

# INVESTIGATION OF DIHAPLOID POTATOES IN THE NETHERLANDS

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*Zusammenfassung, Résumé p. 67*

## SUMMARY

Two hundred primary dihaploids ( $2n = 2x = 24$ ) were obtained relatively easily from Dutch varieties and breeding stocks of the potato ( $2n = 4x = 48$ ). Male fertility is rare in primary dihaploids but increases rapidly with further crossing. It is thus possible to inbreed and cross-breed *Solanum tuberosum* at the diploid level. Tetraploids were obtained by colchicine treatment. Tetraploids were also obtained by backcrosses of dihaploids with tetraploids.

## 1. INTRODUCTION

The feasibility of breeding tetraploid plants at the diploid level has been established for *Cyclamen persicum* (LEGRO, 1959; WELLENSIEK, 1961) and for potato (HOUGAS and PELOQUIN, 1958, 1960; PELOQUIN and HOUGAS, 1959). The potential value of dihaploids for potato breeding was discussed by HOUGAS and PELOQUIN (1958), and a theoretical scheme for their use proposed by CHASE (1963).

The four years of breeding work reported here was initially stimulated by a paper read at Wageningen in 1960 by Dr. R. W. HOUGAS who subsequently kindly supplied to the late Dr. H. J. TOXOPEUS 18 potato dihaploids (U.S.-W. Series) and the pollen donor *Solanum phureja* P.I. 225-682.

## 2. INDUCTION OF NEW DIHAPLOIDS

Dihaploids were obtained from European varieties and home bred parental lines by the procedure of PELOQUIN and HOUGAS (1959). Initial screening was easy if the female parent lacked anthocyanin, the hypocotyl colour conditioned by the pollen parent *S. phureja* serving as a marker in the seedling population. Progeny from red-skinned female parents were first screened for leaf shape and low number of chloroplasts in the stomata – two of several distinguishing characters occurring in dihaploids (RÖTHACKER and SCHÄFER, 1961). The final selection was always made by chromosome counts of root-tip cells (SPECKMANN, 1965).

In 1961, 205 pollinations of 16 varieties by *S. phureja* yielded 137 berries giving, in the following year, approximately 2,600 seedlings of which 8 were dihaploids. In the following years further acceptable dihaploids were induced and there are now 200 available. The total number found was greater than this but some were rejected be-

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cause they failed to set tubers, were dwarfed or chlorotic, or were lost by virus infection or other pest and disease attack. Those retained have the following desirable qualities: resistance to Y, A and X viruses, wart disease and eelworm (biotype A); satisfactory field resistance to *Phytophthora infestans* and hypersensitivity to genes R<sub>1</sub>; R<sub>3</sub> and R<sub>1</sub>R<sub>3</sub>; red skin colour.

### 3. BREEDING AT THE DIPLOID LEVEL

Not all the dihaploids flowered; of those which did, female fertility, tested by pollinating them with *S. goniocalyx*, was generally good. In contrast, only one clone – U.S.-W. 4 – had reasonably fertile pollen. In 1961 it was used to pollinate U.S.-W. 7, U.S.-W. 42 and U.S.-W. 187, the progeny being so-called secondary dihaploids. Most of the 97 plants obtained formed flowers with good fertile pollen, some being self-fertile and the best being used for pollinating primary dihaploids. Although a few of these last crosses produced some chlorotic plants, the hybrids were usually vigorous in comparison with the marked inbreeding depression exhibited by progeny of self-fertilised secondary dihaploids or by a sib-cross (Fig. 1).

Progeny of the next generation of dihaploid crosses set abundant seed and it is thus clear that a breeding programme can be conducted at the diploid level. Some individuals in this generation were homozygous for R-genes (hypersensitivity to *Phytophthora infestans*) and segregation ratios agreed with expectations.

Wide variation in many characters, including tuber shape and eye depth, is found among primary dihaploids bred from the same potato variety and within each population of secondary dihaploids too. Like variation has been noted by other investigators such as JAKUBIEC (1964).

The commercial potential of this new material has not yet been evaluated. The highest yields per plant have been 960 g and 1460 g for primary and secondary dihaploids respectively.

