IDENTIFICATION AND LOCALIZATION OF A TRANSLOCATION IN PISUM CHROMOSOMES BY THE C-BANDING TECHNIQUE

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Gottschalk's mutant collection in Bonn initially included, among others, 120 translocation genotypes. They originated in 1954 from radiation experiments with cv 'Dippes Gelbe Viktoria'. The translocation lines were selected by analyzing pollen sterility. From these 120 genotypes only 30 are now still available.

In the early work, eight translocation genotypes were investigated by examining the mitotic chromosomes of the root tips. Homogeneously stained chromosomes were measured and the values of the mutant lines were compared with those of the controls. The deviations in total length and arm indices were the basis of these investigations (2, 6). Though the participating chromosomes could be identified, the exact localization of the exchange was impossible.

The development of the banding techniques in the early seventies seemed to provide a promising tool for overcoming some of the technical problems. The C-banding method is especially useful for identifying the heterochromatic regions in plant chromosomes. However, this technique proved difficult in Pisum. Attempts in our institute succeeded only in characterizing chromosomes 4 and 7 (nomenclature according to Blixt [1]). The banding pattern corresponded to that published by Lamm in 1978 (3). Because of the difficulties, the investigations in our institute were discontinued. Lamm published some further results on banded Pisum chromosomes in 1981 (4).

In 1982 our institute again took up the problem of banding Pisum chromosomes. The method used is a modification of those described by Lamm (4), Marks and Schweizer (5), and Schweizer (pers. comm.). This new attempt has yielded a heterochromatin pattern by which the single chromosome can more or less be identified. Yet, this time too, many difficulties had to be overcome. Typically fewer than 10% of the slide preparations could be analyzed. This in part is due to an extremely Low and varying mitotic index of Pisum root tips, and in part to the method itself. Only in some few cells is the banding pattern sufficiently clear. In general either the chromosomes are homogeneously stained, or the pattern is incomplete, or the chromosomes are extremely swollen and unstained. Often all these shortcomings are found on the same slide, and we are unable to give a satisfactory explanation for them.

The distribution of heterochromatin in normal Pisum chromosomes **is** shown in Figs. 1 and 2. Heterochromatin is found in the centromere regions and on the telomeres, the amount being different in different chromosomes. A rather large quantity of heterochromatin is situated near the NOR region of chromosomes 4 and 7. Few intercalary bands of heterochromatin are evident. However, banding clearly is visible on the long arm of chromosome 7 proximal to the centromere region, and also on the short arm of chromosomes 4 and 7 as well as near the centromere region of chromosome **3** (long arm) and on the long arm of chromosome 5 (Fig. 2). This pattern is found in all metaphases which can be analyzed reliably.

We have applied this C-banding technique to a reciprocal translocation genotype. We knew from earlier investigations of this genotype that chromosomes 5 and 7 are involved in the exchange processes. Fig. 3

shows the metaphase of a translocation heterozygote, and Fig. 4 shows the translocation chromosomes schematically. A long metacentric chromosome, normally not present, attracts attention in Fig. 3 (arrow). The analysis of this chromosome reveals that part of the long arm of chromosome 7 is involved, characterized by the prominent banding near the NOR region. On the second arm a small banding proximal to the centromere region is visible, this being characteristic for the long arm of chromosome 5. The second translocation chromosome is an extremely small one (Fig. 3, arrow). The one arm is identical with the short arm of chromosome 7. The shortness of the other arm reveals that only a very small part of chromosome 5 is translocated.

As demonstrated above, chromosome 7 is characterized by a clear intercalary banding on the long arm (Fig. 2). This banding is not found in both the translocation chromosomes. This therefore marks the breaking point of chromosome 7. The heterochromatic region evidently was lost during the exchange process. With this presupposition, and by measuring the arm length, we can in addition identify the breaking point of chromosome 5. Thus, the one translocation chromosome, the big one, is composed of the largest part of chromosome 5, including the centromere region and a section of chromosome 7, this being exactly that part from the position of the former intercalary banding on the long arm to the end, including the satellite. The second translocation chromosome, the short one, possesses the short arm of chromosome 7, the centromere region and the adjacent part, up to the former intercalary banding. From chromosome 5 only a very small terminal piece is translocated (Fig. 4).

Thus, our findings have shown that the application of the C-banding technique in connection with translocation problems is useful; yet the measurement of chromosomes seems also to be necessary. Because of the low number of bandings in the Pisum chromosomes it seems impossible to analyze any type of translocation. Only in those instances where a region with a characteristic pattern is involved can detailed analysis give information on the exact point of exchange.

