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Short Communication

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New Winter Wheat Variety: 'GK Délibáb' Developed via Combining of Conventional Breeding and Method of *In Vitro* Androgenesis

In memory of Zoltán Barabás academician

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With 2 figures and 1 table

Abstract

Wheat (*Triticum aestivum* L.) cultivar 'GK Délibáb' was improved and released in 8 years combining traditional breeding steps and plant biotechnology methods. 'GK Délibáb' is derived from a multiple cross made in 1984. After F_2 selection, haploids were induced from F_3 generation in anther culture. Fertile doubled haploid (DH) seeds and lines obtained via colchicine treatment were propagated and tested further in nursery. After three years of official tests of Institute for Agricultural Qualification (IAQ) for registration 'GK Délibáb' was registered and released as a wheat variety in Hungary. The major agricultural advantage of 'GK Délibáb' are excellent winterhardiness, premium bread making quality and earliness. The performance of 'GK Délibáb' suggests that lines originating from DH plants may be as valuable as lines originating from other sources.

Key words: *Triticum aestivum* L. - anther culture - androgenesis - doubled haploid - breeding - release - cultivar

Breeders combine conventional and newly developed methods to produce improved crop varieties in the shortest possible time. Haploid methods (*in vitro* androgenesis,

bulbosum technique etc.) have been used extensively in breeding to achieve genetic homozygosity. Reaching the homozygote state is of a primary importance in production of new breeding lines and varieties (Baenziger et al. 1989, Picard et al. 1990).

Production of microspore-derived plants from anther or microspore culture has been reported in more than 250 species of plant kingdom, but publications in breeding applications are limited. The phenomenon of *in vitro* androgenesis in wheat has been reported by three independent research teams (Ouyang et al. 1973, Picard and De Buyser 1973, Wang et al. 1973) about 20 years ago. On the other hand, we had to wait for a relatively long time for the first androgenic-derived wheat varieties. The first dihaploid wheat variety in the world (Hu et al. 1986) and in Europe (De Buyser et al. 1987) was released in the last ten years.

In CRI's winter wheat breeding programmes, to avoid long selection, in which each cycle requires generally one growing season, we have initiated a wide laboratory/nursery breeding programme in 1984. The purpose of this report is to describe the first result of this breeding programme. After three-year test in the Institute for Agricultural Qualification, 'GK Délibáb' a DH winter wheat variety was registered by the National Council for Agricultural Variety Qualification in December of 1992, in Hungary (IAQ 1993). This paper describes the combination of breeding, plant biotechnology technique and the process leading to the release of 'GK Délibáb'.

In 1984 the Hungarian (Mini Manó, Mv 12), Russian (Jubilejnaja 50) and Bulgarian (Sadovo Super) winter wheat varieties were used for a multiple cross: Mini Manó // Jubilejnaja 50 / Sadovo Super /3/ Mini Manó / Mv 12. The parents possess the genes which are important in Hungarian cultivation, as winterhardiness, premium bred quality, high yielding, resistance to fungus diseases (powdery mildew, stem and leaf rust) etc. Vernalized F_1 plants were grown in greenhouse and the first selection was made in F_2 under nursery condition. The seeds (F_3) of selected heads were grown in greenhouse and anthers cultured therefrom. Before tissue culture work, donor tillers were cold treated as described later (Pauk et al. 1991). For callus and embryoid induction 'P-4' medium was used (Ouyang et al. 1983). Anthers were incubated at 28 ^{0}C in dark thermostat. Every three days calli of

suitable size were transferred on '190-2' regeneration media (Zhuang and Jia 1983). Regeneration cultures were kept at 28 ^{0}C in a 16 h photoperiod provided by cool white fluorescent tubes. Well tillered and rooted plants grown *in vitro* were transferred in pots in standard soil into greenhouse. The cytology of individuals were tested from root tips. To restore fertility for seed collection, the chromosomes of haploid plants have to be doubled. The haploids were treated by 0.02% colchicine, 2% DMSO solution for 5 hours followed by an overnight washing into tap water. The spontaneous and colchicine induced DHs were vernalized at 3-5 ^{0}C in cool chamber for 42 days and grown in greenhouse under standard growing condition. The collected DH seeds were propagated and the selection for positive genotypes was carried out in nursery.

In CRI's Wheat Cell & Tissue Culture Laboratory, the haploid wheat production has been carried on for breeding purposes since 1984. Haploids - one-two thousand per year - are induced from anther cultures of various wheat populations in order to achieve homozygosity rapidly.

