of the species studied (man, cattle, mouse, cat) are the same used in the present study. Thus chromosomes merely represent packages of a given amount of genetic material, i.e., lower diploid number means larger packages and higher diploid number means smaller packages. In most mammalian karyotypes, the variability in the amount of heterochromatin is not expected to significantly alter this conclusion.

Our data suggest that the rate of SCE under identical experimental conditions (same concentration of BrdU; complete darkness) is perhaps determined by the genomic size, especially the amount of euchromatin. It has been found by several investigators 10,11 that constitutive heterochromatin has lower frequencies of SCE per unit chromosome when compared to that of euchromatin. Thus it is possible that the species with a high amount of heterochromatin may show a lower SCE rate relative to the genome size, but such cases are not very frequent.

Very recently, attempts to induce SCE in cells in vivo have been successful. Interestingly, the SCE rate of cells in vivo has been generally lower than that of cells in culture. For example, Allen and Latt<sup>12</sup> reported an average of less than 2 (1.81) SCE per spermatogonial metaphase

of the mouse and Vogel and Bauknecht<sup>13</sup>, about 4 SCE per cell. Schneider et al.<sup>14</sup> reported 4.1 SCE per cell in mouse and about 7 in rat and Pera and Mattias<sup>15</sup> found 1 SCE per metaphase plate in Microtus agrestis. The lowest SCE rate (0.75 per metaphase plate) was reported by Bloom and Hsu <sup>16</sup> in chicken embryos. This discrepancy in SCE rate between the in vivo and the in vitro systems may be due to a number of factors (lack of light exposure, rapid debrominization of BrdU, etc.).

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## Linkage between two loci controlling colour polymorphism in the colonial ascidian, Botryllus schlosseri

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Summary. In the colonial ascidian, Botryllus schlosseri, 2 loci controlling colour polymorphism are closely linked. They represent the nucleus of the first linkage group of this species. Recombination occurs in both male and female gametogenesis.

Several Mendelian loci are known to control colour polymorphism in Botryllus schlosseri<sup>2,3</sup>. 4 of them, which were studied in more detail<sup>4</sup> and designated as A, B, Bl, R, are responsible for the presence of the double intersiphonal band of nephrocytes (B), and of the orange (A, 'Arancio'), blue (Bl), or reddish (R) pigments contained in different types of blood cells. The allele for presence dominates over the allele for absence at all these loci, except Bl. Locus A has proved to be independent of B, Bl, R; locus Bl is independent of A, B and R. The present report concerns the linkage between B and R.

Materials and methods. A colony of B. schlosseri consists of a clone of zooids belonging to 3 sequential blastogenic generations, the adults and 2 generations of buds. A weekly change of generation takes place at a temperature of 18°C involving: resorption of the adults, maturation of the buds of the older generation and initiation of a new generation. The zooids are interconnected by a vascular network running in the common tunic. In so far these connections are maintained, the zooids of each generation keep at the same developmental and sexual stages and do, therefore, behave as a single individual. Self-fertilization within such a clone is prevented by protogyny<sup>3,5</sup>: 1 colony crossed with another colony at

a different sexual stage acts as female and then male in sequence. The embryonic development takes place inside the maternal body; free-swimming larvae are released slightly before regression of the parental zooids.

Linkage between the pigmentation loci B and R in Botryllus schlosseri. Offspring derived from double backcrosses (BbRr $\times$ bbrr)

Cross	Offspring phenotypes					$\chi^{2}_{\mathrm{B}}$	$\chi^2_{ m R}$	$\chi^2_{ m B-R}$	d.f.
No.	BR	Br	bR	br	total		~	<b>X</b>	
1	1	41	41	2	85	0.0	0.0	73.4	1
2	0	21	32	1	54	2.7	1.8	50.1	1
3	2	20	31	0	53	1.5	3.2	45.3	1
4	0	29	23	0	52	0.7	0.7	52.0	1
5	0 -	21	23	0	44	0.1	0.1	44.0	1
6	0	11	10	0	21	0.0	0.0	21.0	1
Total									
series	3	143	160	3	309	0.9	0.9	285.5	1
All families						5.0	5.8	285.8	6
Heterogeneity						4.1	4.9	0.3	5

Colonies heterozygous at the 2 loci B and R were testcrossed to double recessive colonies (bbrr). The larvae were collected directly from the donor; at the time of metamorphosis they were attached to glass slides; the new colonies derived from them were reared in aquaria and scored for pigmentation characters.

