

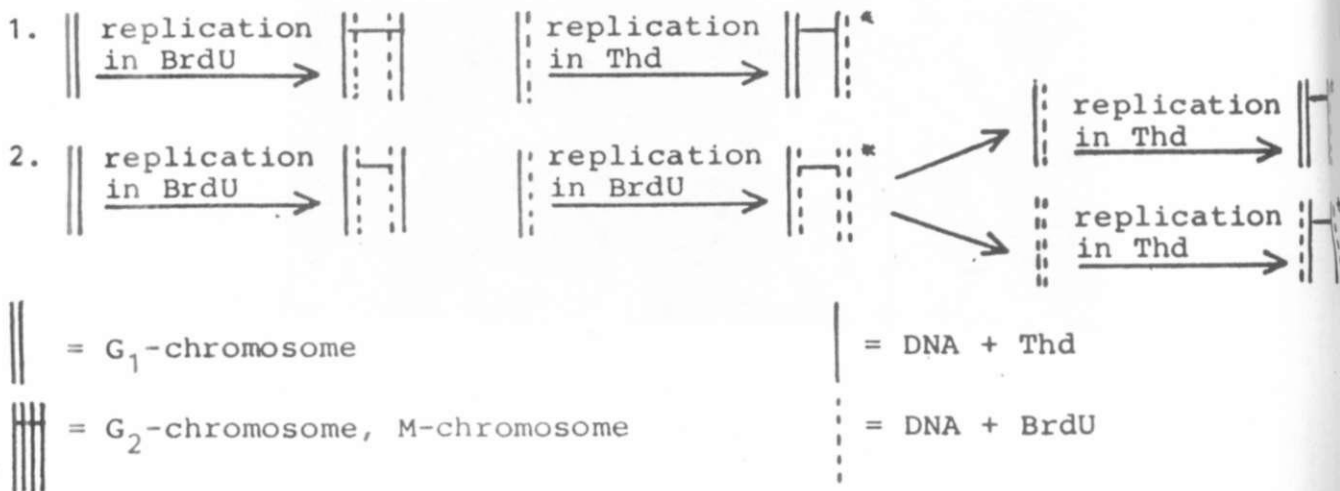
SISTER CHROMATID EXCHANGE (SCE) IN PISUM SATIVUM

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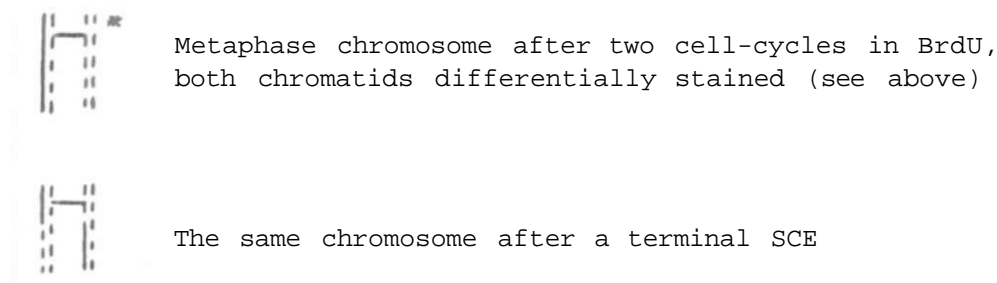
After application of BrdU (Bromdesoxyuridine) spontaneous sister chromatid exchange (SCE) was first demonstrated by Latt (2) in human chromosomes. This method subsequently was improved, and SCE was observed in a great number of mammalian chromosomes as well as in plant cells (*Vicia faba* [1], *Allium cepa* [3], and others). Recently, we could show SCE in *Pisum sativum* root tip chromosomes. We applied the method of Kihlman and Kronberg (1) with a few modifications. Root tips were exposed to BrdU concentrations of  $10^{-4}$  M and  $10^{-6}$  M in tap water. To increase the number of metaphases isovalerylurea was used instead of colchicine. The tissue was macerated in a solution of pectinase (5%) and cellulase (1%). After staining with Hoechst 33258 and Giemsa the two chromatids of the metaphase chromosomes were differentially stained and SCE's could be detected (Fig. 1.1 and 1.2-1.4, arrows indicating SCE's). We investigated two series of preparations:

1. Root tips were exposed to BrdU during one cell cycle, followed by one cycle in Thd (Thymidine).
2. Root tips were exposed to BrdU during two cell cycles, followed by one cycle in Thd.

All solutions were made with tap water, Schematically, the incorporation of BrdU results in the following chromosomes:



The above scheme is based on the assumption that a chromosome is built up on one DNA element. Metaphase chromosomes constructed according to the scheme labelled with (\*) should show differential staining and SCE's; in those labelled with (+) we should find the results of an SCE occurring during the preceding cell cycle, schematically as follows:



After mitosis this chromosome gives two types of chromosome, and after a further cycle in Thd, these result in metaphase chromosomes which show a discontinuous staining in only one chromatid (cf. Fig. 1.5), and schematically:



The incorporation of BrdU influences the length of the chromatids as Fig. 1.5 shows. We did not make any measurements so we cannot decide whether it shortens or lengthens the respective element.