### 4. CROSSES AT DIFFERENT LEVELS OF PLOIDY

The following points of interest emerge from the chromosome number data summarized in Table 1.

a. The progeny of cross 33 [(a hybrid of 60 chromosomes from *S. tuberosum* × *S. demissum*) × *S. phureja*] were expected to be dihaploids with 30 or hybrids with 42 chromosomes, but all proved to have 54. The incorporation of two genomes of *S. phureja* accounts for this.

b. Most progeny from cross 00, used to induce primary dihaploids, have 48 chromosomes instead of the desired 24 or the expected 36. The coloured tetraploids presumably contain two genomes of *S. phureja*.

c. Crosses 69, 121 and 133 are basically similar in that dihaploids have been crossed with tetraploids, cross 121 being, in fact, a dihaploid of var. *Maritta* back-crossed with *Maritta*. The single triploid progeny from this cross and that from cross 133 should be

Fig.1. Plants derived from self-pollinated secondary dihaploid (left), from spontaneous seed of secondary dihaploid (middle), and from dihaploid  $\times$  secondary dihaploid (right).



Abb.1. Pflanzen hervorgegangen aus Selbstung eines sekundären Dihaploiden (links), aus spontanem Samen eines sekundären Dihaploiden (mitte), und aus einem Dihaploiden  $\times$  einem sekundären Dihaploiden (rechts).

Fig.1. Plantes provenant d'un dihaploïde secondaire autopollinisé (à gauche), de semence naturelle d'un dihaploïde secondaire (au milieu), et du croisement dihaploïde  $\times$  dihaploïde secondaire (à droite).

considered triploid *tuberosum* plants. The remaining progeny of these three crosses are tetraploids. It is considered that in these examples the dihaploid parent produced ovules with 24 chromosomes. Two plants are self-fertile and the remainder exhibit good male and female fertility when crossed with tetraploid varieties.

d. Cross 116 was made at the diploid level [(primary dihaploid  $\times$  *S.goniocalyx*) secondary dihaploid] and all progeny had the expected 24 chromosomes.

e. The few plants obtained from cross 123 and 130 did not, as expected, have 48 and 60 chromosomes respectively, though such progeny might have been obtained if more plants had been produced. Four of the 5 progeny from cross 132 did have the expected 48 chromosomes and these plants were found to be fertile when crossed with other varieties.

f. One of the two progeny obtained from cross 125 [(a triploid from *S.tuberosum*  $\times$  *S.phureja*)  $\times$  secondary dihaploid] had the expected 30 chromosomes.

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Table 1. Progeny of crosses between potato plants of different ploidy

| Cross<br><i>Croisement</i><br><i>Kreuzung</i> | 2n             | Number of plants<br>investigated<br><i>Zahl der untersuchten</i><br><i>Pflanzen</i><br><i>Nombre de plantes</i><br><i>examinées</i> | No. of plants with<br><i>Zahl der Pflanzen mit</i><br><i>Nombre de plantes avec</i><br>chromosomes |    |    |    |    |    |    |
|-----------------------------------------------|----------------|-------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------|----|----|----|----|----|----|
|                                               |                |                                                                                                                                     | 24                                                                                                 | 30 | 36 | 48 | 54 | 58 | 60 |
| a. 33 (tbr · dms) · phu                       | 60 · 24        | 17                                                                                                                                  |                                                                                                    |    |    |    |    |    | 17 |
| b. 00 tbr · phu                               | 48 · 24        | 100                                                                                                                                 | 2                                                                                                  |    | 3  | 95 |    |    |    |
| c. 69 htbr · tbr                              | 24 · 48        | 20                                                                                                                                  |                                                                                                    |    |    | 20 |    |    |    |
| 121 htbr · <i>Maritta</i>                     | 24 · 48        | 6                                                                                                                                   |                                                                                                    |    | 1  | 5  |    |    |    |
| 133 (sh · htbr) · tbr                         | (24 · 24) · 48 | 6                                                                                                                                   |                                                                                                    |    | 1  | 5  |    |    |    |
| d. 116 (htbr · gon) · sh                      | (24 · 24) · 24 | 10                                                                                                                                  | 10                                                                                                 |    |    |    |    |    |    |
| e. 123 htbr · dms                             | 24 · 72        | 4                                                                                                                                   |                                                                                                    |    | 1  |    | 1  | 1  | 1  |
| 130 tbr · dms                                 | 48 · 72        | 4                                                                                                                                   |                                                                                                    |    | 1  | 1  | 2  |    |    |
| 132 dms · sh                                  | 72 · 24        | 5                                                                                                                                   |                                                                                                    |    |    | 4  |    | 1  |    |
| f. 125 triploid sh                            | 36 · 24        | 2                                                                                                                                   | 1                                                                                                  | 1  |    |    |    |    |    |

tbr = *S. tuberosum* phu = *S. phureja* sh = secondary dihaploid  
dms = *S. demissum* htbr = dihaploid gon = *S. goniocalix*