1. Blixt, S. 1958. Agri Hort. Genet. 16:221-237.
2. Klein, H. D. 1974. PNL 6:22-23.
3. Lamm, R. 1978. PNL 10:31-32.
4. Lamm, R. 1981. Hereditas 94:45-52.
5. Marks, G. E. and D. Schweizer. 1974. Chromosoma 44:405-416.
6. Muller, D. 1976. Caryologia 29:217-225.

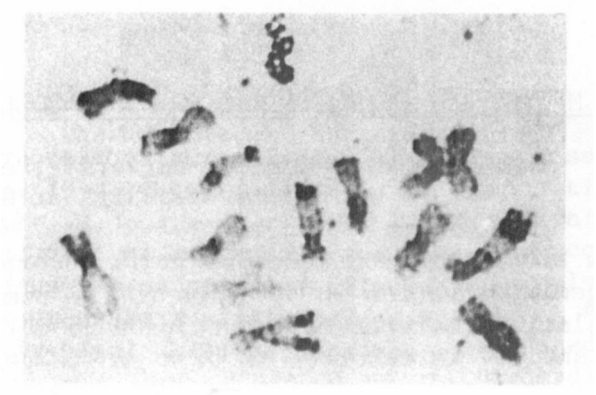

Fig. 1. C-banded metaphase of the root tip of Pisum sativum, var. 'Dippes Gelbe Viktoria'.

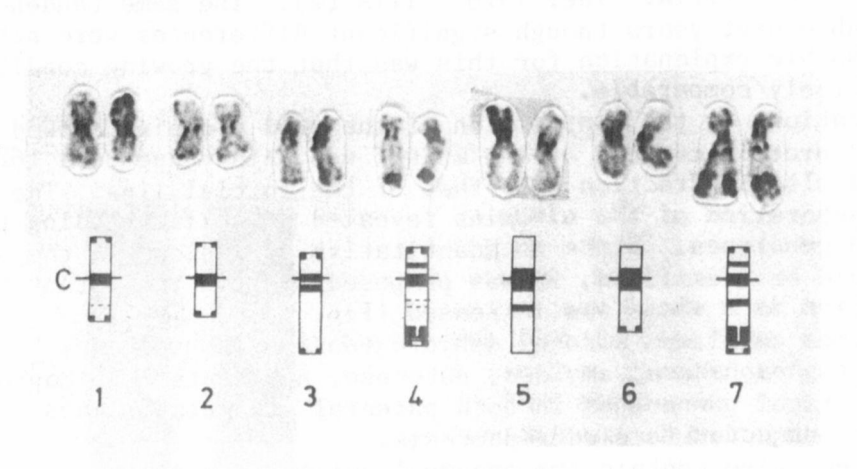

Fig. 2. Karvogram of C-banded pea chromosomes (DGV); below a schematical drawing; C=centromere

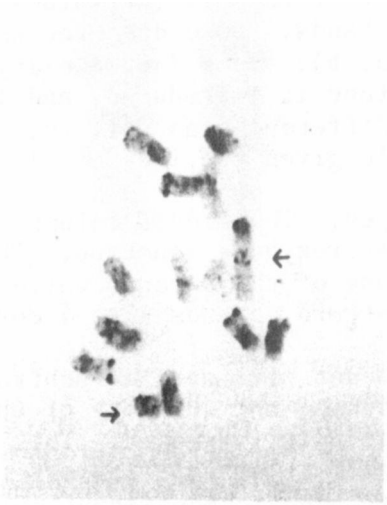

Fig. 3. C-banded metaphase of a translocation heterozygous line (T198) of DGV, arrows pointing to the two trans-chromosomes.

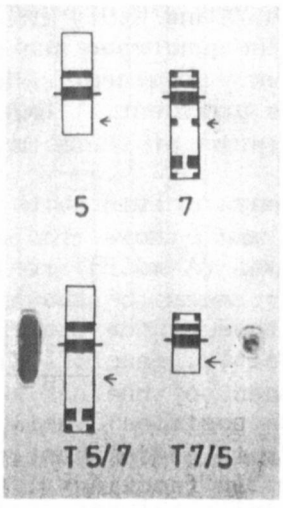

Fig. 4. Schematical drawing of the two translocation chromosomes. Below, normal chromosomes; above arrows pointing to the region of breakage and reunion.