Several questions result from the application of this method. Because of a low mitotic index in *Pisum sativum* root tip meristems and the fact that only a small number of well stained chromosomes is available, we are not yet able to count the SCE's per metaphase. For the same reasons, we are not able to decide whether homologous chromosomes have an identical SCE pattern, or whether a special pattern is characteristic for a specific chromosome. Yet the available material seems to point to the fact that the homologues are not identical in this respect, and this would mean that the pattern is random.

In addition, the results could provide a basis to investigate the question whether a metaphase chromosome of *Pisum sativum* is composed of only two chromatids or of two chromatids which are subdivided into half-chromatids, a question currently under discussion. C-banded metaphase chromosomes of this material exhibit a subdivision of certain heterochromatin blocks, which suggest that the chromatids are subdivided (Fig. 2, arrows). If this is true, then we should find in connection with our SCE experiments exchanges within a single chromatid. Some of the patterns found agree with this assumption (Fig. 1.6). Further investigations are required.

1. Kihlman, B. A. and D. Kronberg. 1975. *Chromosoma* 51:1-10.
2. Latt, S. H. 1973. *Proc. Nat. Acad. Sci.* 70:3395-3399.
3. Schwartzman, J. B. and F. Cortes. 1977. *Chromosoma* 62:119-131.

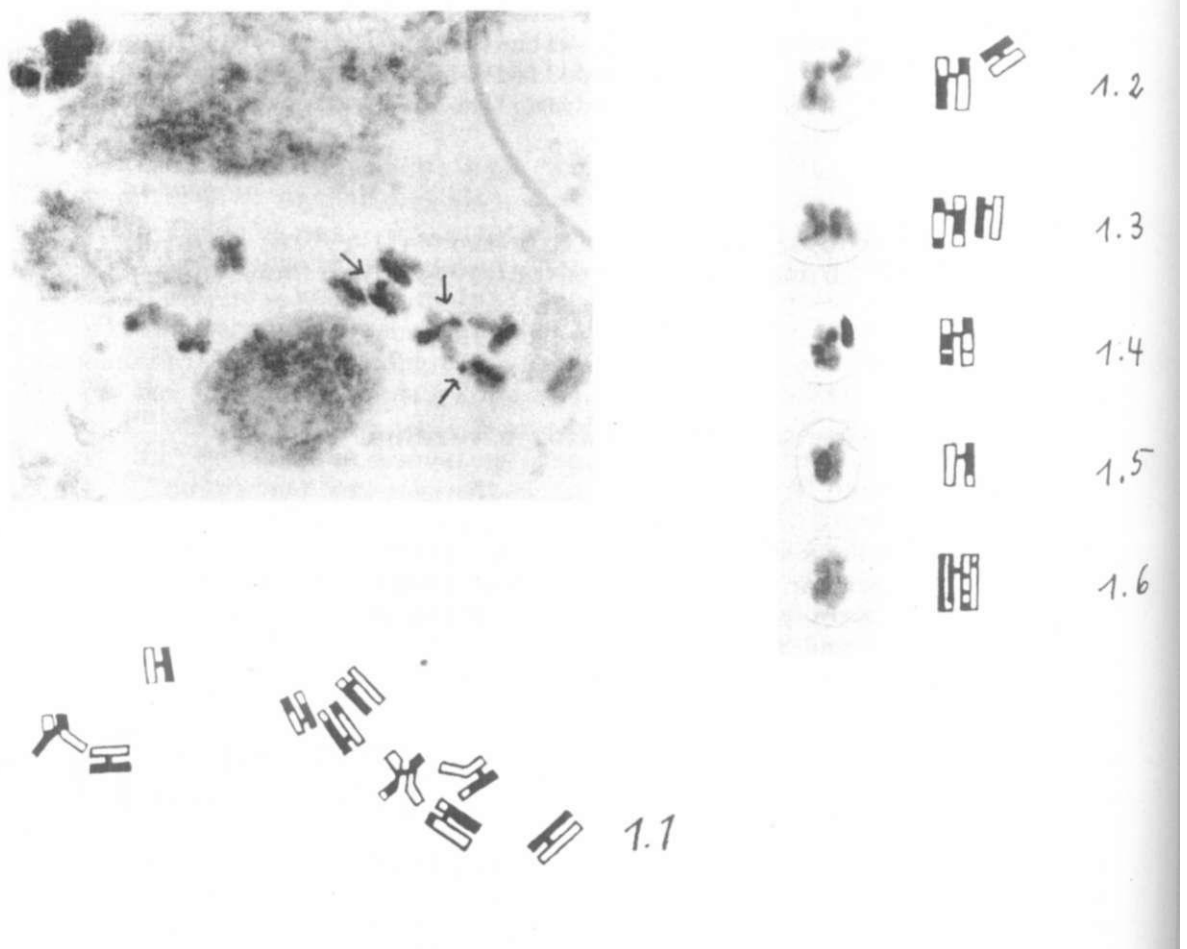


Fig. 1. Differentially stained metaphase chromosomes of Pisum sativum after BrdU incorporation; arrows point to SCE's. See text for details.

Fig. 2. C-banded metaphase chromosomes of Pisum sativum; arrows point to subdivided heterochromatin blocks.



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