Eight principal genotypes according to <u>rf</u>-factors are the basis of the proposed classification. They may be on N-, T- and S-type of cyto-plasm and they would show fertility or sterility depending on the relation-ships between the cytoplasm and the nucleus. The factors not designated in this short classification occur in recessive state.

This classification provides an indication of the type of cytoplasm and the state of <u>rf</u>-factors with every line and shows for what purpose it could be used in hybrid seed production on sterile basis.

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2. Elimination of hidden isolated heterozygous states of Rf-factors in sterile lines and single cross hybrids with the genotype Trf1rf1rf2rf2.

During the maintenance of the sterile lines 171, 144g, 0266a and Wf9 with the genotype $Trf_1rf_2rf_2$, fertile plants without vigorous growth appear systematically. The latter are due to mutation of the recessive factors rf_1 and rf_2 to the dominant factors rf_1 and rf_2 .

The occurrence of fertile plants could not be eliminated by the method of analyzing crosses between individual plants of the sterile and the fertile analogue. In these analyses, not only the ideal genotypes $\frac{\text{Trf}_1\text{rf}_2\text{rf}_2}{\text{Trf}_1\text{rf}_2\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_1\text{rf}_2\text{rf}_2}{\text{nre}} \text{ are considered pure but also the genotypes having single dominant factors are considered likewise since sterility occurs in them too, namely, <math display="block">\frac{\text{Trf}_1\text{rf}_2\text{rf}_2}{\text{Nrf}_1\text{rf}_1\text{rf}_2\text{rf}_2} \times \frac{\text{NRf}_1\text{rf}_1\text{rf}_2\text{rf}_2}{\text{Nrf}_1\text{rf}_1\text{rf}_2\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_1\text{rf}_2\text{rf}_2}{\text{Nrf}_1\text{rf}_1\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_1\text{rf}_2}{\text{Nrf}_1\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_1\text{rf}_2}{\text{Nrf}_1\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_2\text{rf}_2}{\text{Nrf}_1\text{rf}_2} \times \frac{\text{Nrf}_1\text{rf}_2}{\text{Nrf}_$

However, in the next stage of the maintenance of the line, crosses between the following genotypes are possible also: $\frac{TRf_1rf_2rf_2}{TRf_1rf_2rf_2} \times \frac{TRf_1rf_1rf_2rf_2}{TRf_1rf_1rf_2rf_2} \times \frac{TRf_1rf_1rf_2rf_2}{TRf_1rf_1rf_2rf_2} \times \frac{TRf_1rf_1rf_2rf_2}{TRf_1rf_1rf_2rf_2} \times \frac{TRf_1rf_1rf_2rf_2}{TRf_1rf_1rf_2rf_2} \times \frac{TRf_1rf_1rf_2rf_2}{TRf_1rf_2rf_2} \times \frac{TRf_1rf_2rf_2}{TRf_1rf_2rf_2} \times \frac{TRf_1rf_2rf_2}{TRf_1rf_2} \times \frac{TRf_1rf_2rf_2}{TRf_2rf_2} \times \frac{TRf_1rf_2rf_2}{TRf_1rf_2} \times \frac{TRf_1rf_2rf_2}{TRf_2rf_2} \times \frac{TRf_1rf_2r$

We succeeded in eliminating this undesirable phenomenon by selection of pure genotypes through analyzing crosses using two well differentiated testers with the genotypes $\underline{Rf_1}\underline{Rf_1}\underline{rf_2}\underline{rf_2}$ and $\underline{rf_1}\underline{rf_1}\underline{Rf_2}\underline{Rf_2}$, namely:

1. for cleaning of the fertile analogue:
TRf_Rf_rf_rf_2 x lNrf_rf_rf_rf_2 = 100% sterility
Trf_rf_Rf_Rf_2 x lNrf_rf_rf_rf_2 = 100% sterility

2. for cleaning of the sterile analogue after it has been pollinated with pollen from the already cleaned fertile analogue:

 $laTrf_{1}rf_{1}rf_{2}rf_{2} \times NRf_{1}Rf_{1}rf_{2}rf_{2} = 100\% \text{ sterility}$ $lbTrf_{1}rf_{1}rf_{2}rf_{2} \times Nrf_{1}rf_{1}Rf_{2}Rf_{2} = 100\% \text{ sterility}$

This method of cleaning applies also to sterile single cross hybrids with the same genotype when they produce undesirable fertile plants.

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3. A new method for determination of the degree of fertility in hybrids on sterile cytoplasm.

The methods used so far for determination of the degree of fertility determine it as percentage of fertile pollen against the total amount of pollen shed. The sterile stamens which were not extending outside the floweret were not taken into account. From a practical point of view it is more correct that fertility degree should be expressed as percentage of fertile pollen against the total amount of pollen which would be produced by plants with a normal cytoplasm. The only method meeting to a certain extent this requirement is the method of Galleev (CMS v selekcij i semenovodstve kukuruziy, Kiev, 1962).

The method utilized in our studies employs the following procedures: samples from 1000 flowerets are taken several days before or at the time of flowering of the tassels. The flowerets should be chosen from different plants and different parts of the tassels. Cross sections are made on 40 flowerets. The low part of the floweret held with a microscopic needle is observed under a stereo-microscope (25 times magnification). Data are taken on fertile, sterile and semi-sterile stamens. Assuming that the latter contain 50% of sterile pollen, the degree of fertility is determined by the percentage of fertile pollen in relation to the amount of pollen which would be produced by the tassels if all the stamens produce pollen normally.

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