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REVIEW ARTICLE

A comprehensive review on mutation breeding milestones in cereals: Conventional to advanced molecular approaches to achieve sustainable goals in trait improvement

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Abstract

The grass family includes the annual common grasses known as cereals. The 5 main types of cereals are rice, wheat, barley, rye and oats. As a result of their high vitamin (20-80%) and fiber content (90%), these grains can help treat conditions like type II diabetes, complex metabolic syndrome, obesity, gastrointestinal disorders, and high cholesterol. Cereals have inspired a long history of research into their genetics, development, and evolution due to their significance in both economic and scientific terms. They need to be improved because they play a big part in global food production and crop quality is declining. The crop improvement can be done by incorporating desired features from non-adapted landraces/wild resources. That induced mutagenesis has made a significant contribution to crop improvement initiatives by producing mutant varieties with better and desired genetic modifications in the crop plants mainly in agronomically significant features. The mutants produced by the induction of physical and chemical mutagens were highly useful. The notable traits obtained by the induced mutations are semidwarfness and early maturity in rice varieties, disease resistance in barley, salinity, and drought tolerance in wheat, and lodging and disease resistance in oat and rye. In this review, we focused on identifying and developing elite mutants, their genetic function, and their utilization in future breeding programs for various traits in cereal crops.

Keywords

Cereals; Crop improvement; Functional validation; Genomics; Mutation

Introduction

The word "cereal" is derived from the Latin word "cerealis," which means "grain" and refers to a specific kind of fruit called a caryopsis, which is made up of the endosperm, germ and bran (1). The world's population relies heavily on cereals as a source of energy, protein, B vitamins and minerals. They are the most significant food sources (2). Cereal grains were the earliest agricultural initiatives by early man and people still consume them now based on their location as well as what grows there best. Cereal grains are grown in huge amounts and produce more food energy globally than any other crop; therefore, they are staple food crops (1). The consumption of the major cereals is used as a treatment for various human diseases (3). Moreover, the prediction for global grain consumption has climbed by 2735 million tons in 2020, compared to 43 million tons (1.6%) in 2019–20 (4). The profound importance of cereals in economic and scientific realms has sparked extensive research on their genetics, development, and evolution, leading to a rich historical exploration. The almost full rice genome sequence serves as a symbol of the shift to high-throughput genomics and computational biology, which has influenced research on many kinds of cereals (5).

Many genetic variations have been created since Muller and Stadler's groundbreaking work eighty years ago. These variations have been made possible by using a wide range of physical and chemical agents in mutation procedures. These procedures are now essential for plant breeding and genetic studies (6). Mutations are heritable changes in the phenotype of an organism. Chemical changes at the gene level are what cause these changes (7). Mutagens such as fast neutrons, ion beam radiation, X-rays, Gamma rays, and Ethyl methane sulphonate (EMS) are frequently used in mutation breeding. The physical mutation tends to result in double-strand DNA breaks and significant DNA deletions and consequently produces visible impacts on the chromosomal aberrations. Gamma rays are the most popular physical mutagen among plant breeders because they are convenient and may penetrate deeper into the tissues (8). Chemical mutagens like ethyl methane sulphonate and N-methyl-N-nitrosourea are widely used (9). It has most frequently been used to predominantly induce single-point mutation employed in investigations on reverse genetics. Mutation breeding is focused on taking advantage of local cultivars (10, 11). The number of mutant varieties developed so far by physical and chemical mutagens is 2610 and 384 respectively (12). Induced mutations are utilized to produce mutant plant cultivars with altered plant traits, leading to a notable increase in production and improvement in quality. The generated mutants have helped plant breeders overcome many challenges and create new, useful forms, such as semi-dwarf varieties, early maturing, and disease-resistant varieties (13). For many agricultural plants, dwarfism is a favorable trait. The development of dwarf rice and wheat cultivars was a key component in the success of the Green Revolution. Dwarfism can minimize lodging and boost the harvest index in grain crops (14). Mutations in the genes governing the GA signaling pathway or its production frequently result in dwarfism. Over the past 50 years, mutation breeding has gained popularity and as of today, 3362 mutant plant types from 240 different plant species have been introduced in more than 75 nations among them 1602 major kinds of cereal, 501 major legumes, and 86 major oil seed mutant varieties are developed by mutation breeding/induced mutation (12). The review highlights the mutants discovered in the major cereals, their traits and genes the functional validation of mutants, and the application of the mutants in the breeding program to enhance the genetic gain of the cereal crop species (Fig. 1).

Rice:

Rice is the base of the global population's diet about 3.5 billion consume it around the world (15), hence the advancement of the rice sector, particularly in breeding, is essential (16). The limited genetic diversity of rice has also slowed the creation of cultivars resistant to biotic (37.4%) (17) and abiotic (50%-70%) (18) stresses, leading to yield loss (19). One of the finest methods for introducing variations among the mega types with good adaptation is to use induced mutations. To cause mutations in rice, physical agents such as fast neutron, gamma-ray, and ion beams as well as chemical mutagens like ethyl methane sulfonate (EMS), methyl nitrosourea (MNU), and sodium azide (SA) have been used (20). The major unfavourable traits of rice are tall, lodging, and susceptibility to various abiotic and biotic stresses. Semi-dwarfism is one of the most crucial features in many crops, including rice, and was made possible by the Green Revolution in 1966. The

