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RELEASE OF GENE SILENCING IN TRANSGENICS - A NEW ROLE FOR INDUCED MUTATIONS

Gene silencing (GS) is defined as loss of expression of previously expressed gene(s) [1]. In plants GS was found after several reports of non or unstable expression of transgenes in the recipient plants, in spite of the physical presence of the introduced gene [2; 3]. Besides transgenes, silencing of homologous host genes, referred as co-suppression, has been reported [4]. In non-transgenic plants, GS is implicated in epigenetic changes, and host plant responses to viral infections [5;6]. Expression of the gene could be blocked at the transcription level or by the degradation of RNA after trancription. The latter is known as post-transcriptional gene silencing (PTGS). Promoter methylation and chromatin structural changes are correlated with transcriptional gene silencing. No changes in transcription rate but marked reduction in cytoplasmic RNA is observed in PTGS. DNA methylation has also been observed in some instances of PTGS but not in others. Transgene silencing is a source of instability, and a matter of concern, in the development of transgenic crops [7]. Earlier, mutants that enhance gene silencing [8] as well as for decreased methylation [9] were reported in *Arabidopsis thaliana*. Recently, two reports [1; 4] on release of transgene silencing by induced mutations in *Arabidopsis* were published. Important features of *Arabidopsis* mutants that release or enhance GS, increase or decrease DNA methylation are summarised in Table 1.

The presented works clearly demonstrated that using the standard methods of mutagenizing seeds (EMS and N_f treatments), mutants can be isolated to release transgene silencing, increase or decrease methylation of transgenes as well as of the resident genes in *Arabidopsis*. Heritable, mutant phenotypes resulted from alterations in GS and hyper- or hypo-methylation of the genome. It is likely that loci homologous to *ddm*, *egs*, *sgs* and *som* found in *Arabidopsis* exist in other plants. The importance of gene silencing in epigenetic alterations, plant host - viral interactions, defence responses, and of methylation and de-methylation in controlling gene expression during differentiation, flowering, flower development, and vernalization response [13] is challenging new area to identify mutants altering methylation pattern in crop plants. At the same time, there may be many observations, made in earlier mutation, and *in-vitro* experiments that can now be analysed for GS and alterations in DNA methylation.

Table 1. Induced mutants suppressing or enhancing gene silencing and increasing or decreasing DNA methylation in *Arabidopsis*

Mutant(s)- Symbol(s) and References	
Genetic stock/Mutagenesis	Main characteristics of the mutant(s)
***sgs1, sgs2* (suppresser of gene silencing) [4]**	
Transgenic, 35S-***uidA*** silenced line EMS (ethyl methanesulphonate) 0.4% for 16 h at room temperature, 500 seeds treated, seeds of 5 M_1 plants harvested in bulk, seven mutants were isolated, six showed total release - one allele of *sgs1,*	Release of PTGS; restoration of β-glucuronidase (GUS) activity; GUS activity and *uidA* RNA increased 3500 fold; *uidA* transcription rate increased only 3 fold; monogenic recessive; transgene methylation is reduced; methylation of centromeric repeats not

five alleles of *sgs2*. In subsequent screening in progeny of 2,000 mutagenised seeds, *sgs2* mutants isolated at high frequency - indicating that *sgs2* is highly mutable	affected; *sgs* mutants differ from *ddm* and *som* mutants; did not release transcriptional silencing of 35S-*hpt* (hygromycin phosphotransferase) transgene; released co-suppression of host *Nia* (nitrate reductases) and 35S-*Nia2* transgenes; acts in *trans* to release PTGS
<u>*som* (somniferous)</u> [1]	
Transgenic, silenced line with *hpt* gene EMS 0.3% for 12 h, 50,000 seeds; fast neutrons (N_f) 60 Gy, 75,000 seeds; M_2 seeds collected separately from each tray; 1 mutant per 16,000 M_2 plants - EMS; 1 mutant per 5,700 M_2 plants - N_f; eight putative mutants isolated	Heritable, restoration of hygromycin resistance; transgene reactivation; DNA methylation of the reactivated *hpt* locus and at centromeric repeats reduced; reactivate transgenic, test locus in *trans;* some *som* mutants (*som 1*, *4* and *5*) are allelic to *ddm1* which cause DNA hypomethylation (group A); others are nonallelic with *ddm1* and *som* mutants of group A; third group of mutants show slow resilencing after crossing to *ddm1* and the wild type
<u>*egs1* and *egs2* (enhancer of gene silencing)</u> [8]	
Transgenic line with ***rolB*** gene of *Agrobacterium rhizogenes* impaired in shoot regeneration. Shows inhibition of hypocotyl and internode elongation, pronounced growth retardation, altered flower morphology, and early senescence, very low frequency of shoot regeneration is observed. EMS 0.2% for 16 h at room temperature, M_2 seeds bulk harvested, 70,000 M_2 seedlings from 11,000 M_1 plants, five mutants of independent origin, isolated *egs1* with three alleles *1*, *2* and *3* *egs2* with two alleles *1* and *2*	Frequency of *rolB* gene silencing increased and timing altered; 99% *rolB* silencing by *egs1-1;* silencing at earlier developmental stage; silencing of *rolB* results in normal growth and differentiation in plants; silencing correlated with marked reduction in *rolB* transcripts in cytoplasm; nuclear transcription only moderately reduced; evidence supports PTGS; silenced state mitotically stable; silencing is reversed in the sexual progeny; recessive inheritance
<u>*ddm1* (decrease in DNA methylation - hypomethylation)</u> [9; 10; 11]	
Columbia ecotype EMS; 79 pools representing 2,000 plants followed by single plants, screening of pools for hypomethylated DNA using Southern blot analysis, three independent mutants isolated: *ddm1-1* and *ddm1-2*	Non tissue specific hypomethylation of genomic DNA from leaves, stems, flower buds and roots; hypomethylation of centromeric repeats, 5.8S, 18S, and 25S rDNA; 70% reduction in genomic 5 methylcytosine levels; DNA methyltransferase activity similar to wild type; *ddm1/ddm1* homozygotes show increase in the number of cauline leaves, altered leaf shape, delayed flowering and other developmental abnormalities; mitotic stability and meiotic heritability of

	induced developmental abnormalities; re-methylation of DNA sequences hypo-methylated by *ddm1* mutation is slow or nonexistant even in wild type (*DDM1*)

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THE ROLE OF INDUCED MUTATIONS IN THE IMPROVEMENT OF COMMON BEANS (*Phaseolus vulgaris* L.)

An early bush type mutant of *Phaseolus vulgaris* L. was selected, already in 1940s, after x-ray treatment of the variety 'Michelite', a viny type small-seeded common bean [2, 3]. The small, early bush type mutant developed at Michigan Agricultural Experiment Station was backcrossed to the parent Michelite and a large, early bush type was recovered. This type, and other bush type mutants of Michelite origin, were used in crosses with anthracnose resistant beans. After four generations of back-crosses and five generations of selections a large number of anthracnose resistant bush types were available and initially tested for their agronomic traits and canning quality in 1953. After a positive evaluation in multi-location trials, the variety 'Sanilac' was released in 1956. In 1958, seeds of this variety were available to all bean growers. From 1960 to 1988 a large number of new and better bean varieties were developed using the x-ray induced bush type mutants or their derivatives in the pedigree (Table 1). 'Seafarer' (released in 1967), a very early variety with improved disease and virus resistance, was frequently used to develop varieties released in the 80s. In 1955, the parent Michelite was sown on 95% of the 400,000 acres of small-seeded white beans (navy or pea beans) in Michigan [2]. In the next two decades after the release of the first mutant derived variety, most of the navy bean cultivation area in Michigan was covered by Sanilac and other bush varieties developed from the x-ray mutants of Michelite [1].

For the development of the currently grown navy bean varieties in North America, the white-seeded bush bean mutant 'NEP-2' was extensively used as cross parent in bean improvement programmes. NEP-2 was induced from the black-seeded variety 'San Fernando' through mutagenesis [4]. It was one of the various seed-coat colour mutants resulting from a mutation programme using EMS or gamma ray treatment at the Inter-American Institute of Agricultural Sciences in Turrialba, Costa Rica [14, 15]. It was found resistant to common mosaic virus, with good canning quality and good agronomic traits [16]. NEP-2, was released in the mid-seventies [4]. Its use as cross parent in breeding programmes lead to the release of many, more productive bean varieties, with mainly upright short vine plant architecture in the 1980s and 90s (Table 2). Varieties such as 'C-20', 'Laker', 'Norstar, 'Huron' and 'AC Skipper' have derivatives of both mutants in their pedigree. Altogether, more than 40 mutant-derived varieties were released by incorporating one or two mutant types in bean breeding programmes in North America from 1956 until 1998. These varieties mainly belong to the group of small white-seeded beans, some to the related black and pinto bean groups. At present, the small white bean varieties 'AC Hensall', 'Centralia', 'Stinger' and 'Crestwood' are cultivated in Canada [9], whereas the varieties 'Albion', C-20, Crestwood, 'Mayflower', 'Norstar', Seafarer, 'Black Magic' and 'Black Hawk' are grown in the small bean production areas of the USA [5, 6]. In Michigan, 40% of the 300,000 acres of navy bean are covered by varieties developed from C-20 (J.D. Kelly, personal communication).

'IAPAR 57' released in 1992, was the first variety with high tolerance to golden mosaic virus disease released in Brazil. It yields under high virus disease pressure two and half times more than susceptible varieties. It originated from the

cross of lines 'MD 632' x 'BAC 32'. The virus disease tolerance of MD 632 derived from its mutant parent 'TMD-1' which was used in various cross-breeding programmes [18,19]. Tolerance of TMD-1 to golden mosaic virus disease was induced by EMS seed treatment of 'Carioca' [17].

