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The response of the mutation frequency of barley in M_2 to the M_1 harvesting method

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Abstract. This paper deals with the frequencies of albina and xantha seedlings in an M_2 generation of six-rowed Pokko barley treated with sodium azide, NaN_3. The dependency of these frequencies on the harvesting method of M_1 plants is considered.

The results demonstrate that "one-grain-one-ear" harvesting in M_1 produced higher frequencies of both albinas and xanthas in M_2 compared with an M_2 originating from whole ear harvesting of M_1. For albinas the difference is significant. In spite of the increased mutation rate, the laboriousness of the "one-grain-one-ear" harvesting method restricts its feasibility in practical mutation breeding.

1. Introduction

The efficiency of a mutagen treatment can be measured in terms of the frequency of mutants in the generations subjected to selection. In addition to the quality and dosage of a mutagen, many other factors influence the mutation yield obtained. For instance, the mutation frequency is higher in the main ear of the barley plant treated at the seed stage than in the ears of the subsidiary tillers (GAUL 1961). Likewise, it has been surmised that the mutation frequency in the M_2 generation might be high in cases where the M_2 bulk originates from the maximal number of initial cells subjected to mutagen treatment. Such a situation could be attained, for instance, by taking only a single grain from each main ear of M_1 plants, which would ensure that every M_2 plant originates from a different initial cell.

Idea outlined above was suggested to the author in 1977 by Dr. R.D. BROCK, then working at the Joint FAO/IAEA Division in Vienna. On the basis of this idea, a practical mutation breeding programme was initiated on a six-rowed spring barley.

2. Treatment and handling of the M_1 generation

Using sodium azide, NaN_3, as mutagen, a breeding programme was started in May 1978. The subject was a six-rowed barley line, Hja 70352, which was subsequently released under the name Hankkija's Pokko (KIVI and REKUNEN 1980).

Pokko barley carries large spikes. In the control material grown in 1979 in the same field with the M_2 bulk, the average number of grains per spike was 50.3. The chemical mutagen used, NaN_3, has shown very high efficiency, producing a frequency of upto 7.7 % of morphological and physiological mutations in M_2 (KLEINHOFS et al. 1974). Its effectiveness can be increased by an 8—16 h prewetting treatments. In particular, NaN_3 treatment increases the number of point mutations, though not that of chromosomal aberrations (NILAN et al. 1976).

Following the method of NILAN and SANDER (1974), dry grains of Pokko barley were treated with phosphate buffered 0.3 M NaN_3 solution, pH 3. The lot comprised a 2 kg sample of screened barley, with 1000 gwt of 35 g and a germinability of 97 %.

The treated seed was sown in a trial field with irrigation possibilities. An ordinary plot seeder and normal seeding rate were used. The M_1 generation consisted of approximately 20.000 plants. The control material of similar extent was sown in the same field.

M_1 plants were harvested by hand, taking the main ear of each plant. From each ear separately, one grain was taken at random. The harvesting of 20.000 plants in this way took 28 working days.

3. The M_2 generation

In May 1979, an ordinary plot seeder was used to drill the M_2 in 8 m^2 plots. From both the control and the whole ear M_1 harvest samples were taken at random to compose two lots, each of equal weight to the "one-ear-one-grain" M_1 lot. Thus, three lots of approximately the same size were grown in 1979 (Table 1).

The rest of the M_1 yield, corresponding to the total whole-ear lot, was sown in the same field. This excess totalled about 350.000 plants, and was subjected to single plant selection for practical breeding purposes.

Table 1. The frequencies of albina and xantha seedlings in the M_2 bulks of NaN_3 treated Pokko barley originating from M_1:s harvested in various ways.

M_1	M_2				
Harvesting method	No. of seedlings	Albinas no.	Albinas %	Xanthas no.	Xanthas %
Whole ear (WE)	20500	82	0.40	22	0.11
One grain (OG)	20500	127	0.62	31	0.15
Control (CO)	19800	0	<0.01	0	<0.01
t test					
WE/OG		3.13**		1.24	
CO/WE		9.07***		4.69***	
CO/OG		11.31***		5.57***	

4. The albina and xantha mutants in M_2

In order to test the mutation frequency, two rather common chlorophyll aberrations, appearing as albina and xantha seedlings were counted (Table 1).

From among the total of 41.000 seedlings in OG and WE, 262 mutants of albina and xantha types were found. This corresponds to a mutation frequency of 0.64 per cent. The albinas predominated over the xanthas; their ratio being 209:53 or 3.94. This ratio is rather close to the value of 4.3 calculable as the average for different mutagens in the paper by HOLM (1954). In the control comprising approx. 20.000 seedlings, no albinas or xanthas were found. When the frequencies of both albinas and xanthas were compared between the M_2 bulks, it was found that OG harvesting on M_1 gave higher values than did WE. In the case of albina frequencies the difference was significant. The combined frequency of the two types of chlorophyll mutations observed in the OG lot was 0.76 %.

5. The feasibility of "one-grain-one-ear" harvesting

The results of this investigation demonstrate that the highest mutation rate results when the M_2 generation originates from the maximal number of initial cells of the generation subjected to treatment. Such a situation was created in this work by harvesting only one grain from each of the main spikes of the M_1 plants. The difference over an M_2 from whole ears of M_1 was positive both albinas and xanthas, and significant in the case of albinas. The excess of the albinas was 55 % and that of xanthas 43 % compared with the M_2 bulk originating from the whole-ear harvested M_1. A practical consideration restricting the feasibility of this method for increasing mutation yields is the very laborious harvesting involved compared, for instance, with collecting and threshing whole main spikes of the M_1 plants. In this work, an M_2 bulk of 350.000 plants from 20.000 whole M_1 ears and a "one-ear-one-grain" M_2 of only 20.000 individuals, i.e. the same size as M_1, were produced with the same input of labour.

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SELOSTUS

M_1-polven korjuutavan vaikutus mutaatiorunsauteen ohran M_2-populaatiossa

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Monitahoista Hankkijan Pokko-ohraa käsiteltiin kemiallisella mutaatiovaikuttajalla, natriumatsidilla (NaN_3). M_1-polven sadon talteenottotavan vaikutuksen tutkimiseksi korjattiin (1.) M_1-yksiköiden päätähkät sekä (2.) erikseen yksi sattumanvaraisesti otettu jyvä kustakin päätähkästä. Viime mainitulla korjuutavalla päästiin siihen, että jokainen M_2-polven yksilö varmasti polveutui mutagenssikäsittelyn kohteeksi otetun jyväerän eri initiaalisolusta.

Mutaatiovaikutuksen mittaamisessa käytettiin oraiden kahden lehtivihreäpoikkeaman runsautta. Kun korjuu oli suoritettu tavalla (2.), oli M_2-polvessa valkoisten albina-oraiden määrä 55 prosenttia ja keltaisten xantha-oraiden 43 prosenttia runsaampi kuin tavalla (1.) korjatusta aineistosta polveutuvassa M_2-aineistossa. Albina-mutanttien osalta ero oli tilastollisesti merkitsevä.

Suuremmasta mutaatiorunsaudesta huolimatta tekee korjuun hankaluus "yksi jyvä tähkästä" -menetelmän epäedulliseksi käytännön mutaatiojalostuksessa. Samalla työmäärällä, joka menetelmällä (2.) korjaten tuotti 20.000 yksilön M_2-populaation, saatiin kokonaisina päätähkinä korjatusta M_1-polvesta (1.) 350.000 yksilön M_2-aineisto valinnan kohteeksi.

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