Mamestra brassicae wurde bereits 1970 von Aplin und Birch[5] untersucht. Sie fanden nur eine nicht näher identifizierte Verbindung vom Molekulargewicht 166, die jedoch bei unseren Untersuchungen nicht auftrat. Bei Mamestra persicariae fanden wir neben den bereits angegebenen Verbindungen **1** und **4**[5] noch Phenylacetaldehyd (**3**) und Spuren Benzylalkohol (**2**).
Das Gaschromatogramm der Duftstoffe von Polia tincta, dessen Zusammensetzung dem der beiden Mamestra-Arten ähnelt, unterscheidet sich von dem aller anderen untersuchten Tieren charakteristisch durch das Vorhandensein des Signals der später eluierten Verbindung mit Molekulargewicht 164. Dieser Substanz, die möglicherweise terpenoide Struktur besitzt, muss der blütenartige Geruch der Duftpinsel von Polia tincta zugeschrieben werden.

Rate of sister chromatid exchanges in mammalian cells differing in diploid numbers[1]

S. Pathak, Oscar G. Ward[2] and T. C. Hsu

Department of Biology, The University of Texas System Cancer Center M. D. Anderson Hospital and Tumor Institute, Houston (Texas 77030, USA); and Department of Ecology and Evolutionary Biology, The University of Arizona, Tucson (Arizona 85721, USA), 1 December 1976

Summary. The rate of sister chromatid exchanges (SCE) under identical experimental conditions is the same in various mammalian species irrespective of their diploid chromosome numbers.

Visualization of sister chromatid exchanges (SCE) by bromodeoxyuridine (BrdU) treatment and Hoechst 33258 fluorescence[3] has been a well-established procedure. Modifications of the procedure for Giemsa staining[4-6] have made it convenient to examine SCE with light microscopy. Investigators have used 3 principle cell materials for the SCE analyses: human (2n = 46), Chinese hamster (2n = 22) and the Indian muntjac (2n = 6,7). It appears that under similar experimental conditions the rate of SCE per metaphase was similar even though the diploid numbers are quite different[7].
We conduced a series of experiments using mammalian cells with diploid numbers ranging from 7 to 70. All cultures were treated with BrdU (10 μg/ml) for 2 cell cycles in the dark at 37 °C. The cells were harvested after a 1-h colcemid (0.05 μg/ml) and the usual hypotonic solution treatments. Air-dried preparations were treated for SCE by either the procedure of Korenberg and Freedlender or Pathak et al.[5,6]. The figures 1 and 2 show sister chromatid exchanges in Indian muntjac (2n = 7♂) and caribou (2n = 70) metaphase plates, respectively.

Figures 1 and 2. Trypsin-treated metaphase spreads showing sister chromatid exchanges after two rounds of BrdU labelling. Fig. 1. Metaphase plate of a male Indian muntjac (2n = 7). Fig. 2. Partial metaphase plate of a male caribou (2n = 70). ×1200.

Frequency of sister chromatid exchanges (SCE) in cultured fibroblasts of various mammalian species after 2 rounds of BrdU replication

Species	2N	Range of SCE	No. of cells	Average SCE/cell	References
Muntiacus muntjac (Indian muntjac)	7♂	5–15	25	8.8	*
Microtus montanus (Montane vole)	22	5–12	25	7.9	*
Cricetulus griseus (Chinese hamster)	22	5–13	25	8.6	*3[4,17]
Felis nigripes (Black-footed cat)	38	5–15	25	9.4	*
Mus musculus (Swiss mouse)	40	6–17	25	9.8	*[18]
Homo sapiens (human)	46	–	25	9.3	[19–22]
		–	20	8.1	
		–	90	4.2	
		–	50	6.5	
Bos taurus (cattle)	60	5–14	25	7.8	*
Rangifer tarandus (caribou)	70	7–13	25	9.2	*

*This paper.

1 Supported in part by Research grants VC-21 from American Cancer Society and DEB-76-10580 from National Science Foundation.
2 Department of Ecology and Evolutionary Biology, The University of Arizona, Tucson, Arizona 85721, USA.
3 S. A. Latt, Proc. nat. Acad. Sci., Wash *70*, 3395 (1973).
4 S. Wolff and P. Perry, Chromosoma, Berl. *48*, 341 (1974).
5 J. R. Korenberg and E. F. Freedlender, Chromosoma (Berl.) *48*, 355 (1974).
6 S. Pathak, A. D. Stock and A. Lusby, Experientia *31*, 916 (1975).
7 S. M. Galloway and H. J. Evans, Cytogenet. Cell Genet. *15*, 17 (1975).

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[8]. By measuring the total area of metaphase chromosomes of 8 mammalian species (diploid number from 18 to 78), Ohno et al.[9], 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[12] reported an average of less than 2 (1.81) SCE per spermatogonial metaphase of the mouse and Vogel and Bauknecht[13], about 4 SCE per cell. Schneider et al.[14] reported 4.1 SCE per cell in mouse and about 7 in rat and Pera and Mattias[15] found 1 SCE per metaphase plate in Microtus agrestis. The lowest SCE rate (0.75 per metaphase plate) was reported by Bloom and Hsu[16] 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.).

8 H. Reed and R. N. Jones, Int. Rev. Cytol. *32*, 53 (1972).
9 S. Ohno, W. Beçak and M. L. Beçak, Chromosoma, Berl. *15*, 14 (1964).
10 A. V. Carrano and S. Wolff, Chromosoma, Berl. *53*, 361 (1975).
11 T. C. Hsu and S. Pathak, Chromosoma, Berl., in press, 1976.
12 J. W. Allen and S. A. Latt, Nature, Lond. *260*, 449 (1976).
13 W. Vogel and T. Bauknecht, Nature, Lond. *260*, 448 (1976).
14 E. L. Schneider, J. R. Chaillet and R. R. Tice, Exp. Cell Res. *100*, 396 (1976).
15 F. Pera and M. Mattias, Chromosoma, Berl. *57*, 13 (1976).
16 S. E. Bloom and T. C. Hsu, Chromosoma, Berl. *51*, 261 (1975).
17 H. Kato, Nature, Lond. *251*, 70 (1974).
18 R. L. Brown and P. E. Crossen, Exp. Cell Res., in press, 1976.
19 K. Hayashi and W. Schmid, Humangenetik *29*, 201 (1975).
20 S. A. Latt, G. Stetten, L. A. Juergens, G. R. Buchanan and P. S. Gerald, Proc. nat. Acad. Sci., Wash. *72*, 4066 (1975).
21 H. Kato and H. F. Stich, Nature, Lond. *260*, 447 (1976).
22 R. C. Miller, M. M. Aronson and W. W. Nichols, Chromosoma, (Berl.) *55*, 1 (1976).

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

A. Sabbadin[1]

Istituto di Biologia Animale, Università degli Studi di Padova, via Loredan 10, I-35100 Padova (Italy),
29 December 1976

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[2,3]. 4 of them, which were studied in more detail[4] 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[3,5]: 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 × bbrr)

Cross No.	Offspring phenotypes BR	Br	bR	br	total	χ^2_B	χ^2_R	χ^2_{B-R}	d.f.
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