Table 1. Bean varieties/lines deriving from x-ray induced bush mutants of Michelite or their derivatives (compiled from references 4, 7, 8, 10, 11, 12, 13)

Variety/line	Parent contributing the bush type	Year of release	Breeding institution/company
Navy beans (pea beans)			
Sanilac	x-ray induced mutant of Michelite	1956	Michigan State University (MSU)
Seaway	x-ray induced mutant of Michelite	1960	MSU
Gratiot	x-ray induced mutant of Michelite and Sanilac	1962	MSU
Seafarer	x-ray induced mutant of Michelite	1967/1971	MSU
Kentwood	Sanilac	1973	Agriculture Canada (AC)
Fleetwood	Sanilac	1977	AC, University of Guelph (ACUC)
C-20 (see table 2)	Kentwood	1982/1984	MSU
Harofleet	Fleetwood	1983	ACUC
Harokent	Kentwood	1983	ACUC
Midland	Seafarer	1983	Asgrow Seed Company
Northland	Seafarer	1983	Asgrow Seed Company
OAC Seaforth	Seafarer	1983	ACUC
Wesland	Seafarer and Kentwood	1983	Asgrow Seed Company
Laker (see table 2)	Kentwood	1983/1984	MSU
Mitchell	Seafarer	1986	AC, Harrow, Ontario
Neptune	Seafarer	1986	MSU
Suncrest	Seafarer	1986	Gentech
Centralia	Harokent	1988	ACUC
Stinger	Seafarer	1988	Asgrow Seed Company
Norstar (Tab. 2)	Fleetwood	1993	North Dakota State University
Huron (Tab. 2)	Harokent	1994	MSU
Newport	Harokent	1995	MSU
AC Skipper (Tab. 2)	Kentwood	1996	AC, Alberta
AC Hensall	OAC Seaforth	1997	AC, Ontario
MSU x 80101	Kentwood	-	MSU
Albion	Seafarer	-	-
Dresden	Fleetwood	-	-
Pinto beans			
Ouray	Sanilac	1971/1982	Colorado State University
Arapaho	Ouray	1995	Colorado State University
Other beans			
73130-E2-B	Kentwood	-	-
NC Alberta Pink	Swan Valley and Kentwood	1998	AC, Alberta

Table 2. Bean varieties/lines deriving from the induced mutant variety NEP-2, released 1975 or their derivatives (compiled from references 4, 7, 8)

Variety/line	Parent contributing the 'NEP-2' genotype	Year of release	Breeding institution/company
Navy beans (pea beans)			
Swan Valley	NEP-2	1981/1986	MSU
C-20 (see table 1)	NEP-2	1982/1984	MSU
L-226-10	MSU N80051 and MSU N81009	1983	MSU
L-227-1	MSU N80051 and MSU x 80101	1983	MSU, University of Puerto Rico
Laker (see table 1)	NEP-2	1983/1984	MSU
Mayflower	C-20	1988/1989	MSU USDA/ARS
Norstar (see table 1)	C-20	1993	North Dakota State University
Huron (see table 1)	C-20	1994	MSU
AC Skipper (see table 1)	Swan Valley	1996	AC, Alberta
MSU N76012	NEP-2	-	MSU
MSU N80051	MSU N76012	-	MSU
MSU x80101	NEP-2	-	MSU
MSU N81009	NEP-2	-	MSU
Black beans			
Black Magic	NEP-2	1981/1987	MSU USDA/ARS
Domino	NEP-2	1981/1987	MSU USDA/ARS
Black Hawk	Black Magic	1989/1990	MSU
Pinto beans			
JM-126	NEP-2	1986	USDA/ARS Washington State University (WSU)
Maverick	Black Magic	1997	North Dakota State University
Frontier	Black Magic	1998	North Dakota State University
Great Northern beans			
JM-24	NEP-2	1986	USDA/ARS WSU
Other beans			
MSU Sel. #61627	NEP-2	-	MSU
MSU N80043	MSU Sel. #61627	-	MSU

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DEVELOPMENT OF A NEW JUTE (*Corchorus. capsularis*) VARIETY 'BINADESHIPAT-2' THROUGH SODIUM AZIDE MUTAGENESIS

XA0054528

Jute is the most important agricultural resource of Bangladesh for earning foreign currency [1]. In Bangladesh it is generally sown in April. Mustard and some pulses are harvested in February and although the land is available from early March for sowing jute, here is no variety which can be grown at that time. If jute is sown in early March it initiates flowering at a premature stage. Ultimately it reduces the fibre yield and quality. To overcome this, seeds of the widely cultivated variety CVL-1 were treated with 4, 6, 8, 10, 12, 14, 16, 18 and 20 mM concentrations of sodium azide (NaN_3). Two thousand seeds were treated for each concentration and M_1 generation was grown. Seeds were collected from each M_1 plant and kept separately. Collected seeds were divided into three lots and sown in the fourth week of February and the first and second week of March. In M_2 only 200 individual plants were selected from the first March sowing, on the basis of late flowering (after 120 days) and plant growth. Strain C-278 (from treatment with 12 mM NaN_3)

bred true in the subsequent generations and showed better performances in respect to flowering and fibre yield compared to its parent variety CVL-1. The strain was evaluated in preliminary, advanced, regional and farmers' field trials for its flowering behaviour and fibre yield. Agronomic and fibre quality characters are shown in Table 1. The strain C-278 showed taller plant height and higher base diameter than the parent variety. It produced 8.14% higher fibre yield than the variety CVL-1 (average of 15 on-station and 10 farmers` field trials). The strain C-278 also has improved fibre quality. Finally, it was evaluated that the strain C-278 (i) has no problem of early flowering when sown in the first week of March (ii) could be harvested in July when the land should be available for transplanting Aman rice (iii) has possibility to escape early floods in the low lying area and (iv) has also improved fibre quality. Finally, in 1997, the National Seed Board released the strain C-278 as an early sowing and high fibre yielding variety under the name 'Binadeshipat-2'.

Table 1. Some agronomic and fibre quality characters of Binadeshipat-2 (C-278) along with its parent variety CVL-1

Strain/ Variety	Agronomic parameters			Fibre quality parameters			
	Plant height (cm)	Base diameter (cm)	Fibre yield (t/ha)	Elongation at break (%)	Energy to break point (mj)	Linear density (tex)	Bundle strength (lbs/mg)
Binadeshipat-2 (C-278)	317	1.94	2.79	1.29	0.275	2.25	11.18
CVL-1 (parent)	288	1.82	2.58	0.98	0.129	2.03	10.85

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XA0054529

HIGH QUALITY TURFGRASS THROUGH GAMMA IRRADIATION

In 1983, dormant stolons (each with two nodes) of the triploid (2n = 3x = 27) bermudagrass cultivar 'Midiron' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Butt-Davy] were treated with 80 Gy of ^{60}Co gamma rays. Sixty-six fine-textured mutants or sectors were selected as plants began to grow from the stolons. After 12 years of multi-location testing, mutant number 40 was released as 'Tift 94'. Tift 94 has the cold tolerance of Midiron but has improved turf quality under close mowing. It also shows non-preference resistance to mole crickets (*Scapteriscus* spp.) (Table 1).

Table 1. Quality characteristics of Tift 94 turf bermudagrass [1]

Variety	Quality[2]		Quality[3]		Mole cricket damage[4]
	14 Aug.	6 Oct.	16 July	19 Oct.	
Tift 94	8.0	8.0	8.0	8.5	1.0
Midiron	6.0	5.5	2.5	2.5	5.4
LSD (0.05)	1.6	1.6	1.9	2.4	2.3

[1] Ratings for quality: 1 = poor and 9 = best.
[2] Test established in 1983, ratings from 1991. Grass cut at 25 mm 3x per week.
[3] Test established in 1987, ratings from 1993. Grass cut at 12 mm 3x per week.
[4] Ratings for mole cricket: 1 = resistant and 9 = susceptible.

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XA0054530

NEW SEEDED CENTIPEDEGRASS TURF THROUGH GAMMA IRRADIATION

Seeds of common centipedegrass (*Eremochloa ophiuroides*) were recurrently irradiated with 12 Gy ^{60}Co gamma rays. In each cycle, 1500 plants were spaced on 0.3 m centers and allowed to interpollinate. Seed was bulk-harvested in each cycle. 4,500 plants from cycle 3 and 2,300 plants from cycle 5 were space planted on 0.3 m centers at Blairsville, GA. Plants that survived -28^{0}C during the 1984-1985 winter were interpollinated. Seeds from these surviving plants become 'TifBlair', the first seeded centipedegrass cultivar with a known pedigree. Compared to common centipedegrass, TifBlair is more cold tolerant, produces more and faster growing stolons, grows taller, produces larger seeds and grows more rapidly on a soil of pH 4.3.

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XA0054531

DEVELOPMENT OF MUNGBEAN VARIETY 'NIAB MUNG 98' INVOLVING INDUCED MUTANTS THROUGH CONVENTIONAL BREEDING

Genetic improvement of mungbean (*Vigna radiata* L. Wilczek) through induced mutations [1] has been in progress at the Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan for the past two decades. Breeding efforts have resulted in the development of high yielding and short duration mutants 'NIAB MUNG' (NM) viz. NM 28, NM 13-1, NM 19-19, NM 20-21, and NM 121-25, released as varieties for direct utilization. Irradiation of F_1 hybrids between CV 6601 and VC 1973A has resulted in the development of NM 51 and NM 54.

NIAB MUNG 98 was developed through hybridization [2] between an induced mutant variety NM 20-21 and an exotic AVRDC accession VC 1482E, by the pedigree method of selection. Yield evaluation of NIAB MUNG 98 along with parent varieties in different trials (screening nurseries, microplot, advanced, national, and adaptation) indicated that NIAB MUNG 98 produced 14% and 17% higher seed yield as compared to NM 51 and NM 20-21 respectively (Table 1). NIAB MUNG 98 has significantly higher number of pods and seed yield per plant (Table 2). It has also shown resistance against Cercospora Leaf Spot (CLS) and Mungbean Yellow Mosaic Virus (MYMV). Based upon superb yield performance of NIAB MUNG 98, the variety was approved in November 1998, by the Provincial Seed Council for general cultivation in the Punjab Province.

