QUANTITY PRODUCTION OF ANther-Derived Haploids FROM A MULTIPLE DISEASE RESISTANT TOBACCO HYBRID. I. FREQUENCY OF PLANTS WITH RESISTANCE OR SUSCEPTIBILITY TO TOBACCO MOSAIC VIRUS (TMV), POTATO VIRUS Y (PVY), AND ROOT KNOT (RK)1

L. G. BURK2, J. F. CHAPLIN2, G. V. GOODING3 and N. T. POWELL3

2 Tobacco Research Laboratory, Agricultural Research, Science and Education Administration, US Dept. of Agriculture, Oxford, N.C. 27565, USA
3 Department of Plant Pathology, North Carolina State University, Raleigh, N.C. 27650, USA

Received 2 June 1978

INDEX WORDS
Nicotiana tabacum, tobacco, haploids, doubled haploids, tobacco mosaic virus, potato virus Y, root knot nematode, resistance.

SUMMARY
Methods are described for producing large numbers of haploid plantlets from anthers of a flue-cured tobacco hybrid with monogenic resistance to tobacco mosaic virus (TMV), potato virus Y (PVY) and root knot (RK), respectively. Additional details are given on colchicine treatment for converting haploids to doubled haploids (DH’s) and on the frequency of spontaneous DH’s among untreated plantlets. Disparate genetic ratios of TMV-resistant to TMV-susceptible plants were obtained among colchicine-treated haploid plantlets, induced DH’s and untreated haploids when compared with F2 and BC1 progenies. Haploids (gametes) with the gene for TMV resistance occurred more frequently than expected and plantlets with the gene for RK resistance occurred less frequently than expected. Transmission of the gene for PVY resistance differed only slightly from Mendelian expectations. These unexpected ratios, in addition to the frequent occurrence of plastid chimeras among anther-derived plantlets, strengthened our conviction that haploidy is somehow associated with mutation.

INTRODUCTION

L. G. Burk, J. F. Chaplin, G. V. Gooding and N. T. Powell

Melchers & Labib, 1970; Melchers, 1974; Nakata, 1970; Nakata & Kurihara, 1972; Nitsch & Nitsch, 1969; Nitsch, 1972; Vagera et al., 1976). Progenies of individual doubled haploids (DH’s) show extraordinary phenotypic uniformity but interprogeny differences are equally extraordinary (Burk & Matzinger, 1976). Interprogeny differences are not unusual when the anther source is a hybrid, but they also were observed when the source was a 15-generation inbred variety of single-seed descent (Burk & Matzinger, 1976). Marked differences in characteristics among DH progenies suggest that anther-derived haploids exhibit a higher-than-normal rate of mutation (Burk et al., 1972; Burk & Matzinger, 1976).

Methods for converting haploids to DH’s are; 1), the application of colchicine to plantlets or shoots of larger plants (Burk et al., 1972; Jensen, 1974; Nakamura et al., 1975) or 2), the production of haploid callus and the regeneration of DH shoots (Collins & Sadasivaiah, 1972; Devreux & Laneri, 1974; Jensen, 1974; Kadotani, 1969). Kasperbauer & Collins (1972) described a callus culture method for producing adventitious DH shoots directly from leaf discs grown aseptically on an agar medium.

Most plantlets derived from anther culture are haploid, but some become DH spontaneously (Burk et al., 1972; Burk & Matzinger, 1976). Spontaneous DH’s and those from colchicine treatment behave in a similar manner when progenies are compared under field conditions. Burk & Matzinger (1976) indicated that interprogeny differences fall within a similar range of magnitude regardless of the manner in which the haploid was converted to a DH. Some workers have observed distinct differences among progenies of DH’s (Burk et al., 1972; Burk & Matzinger, 1976). Others have reported minimal or no differences (Collins et al., 1974; Collins & Legg, 1974; Collins & Legg, 1975). The observed differences among progenies of DH’s may be due to a release of residual heterozygosity from the haploid induction process. Haploids may also have enhanced mutagenic properties (Burk et al., 1972; Burk & Matzinger, 1976) or have undergone minor (cryptic) gains or losses of chromosomal material.

In this paper, we report on the genetic ratios in haploids and DH’s for resistance or susceptibility to tobacco mosaic virus (TMV), potato virus Y (PVY), and root knot nematode (RK), each of which is controlled by a single gene. Subsequent studies of resistance to other diseases, alone or in combination, may provide a better understanding of the reliability of the haploid method for investigations of this kind.

There was a basic rationale for choosing a hybrid with multiple disease resistance as the anther-source of haploids and for producing such a large number of plantlets. The potential multiplicity of gametes with different combinations of disease resistance factors could be large. Therefore, an increase in absolute numbers of plantlets would assure recovery of a testable quantity of DH’s with the same resistance genotype and also permit further selection of the phenotypically superior plants as varietal candidates or breeding lines. The recently reported development of varieties and breeding lines from Japan (Nakamura et al., 1974) and the People’s Republic of China (The Shantung Institute, 1976) offered considerable encouragement for the use of the haploid method in varietal development.

In this report we compared the effect of TMV inoculations on haploids and DH’s that had been treated with colchicine, on haploids that had not been treated, and on