Results. The distribution of the phenotypes in the offspring of 6 matings is illustrated in the table. The segregation of the alleles at either locus fits the expected 1:1 ratio. The joint segregation reveals a tight linkage between B and R, with the dominant alleles in the repul-

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sion phase. Actually, all but one of the heterozygous parental colonies were derived from 2 matings of the type  $Br/br \times bR/br$ .

For the total series, the recombination value is 1.94%  $\pm$  0.78. The 2 partial series derived from heterozygotes in female and male phase, respectively, were 1 BR, 79 Br, 90 bR, 3 br (total 173), and 2 BR, 64 Br, 70 bR, 0 br (total 136); they have similar recombination values of 2.31 and 1.47. The parental genotypes in family No. 5 were AaBbRr and aabbrr. The joint segregation of A-B (17 AB, 12 Ab, 12 aB, 11 ab) and A-R (12 AR, 17 Ar, 11 aR, 12 ar) agrees with the previously established independence of locus A from both B and R.

Discussion. Little is known of the genetics of ascidians. The colonial species Botryllus schlosseri has proved to be a suitable material which can be easily reared under controlled breeding conditions in the laboratory, where it gives clones that can be maintained for years. In addition to colour polymorphism, other characters are now under study. The linkage between the loci B and R, with recombination in both sexes, is the first to be discovered and represents the nucleus of the first linkage group

within the haploid set of 16 chromosomes of this species<sup>6</sup>.

## Considerations of karyotypic evolution within Vespertilionidae

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Summary. The karyotypes of each 2 species of Nyctalus and Murina are examined. It is assumed that the diploid number of vespertilionid ancestor was 44 with a fundamental number of 50 and that the mechanism of karyotypic evolution within subfamily Vespertilioninae is mainly caused by centric fusion. On the other hand, the karyotypic alteration of subfamily Murininae may be evolved by non-Robertsonian translocation.

Some authors 3-6 have suggested that the diploid number (2n) of vespertilionid ancestor was between 44 and 50 with a fundamental number (FN) of 50, and that karyotypic alteration of the majority of genera in Vespertilionidae was mainly evolved by centric fusion. In this paper, we discuss the hypothetical karyotype of vespertilionid ancestor and suggest mechanisms of karyotypic evolution in this family, and compare the karyotypes of some vespertilionid bats studied by us with those of other species reported so far. The technique used in this study was that described by Uchida and Ando7. For chromosomal classification the method of Patton  $^8$  was adopted. Karyotypes of 16 Japanese species are listed in the table, and out of them the karyotypes of 6 species are represented in figure 1. In figures 2 and 3 are shown respectively the karyotypes of 2 Japanese noctule bats (Nyctalus furvus and N. lasiopterus) and 2 Japanese tube-nosed bats (Murina aurata and M. leucogaster) whose karyotypes are reported for the first time or not yet in detail.

From the facts, as shown in the table and figure 1, and as already described by some authors 3-6, that the FN values are more constant than the 2n ones, it is assumed that the mechanism considered responsible for karyotypic evolution in this family is mainly Robertsonian translocation (centric fusion or fission) which leads to formation of biarmed elements from uniarmed ones or its reverse. Some bat workers 9-11 suggested on the basis of morphological characters that the vespertilionid ancestor might be a Myotis-like bat. Taking their opinions into consid-

eration, it seems that the karyotypic evolution in this family is mainly attributed to centric fusion.

A case of centric fusion may be found within genus Nyctalus. The karyotype of N. furvus (figure 2a) resembles closely to those of Myotis (figure 1a). And similar karyotypes are found in European species of Pipistrellus (e.g., P. nathusii, P. kuhli and P. savii) 12-15, too. Con-

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