Table 1. Yield performance (kg/ha) of NIAB MUNG 98

Trial	Year	Variety		
		NM 20-21	NM 51	NM 98
Screening Nursery	1990	1664	-	1790
	1991	1079	-	1654
Microplot Trials	1992	1394	1624	2138
	1993	1387	1789	2902
Advanced Yield Trials	1994	-	1279	1529
	1995	-	1785	1806
National Trials	1995	-	-	675
	1996	-	-	1244
Adaptation Trials	1996	-	777	1049
	1997	-	1292	1378

Table 2. Seed yield and yield components in NIAB MUNG 98 in comparison with standard varieties

Variety	Pod/plant	Pod length (cm)	Seed/pod	1000 seed Wt. (g)	Yield/plant (g)
NM 98*	47.1	8.4	11.0	38.0	20.7
NM 51	36.3	9.7	12.1	42.1	16.0
NM 54	28.8	9.9	11.8	57.8	15.8
NM 20-21	30.0	7.8	10.7	36.3	13.0
NM 121-25	43.3	8.7	10.9	30.6	14.0
LSD 5%	4.51	NS	NS	0.42	1.94
1%	6.56	NS	NS	0.61	2.81

*/Plant height: 60cm, Resistant to Mungbean Yellow Mosaic Virus and Cercospora Leaf Spot diseases, matures 70-80 days.

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DEVELOPMENT OF HIGH YIELDING MUTANTS FROM AN ELITE RICE CULTIVAR 'SWARNA PRABHA'

XA0054532

Induced mutagenesis serves as an important tool for creating usable genetic variability in crop plants and significant achievements in crop improvement have been made through the mutation approach. It also serves as a supplement to conventional breeding programmes to improve one or two specific characters in a well-adapted and acceptable elite cultivar.

'Swarna Prabha' is a highly physiologically efficient rice cultivar and records stable yields in both wet and dry seasons. The photosynthetic rate is high (32-34mg CO_2 dm^{-2} hr^{-1}) and also, the reduction in both photosynthetic rate and stomatal conductance to CO_2 was very low under low light conditions, which are prevalent in Eastern India during the wet season. The cultivar, however, possesses a weak culm (prone to lodging) and coarse grain.

The mutation approach was followed to improve the cultivar through induced genetic changes for both grain type and stiffer culm. Dry seeds were subjected to gamma ray irradiation (250 Gy) and the M_2 population was screened for mutants. Thirty mutants with alterations for grain characters (grain shape and test weight) were identified but semi-dwarf culm mutants could be only observed in M_3 but not in M_2 generation. In M_2 generation, segregation was observed for panicle and grain characters. One hundred and sixty five selections were made in M_3, of which 30 were semi-dwarf. The M_4 mutant lines were evaluated for stability of grain characters and culm stiffness. It was observed that the alteration from coarse grain to fine has also resulted in weak culm in a few instances and all the short culm mutants showed very low productivity.

Twentyseven M_4 selections were evaluated for their photosynthetic rate and the mutants showed a wide range of values between 29.4 and 35.7 CO_2 dm^{-2} hr^{-1} as against the parental value of 33.4. Four mutant lines surpassed the parental value. However, in the present study, no positive relationship was observed between the photosynthetic rate and grain yield. Further yield evaluation trials resulted in identification of two promising mutants, 'CRM 40' and 'CRM 41' from the twenty-seven selections. The mutants were submitted to the All India Coordinated Rice Improvement Programme for multilocational Initial Evaluation Trial (IET). One of the mutants, CRM 40 - which recorded high yields in several regions, was promoted to the Advanced Variety Trial (AVT) in 1991. The mutant recorded higher yields over the controls in different locations like Orissa, Bihar and Madhya Pradesh in both 1991 and 1992. On the basis of its relative performance, CRM 40 was recommended for release in two states, Orissa and Madhya Pradesh in 1993. Extensive trials conducted in farmer's fields confirmed the results of the Coordinated trials as CRM 40 recorded high yields consistently in both irrigated medium lands and also in bunded uplands. Based on its performance, CRM 40 was released as 'Radhi' by the State of Orissa in 1996 and was recommended for general cultivation in irrigated lands in both wet and dry seasons.

The mutant cultivar, Radhi, is tall in stature (~120 cm) with 7-8 panicle-bearing tillers and matures in 120 days. It possess long bold grains with an average test weight of 25.45 g. It is resistant to brown planthopper (BPH) and moderately resistant to sheath blight and sheath rot. The main advantage of this improved cultivar is its consistent yield output of more than 4 t/ha under very low inputs (N_2~ 40 kg/ha) and the fact that it outyields the parent variety by more than 0.5 t/ha in both dry and wet seasons.

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XA0054533

'NIAB MUNG 92' A HIGH YIELDING AND SHORT DURATION MUNGBEAN VARIETY

Induced mutations has played a significant role in the development of many crop varieties [2] and is instrumental in enhancing genetic variability. Induced mutations in mungbean at the Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan have been successful in developing mutant varieties 'NIAB MUNG' (NM) in an endemic cytoplasmic genetic background i.e. NM 28, NM 13-1, NM 19-19, NM 20-21, NM 121-25, NM 51 and NM 54.

To introgress large seed size into an adapted germplasm, hybridization [1] between a mutant line NM 36 and an AVRDC accession VC 2768B was initiated. The breeding efforts resulted in the development of a high yielding and short duration mungbean variety - NIAB MUNG 92. This variety has higher yield potential (2 tons/ha) with inherent earliness and uniform maturity. It produced 45% and 24% higher seed yield as compared to NM 121-25 and NM 51 respectively (Table 1).

Table 1. Yield performance of NIAB MUNG 92 in different sets of yield trials

Varieties	NIAB Farm Trials 1991-94	Multilocation Trials 1992-94	Average yield (kg/ha)	Yield increase (%)
NM 92	2164	1457	1811	-
NM 121-25	1281	1206	1244	45
NM 51	1609	1315	1462	24

Plants of this variety have an attractive large seed size (56 g/1000 seeds) and are insensitive to photoperiod with determinate plant growth habit. The variety is also resistant to field shattering and amenable to mechanical harvest. The variety translocates higher amounts of photosynthates to the seeds, resulting in high harvest index (Table 2) as compared to other mungbean varieties. Based on the superior yield performance of NIAB MUNG 92 the variety was approved in November 1996, by the Provincial Seed Council, for its general cultivation in the Punjab Province.

Table 2. Morpho-physiological traits of NIAB MUNG 92.

Trait	NIAB NUNG 92	NIAB MUNG 51	NIAB MUNG 121-25
Days to flower	32	40	45
Days to mature	58	68	71
Plant height (cm)	53	80	81
No. of pods/plant	38	38	37
Harvest Index (%)	37	25	23
1000 seed weight (g)	56	45	31
Seed Protein (%)	25.3	24.9	25.1
Reaction to diseases			
i) Mungbean Yellow Mosaic Virus	Highly Resistant	Highly Resistant	Moderately Resistant
ii) Cercospora Leaf Spot	Resistant	Tolerant	Moderately Susceptible

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TWELVE NEW GROUNDNUT (*Arachis hypogaea* L.) MUTATED GERMPLASM REGISTERED IN ICRISAT GENE BANK

XA0054534

Groundnut or peanut (*Arachis hypogaea* L.), is an economically important principal oilseed and cash crop which is cultivated on a large scale throughout the world. In India groundnut occupies 31.3% of the total cropped area under oilseeds (8.35 million hectares) and accounts for 36.1% of the total oil production (8.85 million tons). Many reasons are ascribed to the low productivity of this crop in developing countries. Early leaf spot (*Cercospora arachidicola*) and late leaf spot (*Phaeoisariopsis personata*) are the two most important fungal diseases of groundnut in India. Individually, or in combination, the two diseases can cause more than 50% yield loss [1]. Groundnut is grown in Tamil Nadu only under rainfed conditions during the rainy season in red soils of poor fertility; the crop is usually subjected to drought conditions with consequent reduction in crop yields. Although India has achieved great success in cereal production, there is a large gap between the demand and supply of edible oils. It is estimated that more than 32 million tons of oilseeds are needed every year [2]. One possible way to increase oilseed production is to include efficient oilseed crops in existing cropping system. A number of attempts have been made to produce disease resistant, drought tolerant and productive characters through conventional breeding methods, but the lines developed either possessed only a moderate level of resistance or retained one or

oilseed production is to include efficient oilseed crops in existing cropping system. A number of attempts have been made to produce disease resistant, drought tolerant and productive characters through conventional breeding methods, but the lines developed either possessed only a moderate level of resistance or retained one or more undesirable features. Hence, there is an urgent need to produce and identify new cultivars combining high level disease resistance, early maturity, desirable pod and kernel features, besides increased yield and oil content in groundnut. Induced mutations could form a potent tool to develop new genes for various characters of groundnut, particularly various degrees of resistance to biotic and abiotic stresses, pod number, seed yield and oil content, without radically altering the genetic background of the cultivars under study.

The seeds of important local cultivar VRI-2 ('Virdhachalam 2') with 48% oil content were selected for the present study. The seed materials were procured from Oilseed Research Station, Tamil Nadu Agricultural University, Virdhachalam. Well-developed, uniform and dry seeds, standardised for moisture content at 13% (with glycerol), were irradiated with gamma rays from a ^{60}Co source. The doses administrated were 50, 100, 150, 200, 250, 300, 350, 400, 450 and 500 Gy. The seeds were presoaked in distilled water for 8 h and were treated with various concentrations of ethyl methane sulphonate (EMS) and sodium azide (NaN_3) 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 mM which were prepared in a phosphate buffer solution of pH 6.0 and 3.0 respectively. Soon after the treatment, seeds were washed with running tap water for 4 h and sown on the experimental field along with respective controls. The unexposed dry seeds and seeds soaked in distilled water were served as control. Three hundred seeds for each treatment were sown on a field with 3 replicates each consisting of 100 seeds with a spacing of 30 cm between the rows and between plants during 1992. In M_1 generation, groups of plants were harvested on a bulk basis, while in M_2 generation single plants were selected and single progeny selection method was followed for improvement. One hundred and fifty normal looking plants from each dose/concentration were randomly selected and advanced to M_3 generation during 1993. In M_3, a group of 50 normal looking plants with visual observations for improvement in yield, oil and other economic characters were selected for each dose/concentration. Oil content was estimated by NMR analyser (TNAU, Coimbatore). Selected superior mutant progenies were evaluated in M_4 and M_5 during 1994-95 for yield, oil and other economic characters.