In the case of 'GK Délibáb' the conventional breeding steps were initiated in 1984 (Fig 1.). After F_1 seed propagation and selection of F_2, one thousand anthers - from F_3 greenhouse grown individuals - were plated during the autumn of 1985. Via tissue culture plant regeneration, root tip citology, colchicine treatment and greenhouse seed propagation 49 DH lines were obtained in October of 1986, when the nursery test and propagation was started. In field test, the homogeneity of lines was tested in ear to row system. To screen the outcross and physical mixing the seed of rows constitute the sublines of individual DHs. The maintenance of DH lines is carried out as in conventional breeding in Szeged (Barabás et al. 1987). In the autumn of 1989 - after one individual selection and two yield tests - 'DH 773' line was presented to the Institute for Agricultural Qualification to carry out official tests for registration. During these three years of official tests for registration 'GK Délibáb' has been tested in 5, 14 and 14 locations, respectively; in different locations of Hungary. The mean yields were 107,9%, 106,3% and 93,5% as compared to the control group (Table 1) and it was accepted for registration in December 1992. In the 2nd and 3rd year of the official test, 'GK Délibáb'

successfully passed the test for varietal characteristics, distinctness, uniformity, stability, and it was accepted for patent (Fig. 1.).

'GK Délibáb' (Fig. 2a) is a released and patented winter wheat (*Triticum aestivum* L.) variety. 'GK Délibáb' is awnless, with 10-25 mm short awns on the top region of the head. At present, it is the earliest winter wheat cultivar in Hungary with high yield, excellent winter hardiness and premium baking quality (Fig. 2b). It is moderately resistant to powdery mildew, red rust and moderately susceptible to stem rust, but it can avoid the consequence of epidemics by its earliness under Hungarian climate. Breeder seed of 'GK Délibáb' is maintained (Fig. 2c) by the Cereal Research Institute.

Zusammenfassung

Eine neue Winterweizensorte 'GK Délibáb' gezüchtet mit einer kombinierten Pedigree und *in vitro* Haploidmethode

Die neue Sorte (GK Délibáb) wurde in 8-jähriger Arbeit hergestellt, wobei herkömmliche und biotechnologische Methoden angewendet worden sind. Sie stammt aus einer mehrfachen Kreuzung aus 1984. Von der F_2 Generation wurden die besten Pflanzen selektiert und in der darauffolgenden F_3 dieser Pflanzen wurden Antherenkulturen angelegt. Die daraus resultierenden Haploiden wurden durch Kolchicinbehandlung rediploidisiert (DH Pflanzen) und im Zuchtgarten weitergeführt. Nach dreijährigen Untersuchungen beim Staatlichen Sortenamt und beim Institut für Landwirtschaftliche Qualifizierung wurde die Sorte anerkannt und nach UPOV Regeln registriert. Die Sorte hat ausgezeichnete Winterfestigkeit, Backqualität und ist frühreif.

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Table 1. Yield performance of the doubled haploid (DH) winter wheat variety 'GK Délibáb'*

Year	No. of location	Control varieties (x=100)	'GK Délibáb' value (% of controls)
1990	5	GK Öthalom, Mv 19, Korona	107.9
1991	14	GK Öthalom, Mv 19, Korona	106.3
1992	14	GK Öthalom, Mv19, Korona	93.6
3-year average			102.6

* after results of the Institute for Agricultural Qualification (IAQ 1990, 1991, 1992)

