

ADDITIONAL REMARKS ON THE PRODUCTION OF VIRUS-FREE CARNATIONS BY MEANS OF MERISTEM CULTURE¹

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Recently VAN OS (1964) gave a detailed description of the meristem culture technique (QUAK, 1957, 1961) applied to carnations. This work is now being carried out on a large scale by the Plant Protection Service at Wageningen for the production of carnations free from carnation mottle virus and carnation ringspot virus. In his paper VAN OS summarises research results of 1960 giving as an ultimate percentage of virus-free plants an average of 1.3 %. According to him the factors which may affect this percentage are: a. the effect of the season, b. varietal differences and c. differences between the material supplied by different growers. The object of this short communication is to furnish some results which indicate that the factors mentioned by VAN OS are probably of less importance than at least one other factor in our recent trials, viz., composition of the culture medium.

Information from Denmark and Belgium shows that between 30 % and 80 % of the meristem isolations made from carnation plants affected by viruses result in virus-free plants. These results are obtained using practically the same technique as we do. However, in Denmark and Belgium, the meristems are taken from vigorously growing mother plants, instead of from rooted cuttings as was our normal routine. With this in mind we changed over to using meristems from mother plants early this year, sticking, however, to the traditional medium described by MOREL (1948). During the first three months of 1964 some 1400 isolations were made from mother plants of five varieties and placed on MOREL's medium. At the end of July 1964 not one young plant had yet been transplanted. This result is in agreement with those from previous trials in which meristems of rooted cuttings gave only small numbers of plants after a six to ten months culture period *in vitro*.

In April we started to use a new medium, based on MURASHIGE's medium (MURASHIGE & SKOOG, 1962), modified according to BUYS (personal communication). Meristems originating from rooted cuttings as well as from mother plants were tested on this medium. In one trial we compared the effect of the old and the new medium on the growth of meristems taken from one batch of rooted cuttings of the variety 'William Sim', supplied by one grower. Ninety-six meristems from this source were placed on MOREL's medium, 216 on the new medium, and 144 on a batch of the new medium which by mistake contained five times the prescribed nutrient concentration. The meristems were isolated in April 1964. Already six weeks later the first plantlets could be transplanted into soil. At the end of July the following numbers of plants had been obtained: from the first group on MOREL's medium: one plant out of 96 (about 1 %), from the

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second group on the new medium: 54 plants out of 216 (about 25%) and from the third group on the too concentrated new medium: one plant out of 144 (about 0.6%). The high percentage of successful isolations on the new medium compared with the usual low percentage of successful isolations on MOREL's medium demonstrates the favourable properties of the modified MURASHIGE's medium. Although this trial has not yet been completed we may expect a continuation of the favourable tendency hitherto observed. The prospect of being able to raise young plantlets from meristems in six to eight weeks instead of the usual six to ten months or even longer is very attractive.

In another trial we isolated 384 meristems from the shoots of 'Crowley Sim' mother plants following the normal routine treatment, but using the new medium. Of this group 312 meristems were placed on the new medium, the remaining 72 being placed on the five times concentrated medium. This was done in April and after six to seven weeks only it was possible to transfer the first plantlets to soil. At the end of July we had already obtained 32 plants (about 10%) out of the original 384 meristems, all of them coming from the new medium, none from the concentrated medium.

In all other trials, comprising material of seven varieties from five growers, isolated in May, June and July, the same tendency of a higher percentage of growth in vitro on the new medium was observed, confirming our observation that by using the modified MURASHIGE-medium the percentage of successful isolations from meristems may be greatly increased. In our opinion the trial with the rooted cuttings of 'William Sim' has established that the medium on which the meristems are grown has a much greater influence on successful culture than has the origin of the meristems. Taking meristems from mother plants instead of from rooted cuttings did not result in better growth and a shorter growing period on MOREL's medium.

The combined effect of using the new medium and of mother plants is still under investigation and further details will be furnished in the near future.

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