Several mutants from M_5 were isolated for various agronomic traits. Seeds of all these elite progenies showed an overall increase in oil percentage and seed yield over respective parents. Eighteen promising mutants were supplied to ICRISAT, Patancheru, India, during December 1995. Among them twelve mutants were registered as new germplasm by the ICRISAT Gene Bank (ICG15127 to ICG15138) (Table 1).

Two mutant lines of high yield and oil content, one mutant of disease and one of drought resistance and six mutants for pod, kernel and improvement of other characters were identified. The new mutant lines significantly outyielded over the parental variety and were superior in many agronomic traits. These elite lines are the potential candidates to introduce in the farming system of Tamil Nadu and other places of India. These germplasm are currently under advance evaluation trials in ICRISAT and other centres. For further information and for small quantities of seed contact: Dr. A.K. Singh, Senior Scientist (Germplasm), ICRISAT, Patancheru-502 324, AP, India (Fax:+91 40 241239; E-mail:ICRISAT@CGNET.COM).

Table 1. Description of new registered groundnut mutant lines and their yield and oil content

ICRISAT identity	Mutated character	Pod yield (g)	Oil content (%)
VRI-2 (Virdhachalam)		39.61	48.28
ICG 15127	High yield (500 Gy)	60.20*	57.62**
ICG 15128	Smooth surface pod (300 Gy)	48.40	53.69*
ICG 15129	Deep constriction pod (EMS 20 mM)	46.40	53.20
ICG 15130	High oil content (250 Gy)	62.20**	58.10**
ICG 15131	Smaller size pod (EMS 30 mM)	48.60	54.56*
ICG 15132	Disease resistance (NaN_3 15 mM)	52.60*	55.50*
ICG 15133	High yield (EMS 25 mM)	64.30**	52.48
ICG 15134	High oil content (EMS 5 mM)	60.50**	59.00**
ICG 15135	Bold pod (350 Gy)	44.80	53.90*
ICG 15136	Drought resistant (400 Gy)	57.80*	52.05
ICG 15137	Dwarf (NaN_3 35 mM)	61.60*	52.80
ICG 15138	Medium size pod (450 Gy)	63.80**	52.08

* and ** significant at 5% and 1% level, respectively (t-test)

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DECORATIVE PERICLINAL CHIMERAS OF *Weigela praecox* (Lemoine) Bailey CREATED BY CHEMICAL MUTAGENESIS

Achlorophyllous mutations are remarkable results of plant mutagenesis. They are very popular among gardeners as a component of ornamental periclinal chimeras with motley leaves ("*variegata*", "*marginata*" and "*media picta*"). The "*aurea marginata*" form of Weigela is well-known. If the periclinal chimera is possible because of the apex structure, then the periclinal chimera with the reverse combination ("*media picta*") is possible as well. To make such periclinal chimeras we have received achlorophyllous mutations in the experiment with growing of seedlings from *Weigela praecox* seeds treated with chemical mutagens. The solutions of N-methyl-N-nitrosourea (MNH) 0.02%, N-ethyl-N-nitrosourea (ENH) 0.05%, dimethyl-N-nitrosourea (dMNH) (0.1%) and 18 hours treatment were used. The year-old seedlings with minor achlorophyllous spots on the leaves were noted in M_1. We cut the shoots above the leaves with such spots for the germination of new shoots from the lateral buds. This process was repeated until chimeras became stable.

Two chimeras "*marginata*" and 3 chimeras "*media picta*" were obtained in the result. Several achlorophyllous mutations were fixed as internal elements in apexes of periclinal chimeras of the type "*chlorina media picta*" which were described firstly as belonging to *Weigela*. Because of availability of cells that are able to synthesis of the chlorophyll in the tunica of the apex, chimeras maintained a high viability of the wild genotype in combination with a high ornamentality of the motley leafed mutant. Mutations had been received in 1990 and fixed as periclinal chimeras in 1992 - 1993. These chimeras were multiplied by green cuttings in 1993 - 1995.

The best new form is a shrub up to 2 meters in height with opposite back-egg-shaped leaves 6-8 cm in length and 3-6 cm in width. Leaves are with a sparse pubescence, which is more pronounced on the lower surface. Flowers are lilac-pink tube-bell. Blooming is abundant. This form propagates easily by green cuttings with one or several internodes.

The 3 year old plants *Weigela praecox* "*chlorina media picta*"reach 0.5-0.7 m and at 4 years of age they bloom abundantly. The colouring of their leaves is the main difference of this form. In the middle part of all leaves there is an elongated light yellow-green spot of 2.5 - 4 cm with a clear boundary and curved edges. The spot is brighter on young leaves. The reddish colouring of one year shoots is the second peculiarity of this form. Ornamental properties of the new form are not kept by seed propagation as in all chimeras. New forms of Weigela praecox *"chlorina media picta*" differ from the wild genotype by a high ornamentality during the whole vegetation season, they are hardier than the known Weigela forms of the "marginata" type and can be widely used in greenbelt settings.

We had also selected several new mutant forms of *Pinus montana* and *Pinus sylvestris*. Cuttings and nursery products of new forms are proposed for exchange [1]. Our experience shows that new forms, created by the stabilisation of achlorophyllous mutations from M_1 as a component of periclinal chimeras with motley leaves ("*variegata*", "*marginata*" and "*media picta*"), increase the biodiversity of ornamental plants for greenbelt setting.

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IN VITRO MUTAGENESIS IN SUGARCANE CALLUS CULTURE

Tissue and cell culture are considered an important breeding tool for crop improvement. In vegetatively propagated species, mutagenesis combined with *in vitro* culture techniques may be the only method to improve existing cultivars. Tissue culture and mutagenesis were applied to increase variation in sugarcane by irradiation of dormant buds or cuttings and subsequent *in vitro* development of plants [1]. An alternative method, allowing mutagenic treatment of large numbers of individuals, is *in vitro* mutagenesis of somatic callus or cell suspension followed by regeneration and selection of desired mutants [2]. The study was designed to determine conditions for chemical and physical mutagenesis of sugarcane somatic callus. Callus culture was established from shoot-explants of 8-week old, pot grown, sugarcane (*Saccharum officinarum* L.) plants. The BL4 clone was used, obtained from Dr. S.H. Siddique, Atomic Energy Agricultural Research Centre, Tandojam, Pakistan. The explants of the innermost 5-6 tightly furled leaves were placed on induction agar medium consisting of Murashige and Skoog [3] salts and vitamins, 4 mg/l 2,4-D and 20 g/l sucrose. The induction of callus took place in a dark room, at 28^0C. After one month of culture, the actively growing callus tissues were separated from the explants and treated with physical or chemical mutagens. Two doses of gamma-rays (10 and 15 Gy) or 2 mM solution of N-nitroso-N-methylurea (MNH) from Serva were applied for 2 and 3 hours. After irradiation, part of the calli were directly transferred to fresh, non-irradiated medium (IC method). The remainder of the calli were kept on irradiated medium for 4 weeks as they were subcultured onto fresh medium just one day before irradiation (ICM method). For each mutagen and dose tested, 10 samples of callus (about 1 g of fresh weight each) were placed on induction medium (one sample per petri-dish). After one-month culture on induction medium, the fresh-weight gain of every callus sample was measured and the culture was transferred onto regeneration medium in which 2,4-D was replaced by 2 mg/l kinetin; 2 mg/l IBA and 2 mg/l IAA. After 8-week culture on regeneration medium, data on regeneration capacity of calli were recorded. Additionally, the frequency of shoots displaying chlorophyll deficiency (chlorophyll variants) among regenerants was estimated.

The inhibition of callus growth in relation to the control was observed in every treated culture and reduction in fresh-weight gain ranged from 42 to 91% (Table 1). The influence of both mutagens in all doses applied was also noticed at the level of plant regeneration. In the most harmful combinations (15 Gy - ICM and MNH 2 mM x 3 h) a 72% reduction in regeneration ability was observed. An increase in the frequency of chlorophyll deficient variants was recorded after all doses of MNH or gamma-rays used. In all treated combinations a 1.5 - 8 fold increase over the control

in frequency of variants, was noticed. The highest frequency of chlorophyll-deficient variants, up to 8.0%, was induced when callus and medium were exposed to 15 Gy gamma-rays. A similar frequency of chlorophyll shoots (7.8%) was observed in culture treated with 2 mM x 3 h of MNH. The most frequently observed variants were albino shoots but a few light-green or striped shoots were also noticed. It was noticed that after the same gamma-ray doses, the reduction in fresh-weight gain of callus and number of regenerated shoots was higher when irradiated callus was cultured for a subsequent 4 weeks on irradiated medium (ICM) than when it was transferred onto fresh, non-irradiated medium (IC). The difference between these two regime treatments was also noticed in the frequency of chlorophyll variants, as 10 Gy of gamma-rays resulted in 3.3 and 5.0% and 15 Gy caused 5.0 and 8.0% of chlorophyll deficient shoots in IC and ICM combinations, respectively. Hundreds of sugarcane plants regenerated from control and gamma-rays or MNH-treated calli were planted in the field to estimate the frequency of variation in morphological characters.

Table 1. The influence of gamma-rays and MNH treatment on callus and regeneration ability of sugarcane

Treatment	Dose	Fresh-weight gain of callus (g)	Callus growth reduction (%)	Regenerated shoots (No.)	Regeneration ability reduction (%)	Frequency of chlorophyll-deficient variants (%)
			Gamma-rays (Gy)			
	0	0.66 ± 0.35	0	1351	0	2.1
IC	10	0.39 ± 0.18	42	847	27	3.3
	15	0.16 ± 0.05	77	486	65	5.0
	0	0.65 ± 0.12	0	925	0	1.0
ICM	10	0.08 ± 0.02	88	350	62	5.0
	15	0.05 ± 0.02	91	258	72	8.0
			MNH (mM x h)			
	0	0.54 ± 0.12	0	1092	0	0.9
	2 x 2	0.15 ± 0.05	72	398	64	4.5
	2 x 3	0.06 ± 0.02	89	305	72	7.8

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XA0054537

IN VITRO INDUCED VARIATION FOR SELECTION OF VALINE RESISTANCE IN CALLUS CULTURE OF *Arabidopsis thaliana* (L.) Heynh.