Tabelle 1. Nachkommenschaft von Kreuzungen zwischen Kartoffelpflanzen mit ungleicher chromosomenzahl

Tableau 1. Descendances de croisements entre plantes de Pomme de terre de différentes ploidies

## 5. BUILDING UP MULTIPLEX PARENTS

In breeding autotetraploid plants it is advantageous to have parental stocks with important major genes in the duplex or quadruplex condition. Twenty-five "doubled dihaploids" were obtained by treating germinated seed, callus tissue of cuttings, or meristematic eyes with colchicine (ROTHACKER and FIEDLER, 1957). These plants could not be used for further breeding because none flowered.

The tetraploid plants obtained from crosses 69, 121 and 133 resulted from fertilisation of unreduced female gametes of the dihaploid parent (JAHR, SKIEBE and STEIN, 1963). In contrast to the mitotically derived plants produced by colchicine treatment, these meiotically produced polyploids have excellent fertility.

## ZUSAMMENFASSUNG

### UNTERSUCHUNG DIHAPLOIDER KARTOFFELN IN DEN NIEDERLANDEN

Die Untersuchungen mit dihaploiden Kartoffeln wurden im Jahre 1961 begonnen. Wir verfügen derzeit über eine Kollektion von etwa 200 dihaploiden. Darunter befinden sich Klone mit Resistenz gegen *Phytophthora*, Kartoffelkrebs und

verschiedene Viren. Männliche Fertilität ist nur bei einem Klon gefunden worden. Indem wir amerikanische männliche fertile Haploiden verwendeten (U.S.-W.), konnten wir eine Anzahl Kreuzungen zustande bringen. Viele dieser

Nachkommenschaften (sekundäre Dihaploiden) besitzen guten Pollen. Einzelne sekundäre Dihaploiden sind selbstfertil.

Die Nachkommenschaft zeigt eine starke Inzuchtdepression (Abb. 1). Dihaploide Klone, homozygot für bestimmte Eigenschaften, sind bereits aufgebaut worden.

Tabelle 1 zeigt die Chromosomenanalyse einiger Nachkommenschaften in Valenzkreuzungen. Die so hervorgegangenen Tetraploiden sind männlich und weiblich fertil.

Mit Hilfe von Colchizin kann die Chromosomenanzahl dihaploider Pflanzen verdoppelt werden. Diese Tetraploiden bilden aber keine Blüten.

## RÉSUMÉ

### RECHERCHE SUR LES POMMES DE TERRE DIHAPLOIDES EN HOLLANDE

Les recherches sur pommes de terre dihaploïdes ont débutées en 1961. Depuis quatre années, nous disposons d'une collection de 200 dihaploïdes approximativement. Il existe parmi ces génotypes des clones résistants au *Phytophthora infestans*, à la galle verruqueuse et à différentes maladies à virus. La fertilité mâle n'a été constatée que chez un seul clone. On a pu réaliser un certain nombre de croisements en utilisant des haploïdes "mâles fertiles" américains (US-W). Beaucoup de descendance secondaires (dihaploïdes secondaires) possèdent du bon pollen. Rarement les dihaploïdes secon-

dares sont autofertiles.

La descendance montre un effet marqué de consanguinité (Fig. 1). On a déjà réalisé des clones dihaploïdes homozygotes pour certains caractères. Tableau 1 mentionne les résultats de l'analyse des nombres chromosomiques de quelques descendance de croisements de génotypes de ploidies différentes.

Les tétraploïdes obtenus sont également "mâles et femelles fertiles". On peut doubler le nombre de chromosomes des plantes dihaploïdes par action de la colchicine chimique. Cependant ces tétraploïdes ne produisent pas de fleurs.

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