INTROGRESSION OF RESTORER GENES FROM RAPHANUS SATIVUS INTO CYTOPLASMIC MALE STERILE BRASSICA NAPUS AND THE GENETICS OF FERTILITY RESTORATION

By Fritjof W. Heyn
Inst. of Plant breeding
Theodor Heuss-Str. 18
34 Göttingen

In Raphanus sativus Ogura (1968) detected a system of cytoplasmicgenic male sterility, which can be used for F_1-hybrid seed production. By successive backcrosses Bannerot et al. (1974) introduced the nucleus of Brassica oleracea and B. napus into the S-Raphanus cytoplasm. The resulting plants are completely male sterile. This material was kindly supplied to me by Bannerot in February 1975.

As Brassica napus is widely used as a grain crop, restorer genes are necessary for having complete male fertility in the F_1-hybrids.Test-crosses with a great number of different B. napus cultivars and accessions resulted in male sterile offspring only. Thus all euplasmic forms are functionally maintainers for this form of male sterility. The only way of establishing restorer lines in B. napus is to introgress restorer genes from Raphanus. They are rather frequent in the European radish cultivars as shown by the work of Bonnet (1975). As polyploids cross more readily than diploids, Raphanobrassica forms were chosen for the intergeneric gene transfer. A hexaploid form (2n=56, AACCRR),white flowered and selffertile, obtained from Bannerot, originally produced by Chopinet, prooved to be the best pollinator. Seed set on the cms B. napus plants was poor, but as several thousand pollinations could be done without great expenditure, the quantity of B. napus-Raphanobrassica-hybrids was reasonably high; 22 successfully pollinated cms plants gave rise to 331 male fertile white flowered hybrids, the genome constitution of which will be AACCR. The flowers of the hybrids exhaled a strong fragrance and the leaves were dark green. These characteristics are improved compared with those of the cms plants, which show a slight chlorophyll deficiency and a faint fragrance.

The male fertile S-plasm B. napus-Raphanobrassica-hybrids were backcrossed as pollen parents to cms B. napus in order to eliminate the superflouos R chromosomes not carrying restorer genes. As there is a partial homology between R, C and A chromosomes (Fukushima, 1945; Mizushima, 1968) a substitution by the restorer carrying chromosomes should be possible. An example of an introgression of two chromosome pairs or parts of them is given by the white flowering B. napus, produced by Stefansson in Winnepeg, Canada. This characteristic is conditioned by two complementary dominant genes (Heyn, unpubl.), which are introduced from a Raphanobrassica.

One of the resulting male fertile plants TR 163-9 was a very efficient pollinator; the seed set produced on a number of cms plants was; one plant with 13 seeds per siliqua, five with 10, eight with 5 to 9 and three plants with less than five seeds per siliqua. The pollinator itself did not set any seed on selfing or crossing with any other plant.

All crosses with cms plants and a restorer plant, which itself is derived from such a kind of cross, give rise to backcross progenies, the segregation ratios of which can be interpreted rather easily. The first striking observation is that of the absence of yellow flowered male fertile segregants. White flowered plants can be male fertile or male sterile.

Segregations for male fertility : male sterility can be explained by assuming the action of two complementary dominant factors Rf_1 and Rf_2. As there are no yellow male fertile plants the white flower characteristic is a prerequisite for the Rf-genes to function. Rf_1 is linked to W_1, one of the genes for white flower colour. From the segregation ratios obtained and from its derivation it became clear that the pollinator TR 163-9 was heterozygous for all the genes conditioning white flower colour and restoration of male fertility. Its genotype is: W_1Rf_1/w_1rf_1 W_2/w_2 Rf_2/rf_2. Crosses to yellow male sterile plants yielded segregations as follows:
24 white male fertile : 41 white male sterile : 157 yellow male sterile; two other ones 23 : 35 : 111 and 38 : 47 : 160. These segregation ratios represent 1 : 2 : 5 segregations which result from a cross W_1rf_1/w_1rf_1 w_2/w_2 rf_2/rf_2 x W_1Rf_1/w_1rf_1 W_2/w_2 Rf_2/rf_2. The segregation 56 : 43 : 186 may be a 3 : 3 : 10 from the combination W_1Rf_1/w_1rf_1 w_2/w_2 rf_2/rf_2 x the same above mentioned same pollinator genotype. The cross white male sterile x TR 163-9 resulted the following ratios: 30 : 84 : 75 : 15 : 18 : 34 and 51 : 30 : 83. Only the first one fits fairly well to an assumed 3 : 7 : 6 ratio. The others deviate considerably. Some of the other data not mentioned here are difficult to interpret. Deviations may be due to the small size of the progeny and/or aneuploidy of the pollinators, which have a low seed fertility on selfing and sibbing. The best restorer plant to the time being has about one fourth of a complete seed set. Another possible reason may be that additional genes could play a role in the inheritance of male fertility restoration like in Raphanus (cf. Bonnet, 1975).

Several generations of outcrossing, selfing and selection of restorer plants will be needed in order to stabilize these till now aneuploid plant on a 38-chromosomes level. The genetic data show that the necessary genes which are to be introgressed from the R genome are located on three chromosomes; this aim should be achievable without not too many difficulties. This expectation is sustained by the experience that fully seed fertile white flowering B. napus plants, derived from Stefansson's material could be obtained without any difficulty in a few generations only. This material gives an example of an introgression of two chromosomes or chromosome pieces from Raphanus into B. napus as evidenced by the genetic data. As for the restorer lines only an additional third gene pair is to be introgressed. This process may especially be furthered by crossing to euploid plant with a F-plasm. The dominant white flower colour can be used as a marker for the presence of the restorer gene Rf_1.

The problem of the slight chlorosis in cms B. napus due to the radish cytoplasm can be solved by selecting for chlorosisfree restorer lines.

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