Studies on selection of valine-resistant tobacco mutants indicated the high efficiency of recovery of such mutants [1]. The presented study has been designed to screen for mutants of *Arabidopsis thaliana* (L.) Heynh. in callus culture derived from mutagenised leaf-explants. This system combines *in vitro* mutagenesis followed by selection of culture growing in the presence of a toxic concentration of valine. Two ecotypes of *A. thaliana* C-24 and RLD were used in the study. Culture of arabidopsis leaf-explants followed the regeneration system proposed by [2]. The protocol includes 5-7 day culture of explants in liquid induction medium and then culture on agar shoot regeneration medium. The culture was established from untreated (control) and mutagenised explants. N-nitroso-N-methylurea (MNH, Serva) was applied as a chemical mutagen. Small rosette-leaves or fragments of bigger leaves (0,5-1,0 cm^2) excised from donor plants were incubated for 3h on the gyratory shaker in callus induction medium containing three concentrations of filter-sterilised MNH (0,25; 0,5 and 1,0 mM) and thoroughly rinsed in the mutagen-free-medium. For physical mutagenesis, gamma radiation (^{60}Co, intensity 64 rad/sec) at two doses (15 and 30 Gy) was applied to plants growing on MS10 medium. On the same day the explants were excised from irradiated plants to start the culture.

In spite of a very strong toxic effect of valine on germination and growth of *A. thaliana* plants, no distinct influence of 10 mM valine was noticed on callus-formation ability of cultured leaf-explants (Table 1). The high efficiency of callus formation was observed in all combinations studied. Over 88 and 91% of explants, for C-24 and RLD respectively, developed callus tissue. The strong inhibitory effect of valine on shoot regeneration ability was more pronounced in culture derived from untreated explants than in culture of mutagenised explants. The highest number of calli able to develop shoots (10.8%) was induced after gamma-irradiation with 15 Gy. Among MNH doses applied the most efficient for both genotypes was 0.5 mM x 3h which induced shoot regeneration in 6.4 and 5.7% of calli for C-24 and RLD genotype, respectively. In total, 924 control and 2077 mutagen-treated explants were subjected to selection on valine-containing regeneration medium and 17 and 294 shoots were regenerated, respectively (Table 2). The average frequency of regenerated shoots was much higher in cultures established from mutagenised explants and reached 14.2 shoots per 100 calli in comparison to 1.8 shoots in the control culture. Although in total, over 300 shoots were regenerated on selection medium, a vast number of them failed to grow after transfer onto rooting medium. Only 28 of the regenerants selected on valine-medium developed seeds. Two fertile arabidopsis shoots were developed under valine selection without any mutagenic treatment, as a result of somaclonal variation. The phenomenon of somaclonal variation has been described as a source for *in vitro* regenerated mutants or variants

in many species [3] including *A. thaliana* [4]. However, the data obtained in the presented study on regeneration frequency in culture established from mutagenic treated explants has indicated a higher efficiency of selection and mutant/variant recovery when *in vitro* culture was combined with chemical or physical mutagenesis.

Table 1. Callus formation and shoot regeneration efficiency from untreated and mutagenised *A. thaliana* explants under selection on valine medium

Genotype	Dose of mutagen	No. of explants	Explants forming calli (%)	Regenerating calli No.	Regenerating calli (%)	Regenerated shoots (No.)
C-24	-	488	88.3 ± 7.9	2	0.5	4
RLD	-	436	91.2 ± 6.0	5	1.3	13
			MNH (mM x 3 h)			
C-24	0.25	263	84.1 ± 6.6	8	3.6	25
	0.5	448	78.2 ± 10.5	23	6.4	108
	1.0	288	74.3 ± 8.8	4	1.9	16
RLD	0.25	207	88.4 ± 5.6	5	2.1	8
	0.5	345	81.7 ± 7.1	16	5.7	49
	1.0	325	73.8 ± 7.6	6	2.5	17
			Gamma - rays (Gy)			
C-24	15	166	82.9 ± 8.6	16	10.8	26
	30	163	64.1 ± 9.6	5	4.8	14
RLD	15	186	84.5 ± 9.9	2	1.3	14
	30	162	68.1 ±4.7	7	6.4	17

Table 2. The effect of mutagenic treatment on regeneration ability of *A. thaliana* callus culture under selection on valine medium

Culture	No. of calli under selection	No. of regenerated shoots Total No.	No. of regenerated shoots Per 100 calli	Fertile R_1 plants No.	Fertile R_1 plants (%)
Untreated	924	17	1.8	2	11.8
MNH-treated	1558	223	14.3	19	8.5
Gamma-ray treated	519	71	13.7	7	9.9
Mutagenised (Total)	2077	294	14.2	26	8.8

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INDUCTION OF RESISTANCE TO BLAST DISEASE IN RICE CULTIVAR 'IR 50'

The elite cultivar, 'IR 50' was developed at the International Rice Research Institute and was released in India for the State of Tamil Nadu in 1982. It is highly responsive to fertilizer, records high yields and possess good grain characters. It matures in just more than 100 days and is ideal for both samba and navarai seasons in Tamil Nadu. The cultivar was shown to be highly susceptible to blast (*Magnaportha grisea*) causing extensive losses. This generated the need for improved resistance to blast in this cultivar. One of the most promising techniques for producing disease resistant forms of plants is the use of mutagenic agents.

With the objective of developing high yielding blast tolerant mutant lines from IR 50, the mutation approach was adopted. Gamma-rays from ^{60}Co and chemical mutagens (ethyl methanesulphonate - EMS and sodium azide - NaN_3) were employed on dry seeds for induction of mutations. The M_1 generation was grown in 1983. In 1984, more than 100,000 plants were grown in M_2 generation. One hundred and sixty eight M_2 derived families were grown in M_3 generation in 1985. For further evaluation in M_4 and M_5 generations, 128 M_3 mutant lines were selected. Based on yield and other attributes, a total of 85 mutant lines were finally selected. All selected mutants resemble the parent for both agronomic and quality characteristics.

The evaluation of these mutant lines for their level of tolerance to blast disease was conducted at the Central Rice Research Institute (CRRI) over a number of years under both artificial and natural conditions. These mutant lines showed varied levels of tolerance to blast in comparison to the parent cultivar. To confirm these findings the mutants were tested at different 'hot spot' locations of blast like Hazaribag in Bihar, Maruteru in Andhra Pradesh and Jagdalpur in Madhya Pradesh. In addition, they were also screened under greenhouse conditions at the Directorate of Rice Research, Hyderabad. Experimental data from all these centers support the earlier finding that variation for tolerance to blast exists in these mutant lines.

The mutant lines were further evaluated under artificial screening at CRRI and highly tolerant individual plants with individual scores of 1 and 2 as against the parent score of 7 to 9 (in the IRRI disease score scale of 1 to 9) were selected. After seed multiplication, yield evaluation trials were conducted on fourteen different individual plant derived lines. The field evaluation data on the selected fourteen mutant lines i.e. CRM 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54 and 58 indicate that all these mutant lines will yield either at par to the parent, or higher.

The mutants were further tested for their suitability in the replacement of the parent variety in the State of Assam. In the yield evaluation and adaptation trials conducted at Kokilabari Farm, Assam, the mutants performed consistently giving a yield of over 3 t/ha. Further evaluation of CRM mutant lines over a four year period at Regional Agricultural Research Station, Assam Agricultural University, Diphu, Assam revealed that three mutants 'CRM 49', 'CRN 51' (after sodium azide

treatment - 0.001 M), and 'CRM 53' (after EMS treatment - 0.66%) consistently yielded double that of the parent (1.25 t/ha for parent). Further, in the trials conducted at the Zonal Agricultural Research Station of Indira Gandhi Krishi Viswa Vidyalaya, Jagdalpur, the CRM mutants performed well on both the yield and the disease scores. Based on the performance of these mutants, the Government of Assam is proposing the release of three mutants namely, CRM 49, 51 and 53 and wishes to replace the parent cultivar IR 50 with these high yielding and blast tolerant mutants.

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RESULTS AND PROSPECTS OF THE USE OF MUTANTS IN SUNFLOWER BREEDING

Modern agriculture requires a significant yield increase of farm crops. In the case of sunflower, this problem may be solved by developing varieties and hybrids with new plant architecture, tolerant to high density conditions. Some mutants were developed to solve this problem by long-term breeding through chemical mutagenesis (1980-1995). Among them were mutants M-2006, M-2007, M-2008, which are characterized by a short leaf petiole, ranging from 1.3 to 5.8 cm, and with increased tolerance to high plant density (80-150 thousand plants/hectare). In the mutant M-1991 the petiole is absent completely.

Super-early, cold tolerant, sunflower mutants (M-1700, M-1925, M-1927) which are resistant to high density growing conditions (100-120 thousand plants/hectare) and with a vegetation period of 45-75 days are of great interest for northern regions throughout the world. Their usage makes it possible to obtain two yields per season. Other mutants (M-1584, M-1155, M-1624, M-1967) are very important for sunflower breeding. They possess a unique combination of dwarfness (35-140 cm) and large seed size (50-53% increase).

The above-mentioned, as well as other valuable mutants, have compact growth habit, short petiole, small leaves, dwarf and erect growth and are resistant to lodging and stalk breakage. They give an increment yield up to 10 t/ha under high density plant stands. Promising mutants were tested in various agroecological zones of Ural, Novosibirsk, Moscow, Byelorussia and Kuban. Results of the two mutants trial performed in 1995 on the Ust-Labinsk (Krasnodar Territory) testing plot are presented in Table 1. Mutants M-1925 and M-1927 were tested in 35x35 cm sowing density, and plant stand was 100 thousand plants/ha.

Through the application of chemical mutagens we have succeeded in changing the ratio of vegetative and generative organs biomass to the total biomass (Table 2). In this way we contributed to solving some significant problems of sunflower breeding.