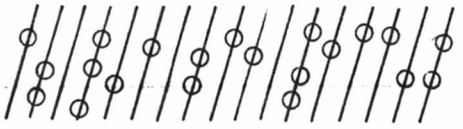
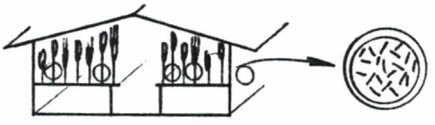
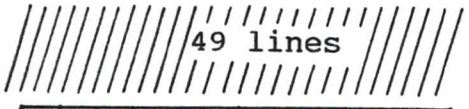
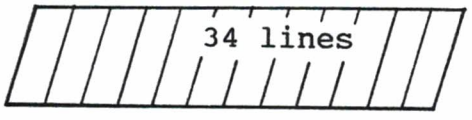
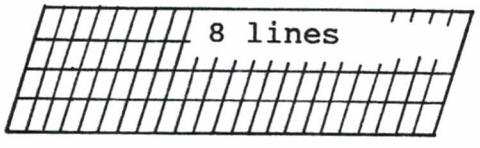
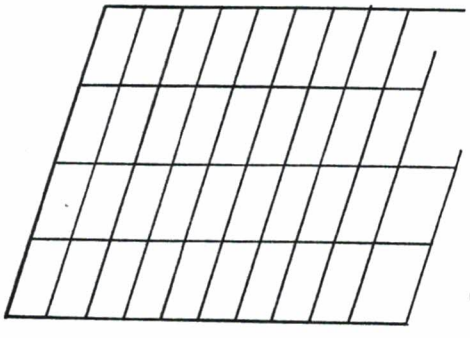
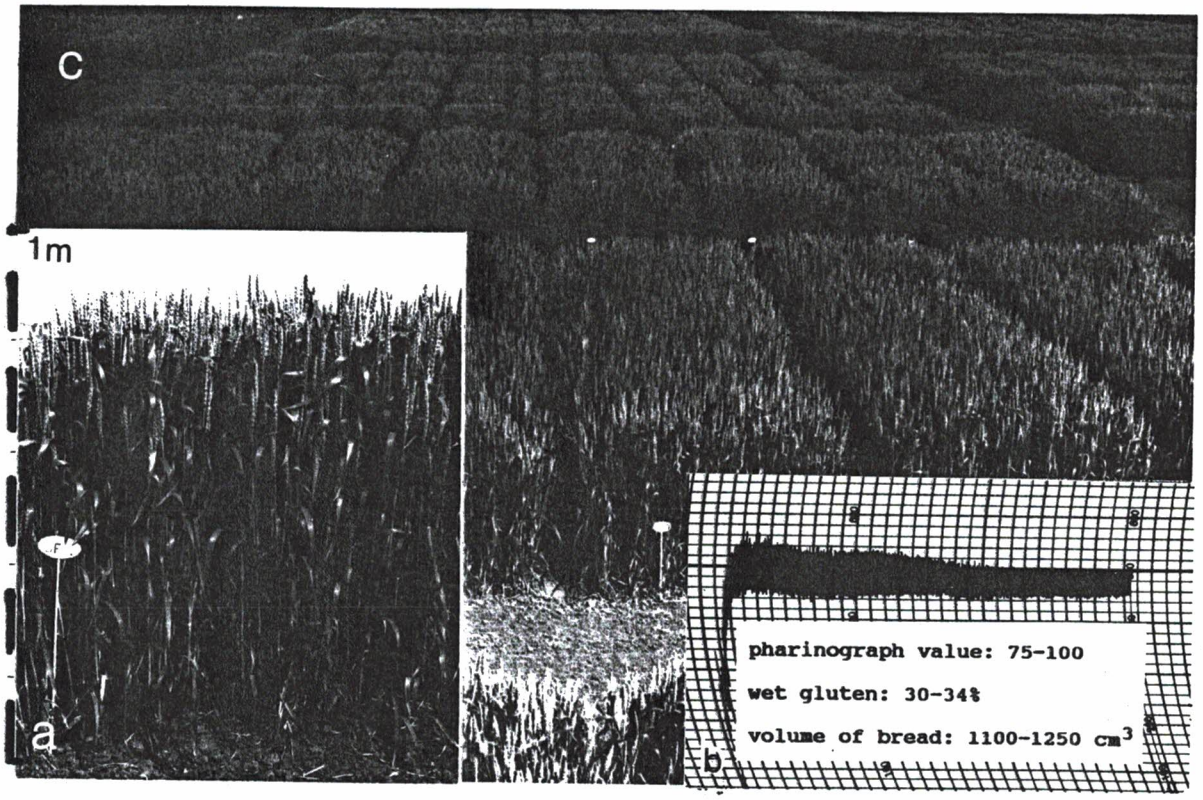
Time (year)	Breeding steps schematic	comment
1984	A x B,C..	crossing
1984		growing F1 in greenhouse
1985		individual selection of F_2 plants in nursery
1985/86		anther culture of F_3 plants grown in ghouse
1986/87		ear to row test of DHs
1987/88		yield test in no repl., at 5 m^2
1988/89		yield test at 2 locations, 4 replications
Sept. 1989	'DH 773'	the superior line
1989/90		official test for registration by the Institute for Agricultural Qualification
1990/91		
1991/92		
Dec. 1992	'GK Délibáb'	registration and patent
1993-		seed market

Fig. 1. Development scheme of 'GK Délibáb' combining of conventional pedigree- and *in vitro* haploid technique, androgenesis


c
1m
a
b
pharinograph value: 75-100
wet gluten: 30-34%
volume of bread: 1100-1250 cm3

Subtitle of Fig. 1. and Fig. 2.

Fig. 1. Development scheme of 'GK Délibáb' combining of conventional pedigree- and *in vitro* haploid technique, androgenesis

Fig. 2. 'GK Délibáb' doubled haploid winter wheat variety /a/, its pharinogram with value, data of wet gluten, volume of bread /b/, and variety maintenance /c/