Table 1. Performance of new sunflower mutants in advance trials

Mutant, hybrid	Vegetation period (days)	Plant height (cm)	Seed yield (t/ha)	Oil content (%)
M-1925	62	140	2.39	51.5
M-1927	70	121	3.08	51.8
Cargil 187	114	133	3.60	49.0
Cargil 207	114	175	3.82	49.2
Zemundo 1213	117	196	4.45	48.9
Yubileiny 60	124	213	3.65	53.6
Rodnick (st)	82	149	3.38	52.0
LSD 0.05			0.28	1.2

Table 2. The ratio of vegetative and generative organs biomass to the total biomass (dry matter content)

Mutant, variety	Seeds	Stalk	Leaf	Calathide	Leaf petiole
M-1927	35.8	29.3	15.6	16.2	0.0
M-1991	39.0	30.8	16.7	13.5	0.0
Rodnick (st)	29.2	31.6	12.2	23.5	3.5

*(Contributed by **KALAIJUN, A.A.,** Seed Company "Mutant", Krasnodar, Russia)*

MODIFICATION OF A RAPID SCREENING METHOD OF RICE MUTANTS FOR NaCl TOLERANCE USING LIQUID NUTRIENT CULTURE

The isolation or identification of mutants requires an efficient screening method. The screening technique must be reliable and able to evaluate large amounts of mutated material. Salinity screening under field conditions is often inaccurate and difficult due to strong environmental effects. The screening method developed by the International Rice Research Institute (IRRI) for selecting salt tolerance at the seedling stage was evaluated in the greenhouse of the FAO/IAEA Laboratory, Seibersdorf, Austria.

The set-up developed at IRRI includes styrofoam floats (36.5 x 26.5 cm) having 100 circular holes (10 x 10 milimeters) and a nylon net bottom, placed on top of a rectangular 18 litre plastic tray. However, the set-up with styrofoam can be washed only a few times, is not very stable and can screen only a limited number of seedlings. After 2-3 times of use, the seedling float becomes contaminated with algae and needs to be replaced.

Modifications were done to make this screening method applicable to evaluate a large mutated plant population. The set-up includes PVC (polyvinylchloride) plates (56 x 36 cm) with 24 rectangular holes (6 x 7 cm) and a nylon net bottom, placed on top of a rectangular 25 litre plastic tray. The plastic seedling plate can be used unlimited times and is easy to handle. It is washable with any detergent and even with a hard brush to remove algae or other contamination. The plastic plate is hard and stable and it can carry heavy weight. It can accommodate more than 30

seedlings per hole (total of more than 700 seedlings) as compared to 1-2 seedlings per hole with the old set-up (total of 100-200 plants).

Using the modified set nine different varieties of known levels of tolerance (Susceptible: 'IR29', 'PP2462-11', 'Taipei 309' and 'Wagwag'; Moderate: 'IR58430-6B-14-1-2', 'IR51500-AC-11-1' and 'IR63731-1-1-4-3-2' and Tolerant: 'Nona Bokra' and 'Pokkali') (Table 1) were tested in five different salinity levels (electrical conductivity - EC 2, 6, 8, 10 and 12 dS/m) to determine the optimum salinity level and best time for scoring injury symptoms. Seeds were pre-germinated for two days then sown on the seedling plate on the tray filled with distilled water. Salinity was introduced after three days by adding NaCl to the nutrient solution described by Yoshida *et al.* [2] up to the desired EC. The pH was maintained daily at 5.5 by adding 1*N* of NaOH or HCl.

Salinity injury rating was based on visual symptoms (1- tolerant and 9 - sensitive) at seedling stage, according to the modified Standard Evaluation System of IRRI Gregorio *et al* (1997). Salinity injury was scored four times: 5, 7, 9 and 12 days after salinization. The screening was done in the greenhouse with day/night temperatures of 30/20 ^{0}C and relative humidity of at least 50% during the day. The experiments were repeated three times using ten seedlings of each variety per replication.

Salinity tests of EC 6 dS/m showed very low salt injury even after 12 days. The varietal differences, were observed 9 days after salinization at EC 8 dS/m. The sensitive check IR 29 had an average score of 6.6 which could be classified as moderate.

At EC 10 dS/m and 12 days after salinization, a clear distinction of the three categories (tolerant, moderate and susceptible) of the varieties tested was established. Results showed that the optimum salinity level for screening rice in the greenhouse of the FAO/IAEA Laboratory, Seibersdorf, should be EC 10 dS/m and that visual symptom scoring be done starting 12 days after salinization.

Most varieties were severely injured at EC 12 dS/m within 10 days of salinity stress, except the two tolerant checks Pokkali and Nona Bokra. After 20 days at this salinity level, all varieties had died, including the tolerant varieties. Thus EC 12 dS/m was too high for isolating the moderately tolerant lines.

Our results, using plastic plates, were similar and confirm the results obtained at IRRI using styrofoam floats. Moreover, our method offers the screening of large numbers of seedlings in a short time and it is cheap in the long run. Presently this technique is being used for screening advanced mutated populations of rice for selection of mutants at the seedling stage. This technology is being transferred to National Programmes in FAO/IAEA Member States through scientists participating in our training programme.

Note: 10 dS/m is approximately 6.4 gm/l NaCl.

Table 1. Rating of different rice varieties at various salinity levels and time of rating using the Standard Evaluation System of IRRI (1= no symptoms, 9 = dead seedlings)

Variety	Days after salinization	Salinity levels (dS/m)				
		2	6	8	10	12
IR 29	5	1.2*	2.3	3.1	3.6	4.7
susceptible	7	1.5	4.3	5.6	5.7	7.8
	9	1.6	5.8	6.6	6.9	8.5
	12	1.6	7.3	7.7	7.8	9.0
Wagwag	5	1.1	2.3	2.3	2.8	3.3
susceptible	7	1.2	3.0	3.9	4.2	6.0
	9	1.4	3.9	4.7	5.3	7.9
	12	1.4	5.0	7.1	7.3	8.9
Taipei 309	5	1.0	1.2	1.8	3.0	3.6
susceptible	7	1.0	1.7	5.2	5.3	6.3
	9	1.0	2.3	7.2	7.5	8.0
	12	1.0	3.4	7.5	9.0	9.0
PP2462-11	5	1.0	1.1	1.3	1.8	3.0
susceptible	7	1.0	1.2	1.8	2.3	4.4
	9	1.0	1.3	3.1	3.2	5.6
	12	1.0	2.4	3.9	4.3	7.9
IR58430-6B-14-1-2	5	1.0	1.0	1.0	1.5	1.5
moderate	7	1.0	1.2	2.2	2.4	3.5
	9	1.0	1.8	3.3	3.4	4.9
	12	1.0	2.4	4.1	5.0	6.6
IR51500-AC11-1	5	1.0	1.8	2.5	2.8	4.5
moderate	7	1.0	3.3	3.4	3.6	5.7
	9	1.3	4.0	4.2	4.5	7.3
	12	1.6	4.4	5.6	5.8	8.4
IR63731-1-1-4-3-2	5	1.0	1.0	1.5	1.6	1.9
moderate	7	1.0	1.7	2.2	2.6	3.4
	9	1.0	1.7	2.4	3.2	4.4
	12	1.0	2.7	4.0	4.4	5.3
Nona Bokra	5	1.0	1.0	1.0	1.0	1.4
tolerant	7	1.0	1.1	1.6	1.9	2.3
	9	1.0	1.2	1.8	2.2	3.2
	12	1.0	1.6	2.5	3.0	3.8
Pokkali	5	1.0	1.0	1.2	1.2	1.4
tolerant	7	1.0	1.3	1.5	1.8	2.2
	9	1.0	1.5	1.8	2.1	3.4
	12	1.0	1.6	2.5	2.8	3.7

*Average score from 30 seedlings

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HIGH FREQUENCY OF MUTATIONS AFTER MUTAGENIC TREATMENT OF BARLEY SEEDS WITH NaN_3 AND MNH WITH APPLICATION OF INTER-INCUBATION GERMINATION PERIOD

N-methyl-N-nitrosourea (MNH) and sodium azide (NaN_3) belong to the most potent chemical mutagenic agents. The high mutagenic effect of MNH can be significantly increased by application of 5-32 hours of germination prior to the treatment of pre-soaked seeds [1], and even further, by application of double treatment with inter-incubation germination period (iig) between them [2].

Sodium azide, a respiration inhibitor is metabolized *in vivo* to a powerful chemical mutagen in many plant species, including barley, rice, maize and soybean [3]. The mutagenic effect of sodium azide greatly depends on the pH of treatment solutions and similar to the MNH, can be further increased by pre-germination of barley seeds prior to NaN_3 treatment [4]. In the experiments presented below the efficiency of single, double and combined treatment of sodium azide and MNH in barley was compared. Seeds of barley variety 'Aramir' were pre-soaked in distilled water for 8 hours and mutagenically treated either with MNH or sodium azide for 3 hours. Sodium azide treatment was carried out in a phosphate buffer at pH = 3. In combinations with double treatment, seeds after the first treatment, were rinsed several times in tap water and germinated on trays with filter paper for 6 hours before a second 3 hour treatment with mutagen.

The highest frequency of chlorophyll mutations was observed after combined treatment with 1.5 mM NaN_3 followed by 0.7 mM MNH with 6 hour inter-incubation germination period between treatments (Tab. 1). In this combination 6.4% of chlorophyll mutations in M_2 was obtained, with a fertility reduction of M_1 plants lower than after the double treatment with MNH.

Similarly, a very high frequency of point mutations was observed for other barley genotypes after combined treatment with NaN_3 and MNH. On an average, 5.6% chlorophyll seedlings were found in M_2 derived from this treatment for 6 barley varieties tested and about 30-50% of M_1 plants carried a chlorophyll mutation (Tab. 2). It should be noted that the reduction of M_1 plant fertility did not exceed 55%, which makes this combination a very efficient treatment for induction of point mutations in barley. It was proven that the combined treatment of NaN_3 and MNH yielded a wide spectrum of gene mutations in many barley genotypes, including dwarf and semi-dwarf characters, changes in root system development and structure. Also, mutants with an increased level of tolerance to Al^{+3} toxicity were selected [5]

and high frequency of DH barley mutants were obtained using this procedure and anther culture [6].

Table 1. Mutagenic effect of single, double and combined treatments with MNH and sodium azide applied on barley seeds variety 'Aramir'

Treatment	M_1 seedlings emergence reduction (%)	M_1 plant height reduction (%)	M_1 plant fertility reduction (%)	M_2 seedlings tested (No.)	M_2 chlorophyll mutants (%)
0.7 mM MNH	27.9	9.1	29.2	5083	1.5
0.7 mM MNH - 6h iig - 0.7 mM MNH	27.6	27.2	84.5	4501	4.0
.12 0.75 mM Na N_3	17.3	4.2	48.5	4732	3.8
0.75 mM NaN_3 - 6h iig - 0.75 mM Na N_3	32.5	4.7	48.8	4677	4.2
0.75 mM NaN_3 - 6h iig- 0.7 mM MNH	18.9	6.0	62.4	3932	4.5
1.0mM NaN_3 - 6h iig - 0.7 mM MNH	22.6	12.4	76.7	4427	6.4

The following protocol can be recommended as a highly efficient method of mutagenic treatment in barley:

- pre-soaking of seeds in distilled water - 8h
- treatment with 1.5 mM sodium azide (pH=3, phosphate buffer) - 3h
- inter-incubation germination period (seeds on trays with wet filter paper) - 6h
- treatment with 0.7 mM N-methyl-N-nitrosourea (MNH) - 3h.

Table 2. The frequency of chlorophyll mutations after combined treatment of NaN_3 and MNH in different barley varieties

Genotype	M_1 plants tested (No.)	M_1 plants carrying chlorophyll mutations (%)	M_2 seedlings tested (No.)	M_2 chlorophyll mutants (%)
Apex	710	51.0	14,195	6.3
Bielik	732	47.0	11,102	5.4
Dema	3220	35.0	43,673	5.2
Grosso	980	28.6	9,219	5.3
Lot	740	52.0	14,001	6.3
Rudzik	1617	39.0	9,457	5.0
average		*42.0*		*5.6*

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LIST OF NEW MUTANT CULTIVARS

The Plant Breeding and Genetics Section of the Joint FAO/IAEA Division undertakes the collection and dissemination of information on commercially used agricultural and horticultural cultivars developed through the utilization of induced mutations.

This list does not claim to be compprehensive. Its content is strictly based on information transmitted by breeders themselves and/or other institutions involved. Listing of a cultivar does not imply its recommendation by FAO/IAEA.

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Agropyron cristatum L. Gaertner (crested wheatgrass)			
CD-II	USA, 1996 Asay, K. Forage & Range Res. Lab. Utah State Univ. Logan, UT 84322-6300	cross colchicine induced tetraploid from 'HYCREST' x natural tetraploid	vigour
Arachis hypogaea L.	***(groundnut)***		
8130	China, 1988 Qiu Qingshu *et al.* SPRI, Laixi, Shandong 266601	cross F_1 (Luhua 4 x RP1) x RP1 (treated with gamma rays, 250 Gy)	seed quality yield
Huayu 16	China, 1996 Qiu Qingshu et al. SPRI, Laixi, Shandong 266601	cross	yield seed quality
Luhua 13	China, 1991 Qiu Qingshu *et al.* SPRI, Laixi, Shandong 266601	cross [F_1(Changda 6 x MA 143) x Baisha 1016] x Luhua 6	yield seed size
Luhua 15	China, 1994 Qiu Qingshu *et al.* SPRI, Laixi, Shandong 266601	cross F_2(7896 x Runner) x Irradiated Runner (250 Gy gamma rays)	seed quality earliness

TG-22	India, 1994 Mouli, Chandra, BARC, NABD Trombay, Mumbai	cross Robout-33-1 x TG-17	yield
TG-26	India, 1996 Mouli, Chandra *et al.* c/o BARC-NABD Trombay, Mumbai	cross JL-24 (gamma rays, 1983) xTG-23 (TGS-2 x TG-1(x-rays, 1958))	yield semi-dwarfness
TKG-19A	India, 1996 Mouli, Chandra BARC Nucl. Agr. Biot. Div Trombay, Mumbai	cross TG-17 x TG-1	seed size
Avena sativa L.	***(oat)***		
Bay	USA, 1995 Forsberg, Käppler, Dürst Dept. of Agronomy Univ. of Wisconsin Madison	cross Hazel/6/Holden/ Irr. 4 (thN irradiated)/Garland/2/ 6x amphiploid/2CIav 6936/3/Garland/5/Noble	disease resistance yield
Belle	USA, 1995 Forsberg, Käppler, Dürst Univ. of Wisconsin Madison	cross Don/7/Ascensao/Fayette/4/Clintland/3/Garland/ Hawkeye/ Irr. 4 (thN irradiated/Victoria/5/Goodfield)	disease resistance lodging resistance yield
Gem	USA, 1996 Forsberg, Käppler, Dürst Dept. of Agronomy Univ. of Wisconsin	cross MO 0768/6/Holden/ Irr. 4 (thN irradiated)/Garland/2/ 6x amphiploid/s*Ciav 6936/3/Garland/5/Froker/.	disease resistance yield

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Brassica napus L.	***(rapeseed)***		
Abasin-95	Pakistan, 1995 Syed Anwar Shah PAEC-NIFA, P.O. Box 446, Peshawar	gamma rays, 1400 Gy [Tower]	earliness yield adaptability
Binasharisha-3	Bangladesh, 1997 Rahman, A., Das, M.L. BINA, P.O.Box 4, Mymensingh	gamma rays, 800 Gy	oil content earliness
Binasharisha-4	Bangladesh, 1997 Rahman, A., Das, M.L. BINA, P.O.Box 4, Mymensingh	gamma rays, 800 Gy	oil content earliness
Capsicum annuum L.	***(pepper)***		
Gornooriahovska kapia	Bulgaria, 1997 T.Christov, S.Daskalov *et al.* Vegetable Research Station G. Oriahovitza Institute of Genetics	cross Zlaten medal ms-8 (gamma rays,135 Gy-1973) x GO-201B	earliness yield
Chrysanthemum sp.	***(chrysanthemum)***		
Raktima	India, 1998 Datta, S.K. Jugran, H.M.	gamma rays [Shyamal]	flower colour
Repin Rosa	Brazil, 1996 Latado, R. Radiogenetics Section CENA-USP Caixa Postal 96	gamma rays, 8 Gy	flower colour

Cicer arietinum L.	***(chickpea)***		
NIFA-95	Pakistan, 1995 PAEC-NIFA, Peshawar	gamma rays, 200 Gy [line 6151]	blight resistance
Citrus limon L. Burm.	***(lemon)***		
Eureka 22 INTA	Argentina, 1987 Zubrzycki, H. INTA-Bella Vista, CC.#5,Col 3 de Abril 3432 Bella Vista	x-rays, 10 Gy [Frost Eureka]	fruit set fruit quality
Citrus sinensis L. Osbeck	***(orange)***		
Valencia 2 INTA	Argentina, 1987 Zubrzycki, H. INTA-Bella Vista, CC.#5,Col 3 de Abril 3432 Bella Vista	x-rays, 20 Gy [Valencia Late]	fruit set fruit quality
Corchorus capsularis L.	***(jute)***		
Binadeshipat-2	Bangladesh, 1997 Saha, C.S. BINA, P.O. Box 4, Mymensingh	NaN_3, 12mM [CVL-1]	fibre yield
Cynodon sp.	***(bermuda grass)***		
Tift 94	USA, 1995 Hanna, W.W. University of Georgia Dept.of Crop&Soil Sc Griffin,GA 302231797	gamma rays, 80 Gy [Midiron]	leaf quality insect resistance

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Eremochloa ophuiroides Hack	***(centipedegrass)***		
Tifblair	USA, 1995 Hanna, W.W. University of Georgia Dept.of Crop&Soil Sc Griffin,GA 302231797	gamma rays, 120 Gy	vigour quality
Eustoma grandiflorum	***(eustoma)***		
Purple Fantasy	Japan, 1996 Nagatomi, S. Inst.of Radiation Breed. NIAR-MAAF Ohmiya-machi,Ibaraki	gamma rays chronic, 0.25 - 1.5 Gy [Pastel Murasaki]	flower size
Purple Robin	Japan, 1996 Nagatomi, S Inst.of Radiation Breed. NIAR-MAAF Ohmiya-machi,Ibaraki	gamma rays chronic, 0.25 - 1.5 Gy [Pastel Murasaki]	flower colour flower size
Red Robin	Japan, 1996 Nagatomi, S. Inst.of Radiation Breed. NIAR-MAAF Ohmiya-machi,Ibaraki	gamma rays chronic, 0.25 - 1.5 Gy [Morgen Rot]	flower size flower colour
Glycine max L.	***(soybean)***		
Heinong 34	China, 1988 W. Wang	cross Heinong 16 x Tokachinogaha	yield protein content

	Soybean Institute HAAS		
Heinong 35	China, 1990 W. Wang Soybean Institute HAAS, Heilongjiang	cross Heinong 16 x Tokachinogaha	yield protein content
Heinong 41	China, 1997 W. Wang Soybean Institute HAAS, Heilongjiang	cross Ha-Co73-8955 (gamma rays induced mutant of Fengshan) x Ha90-9825 (thN induced mutant)	seed size protein content disease resistance
M-103	Vietnam, 1986 Tran Dinh Long Vietnam Agr. Sci. Inst. Thanh Tri, Hanoi	gamma rays, 50 Gy and EI, 0.01%	yield lodging resistance grain quality
Sui Nong 12	China, 1996 Lui Dechang, Jian Chenxi Suihua Agric. Res. Institute HAAS, Heilongjiang	gamma rays, 120 Gy [F_6 (Suijio 83-432 x (Heihe 4 x Te 7604))]	yield
Hordeum vulgare L.	***(barley)***		
Akdeniz M-Q-54	Turkey, 1998 M.I. Cagirgan Akdeniz University Ziraat Fakultesi P.O.Box 510, Antalya	gamma rays, 150 Gy [Quantum]	drought tolerance cold tolerance

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Kormovy	Ukraine, 1997 O. Zayats, M. Pavlishin Inst. of Agric. and Animal Biology 292084 Obroshynov Pustomyty district, Lviv Regio	EI, 0.005%	yield
Kosmos	Poland, 1977 Slabonski, A.	cross Pallas x Firlbeck Union	semi-dwarfness
Humulus lupulus L.	***(hop)***		
Santiam	USA, 1998 J. Henning & A. Haunold USDA-ARS & TSARC	cross Tettnanger x USDA 21618M (=Hallertauer mittelfrüh x USDA 21381M)	oil quality yield
Ultra	USA, 1995 Haunold, A. Oregon State University Dept. Crop&Soil Sci. Corvallis, OR	cross Hallertauer mittelfrüh (colchicine induced tetraploid) x Saazer (diploid)	yield
Lens culinaris Medik.	***(lentil)***		
Mutant 17 MM	Bulgaria, 1999 Miho Iliev Mihov Inst. for wheat & sunflower Doubroudja Gen. Toshevo 9520	gamma rays, 40 Gy	seed size pod size

Linum usitatissimum L.	***(flax)***		
Linola 989	Canada, 1996 Dribnenki, J.C.P. United Grain Growers Ltd. Linola Breeding Prog Morden, Manitoba	cross McGregor/Zero (mutant from 'Glenelg')//CPI84495/3/McGregor	oil quality
Manihot esculenta L. Crantz	***(cassava)***		
Tekbankye	Ghana, 1997 O. Safo Kantanka Dept. of Crop Science Univ. of Sci & Tech. Kumasi	gamma rays, 25 Gy [Isunikakiyan]	cooking quality vigour
Mentha arvensis L.	***(peppermint)***		
TN-8	Vietnam, 1995 Phan Phai	gamma rays [NV-74]	oil quality disease resistance pest resistance
Musa sp.	***(banana)***		
Novaria	Malaysia, 1993 F. Novak & C. Mak	gamma rays, 60 Gy [Grand Naine]	earliness fruit quality
Oryza sativa L.	***(rice)***		
A-201	USA, 1996 Tseng, S.T., McKenzie, CCRRF & CAES & USDA-ARS Biggs, California	cross L-202/PI 457920//L-202 (PI 457920 is a semi- dwarf mutant from 'Basmati 370')	semi-dwarfness
Binadhan 6	Bangladesh, 1998 BINA P.O. Box 4, Mymensingh	gamma rays, 300 Gy [F_2 of (Iratom 24 x Dular)]	yield

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Cilosari	Indonesia, 1996 Mugiono Nat. Atomic Energy Agency Jakarta	cross SM 268/PsJ (mutant from 'Seratus Malami' (200 Gy gamma rays)) x IR 36	yield
IRI 307	Korea, 1970 KAERI	thN, 15 x 10^{12} [Palkweng]	semi-dwarfness
Khao Jao Hawm Pitsanulok 1	Thailand, 1998 Rice Res Institute Chatuchak	cross cross with mutant of Leuang awn (LA29-73-NFU-14-3-1-1)	
Lafitte	USA, 1995 Lindscombe,Jodari,Bollich LSU - Rice Res. Stn. P.O. Box 1429 Crowley,LA 705271429	cross Mercury/Koshihikari	semi-dwarfness earliness
Oltenita	Romania, 1992 Alionte, G. ICCPT Research Institute Fundulea, Calarasi	gamma rays, 350 Gy [Krasnodar 424]	lodging resistance earliness yield
Pusa-NR-550-1-2 (JD-8)	India, 1997 IARI New Delhi	cross Dular mutant x N-22 mutant	semi-dwarfness yield
Pusa-NR-551-4-20 (JD-6)	India, 1997 IARI New Delhi	cross Dular mutant x N-22 mutant	semi-dwarfness yield

Pusa-NR-555-28 (JD-10)	India, 1997 IARI New Delhi	cross Dular mutant/N-22/PNR 351/Mutant 17	semi-dwarfness yield
Pusa-NR-555-5 (JD-3)	India, 1998 IARI New Delhi	cross Dular mutant/N-22/PNR 351/Mutant 17	yield
Pusa-NR-571	India, 1990 IARI New Delhi	cross MW 10 mutant x PMR 351 (Tainan 3 mutant/ Basmati 370)	semi-dwarfness earliness
S-102	USA, 1996 McKenzie, Johnson, California Coop.Rice Research Foundation Biggs, Ca 95917	cross Calpearl/Calmochi-101//Calpearl	earliness glutinous endosperm
VND95-26	Vietnam, 1995 Thinh, D.K., Nam, T.T.H. Inst. of Agric. Sci. S.Vi 121Nguyen Binh Khiem Dist.1 Hochiminh Cty	gamma rays, 200 Gy [IR 9729]	earliness grain quality yield
Pyrus pyriforia Nakai	***(japanese pear)***		
Gold Nijisseiki	Japan, 1993 Sanada, T. Fruit Tree Research Station MAFF, Tsukuba, Ibaraki 305	gamma rays chronic, 0.12-0.15 Gy [Nijisseiki]	disease resistance
Kotobuki Shinsui	Japan, 1996 Yoshioka, T. Institute of Radiation Breeding	gamma rays chronic, 80 Gy [Shinsu]	disease resistance

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Sesamum indicum L.	***(sesame)***		
Seodunkkae	Korea, 1997 Kang, C.W. RDA, 209 Seodun-Dong, Suwon 441-100	NaN_3, 2mM - 3 hrs. [Danbaeckkae]	disease resistance yield
Suwon 155	Korea, 1998 Kang, C.W. NCES, Suwon	gamma rays, 200 Gy	oil quality yield
Sorghum bicolor L.	***(sorghum)***		
Djeman	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 100 Gy [CSM 228]	grain colour grain yield
Djemanin	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 100 Gy [CSM 228]	grain colour grain yield
Fambe	Mali, 1992 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 300 Gy [CSM 388]	lodging resistance grain yield
Gnome	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 300 Gy [IPS 0001]	lodging resistance grain yield
Gnoumanin	Mali, 1998	gamma rays, 100 Gy	grain colour

	A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	[CSM 228]	grain yield
Sadje	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 300 Gy [IPS 0001]	earliness shortness
Sofin	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 250 Gy [CSM 388]	earliness lodging resistance
Tiedjan	Mali, 1998 A. Bretaudeau Institut Polytechnique Rural de Katibougou Koulikoro, BP 06	gamma rays, 100 Gy [CSM 228]	panicle size grain size
Syringa vulgaris L.	***(lilac)***		
Prairie Petite	USA, 1995 Lindgren, D.T. Univ. of Nebraska, WCREC/Horticulture Route 4/Box 46A	thN	dwarfness leaf morphology
Triticum aestivum L.	***(wheat)***		
Bakhtawar-92	Pakistan, 1994 NIFA	gamma rays	disease resistance yield
Darkhan-35	Mongolia, 1992	cross MS-77 (mutant from 'Sarruvra') x Mironovskaja jarovaya	protein content yield

Name of cultivar	Country and date of release (or approval) Name of principal worker(s) and institute	Mutagenic treatment [parent variety] or cross with mutant or with mutant derived variety	Main character improved
Darkhan-49	Mongolia, 1995	cross mutant of 'Orkhon'.	yield
Khara-86	Mongolia, 1986	gamma rays, 100 Gy [Orkhon]	earliness yield
Nishte-95	Pakistan, 1995 NIFA P.O.Box 446, Peshawar 25000		
Tatara	Pakistan, 1996 Syed, A.S. NIFA P.O.Box 446, Peshawar 25000	gamma rays, 1400 Gy	drought tolerance disease resistance semi-dwarfness
Xinong-Mai 2	China, 1993 Liu Zhongqi Crop research Institute Sichuan Ac. Agr. Sci Chengdu 610066	gamma rays, 200 Gy [77 Zhong 2882]	earliness disease resistance
Vigna radiata (L.) Wil.	***(mungbean)***		
NIAB Mung 92	Pakistan, Malik, I.A. NIAB P.O. Box 128, Faiselabad	cross NIAB Mung 36 x VC 2768B	disease resistance earliness
NIAB Mung 98	Pakistan, M. Siddiqui NIAB, P.O. Box 128, Faiselabad	cross NIAB Mung 20-21 (mutant) x VC 1482E	seed size yield disease resistance

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TO THE READER

This month, the fourth issue of the **PLANT BREEDING AND GENETICS NEWSLETTER** will also be published. The Newsletter will inform you about current activities of the FAO/IAEA sub-programme on plant breeding and genetics which is implemented by the Plant Breeding and Genetics Section of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture (Vienna) in close collaboration with the Plant Breeding Unit of the FAO/IAEA Agriculture and Biotechnology Laboratory (Seibersdorf). The Newsletter (PBGN) is published every 6 months so that you have more information about current and planned activities of the FAO/IAEA Plant Breeding and Genetics sub-Programme. It is expected that in the near future, it will also be available on the Joint Division's home pages on the Internet. I would like to emphasize, however, that this Newsletter **does not replace the MUTATION BREEDING NEWSLETTER**. In fact, the MBNL will continue to publish scientific papers related to the application of mutation techniques in plant breeding and genetics. Requests for inclusion on the mailing list of the PBGN should be sent to the address indicated on the back cover.

Miroslaw ***MALUSZYNSKI***

LAST BUT NOT LEAST

This Newsletter is distributed free of charge. To have your name added to our mailing list, please send your request to the address shown on the back cover. In addition to your full name, the request should indicate the detailed name of your institute, university or plant breeding station. Please note that if a copy is available in your library, a duplicate cannot be sent.

All published papers are reviewed. Please submit your contribution to the Mutation Breeding Newsletter by 1 June and 1 December of each year. Authors are kindly requested to take into account that readers want to learn about new findings and new methods but would also like to see the most relevant data on which statements and conclusions are based. Conclusions should be precise and distinguish facts from speculations. The length of contributions should not exceed 2-3 double-spaced typewritten pages including tables. We regret that for technical reasons photographs cannot be accepted. References to publications containing a more detailed description of methods for evaluation of findings are welcome but should generally be limited.

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