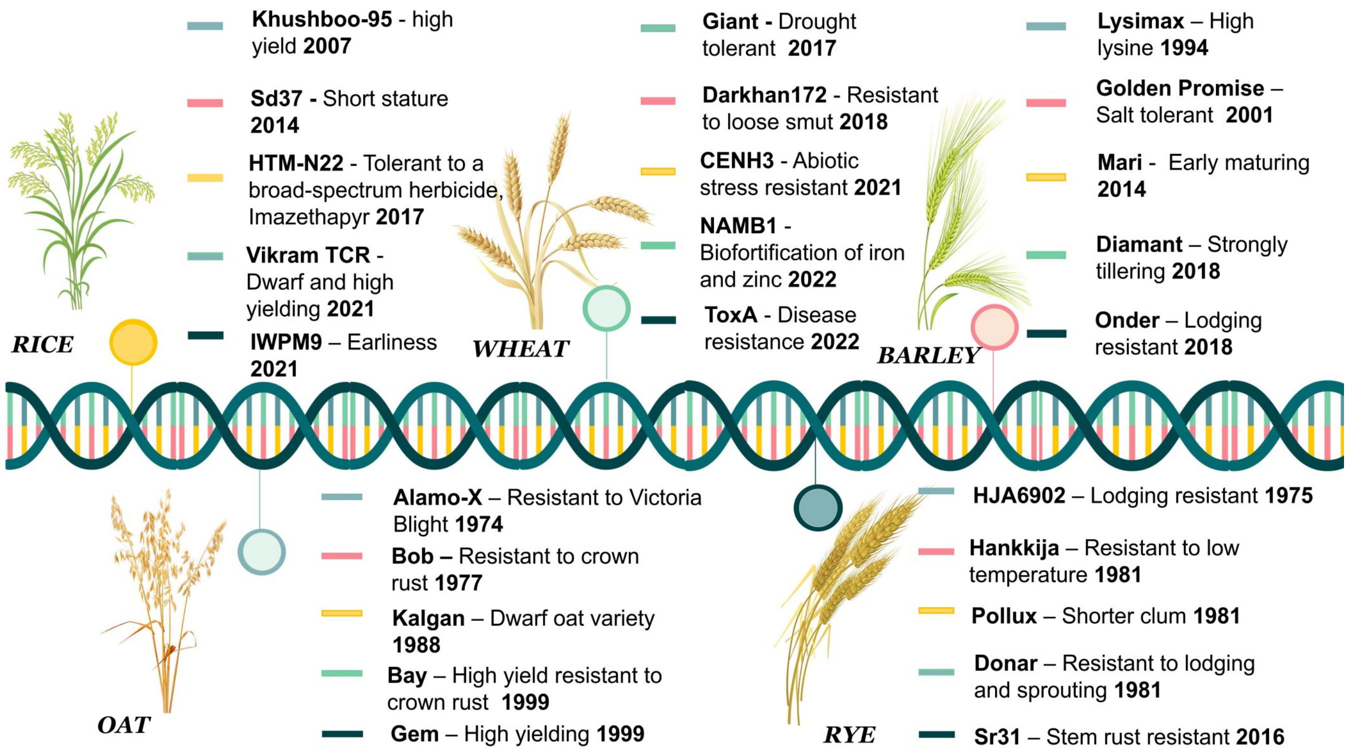

Fig. 1. Developmental milestones of desirable and beneficial mutants in cereal crops of rice, wheat, barley, oat, and rye.

"green revolution gene" is commonly referred to as the rice semi-dwarf 1 (*sd-1*) gene. *Sd-1*, which was produced from the Chinese cultivar Dee-Geo-Woo-Gen (DGWG), gives rice cultivars short, thick culms, increases the harvest index, enhances lodging resistance, and improves sensitivity to nitrogenous fertilizers, producing large yields without degrading panicle and grain quality (21). Mutations in the genes regulating the production or signaling pathway of the plant hormone GA frequently result in dwarfism (22). According to recent research, 85 genes and at least 76 plant-height mutants have been found in rice (23). In India, several rice cultivars from the "PNR" series were introduced. A few of these cultivars grow quickly and have short plant stature. Two early ripening, aromatic mutation-derived rice varieties namely, PNR-381 and PNR-102, are being preferred by farmers in the states of Haryana and Uttar Pradesh (24), these are the main contributions to crop improvement. The identified elite mutants for various characters in rice are listed (Table 1).

Barley

Barley is a fast-growing plant that can survive in a variety of environments. Ancient Egypt and the Roman Empire cultivated cereals as early as 15,000 BC (3). Barley grows well in cold climates and contains less gluten and high fiber. Additionally, the nutrient status of barley includes minerals like sodium, potassium, calcium, magnesium, iron, zinc, and selenium, proteins, and vitamin B. Cultivated barley (*Hordeum vulgare*), is primarily farmed for animal feed, particularly for pigs and for malting and making beer, and for distilling whisky. Barley is utilized as a minor portion of the diet. In the UK and the Far and Middle East, pearled barley is consumed in soups and stews. In some nations, barley is also mashed into porridge and used in baking as flour (25). The discovery of the first disease-resistant mutant against "barley powdery mildew" dates back to 1942, marking the beginning of mutation breeding in barley. This achievement was made possible through the application of X-ray-induced mutagenesis (6) and the first, chemically induced mutant 'Luther' in barley was released in 1966 in the USA which gave 20% higher yield and lodging resistance (10). Nilsson-Ehle crossed erectoides mutants with the Danish Kenia variety, superior malting barley, and obtained lodging-resistant and high-yielding recombinants (26). The major traits of barley improvement are semidwarf variety, lodging, good malting quality, and biotic and abiotic stress resistance (Table 2).

Wheat

Agronomically significant features have been successfully bred in a variety of crops using mutations. By using several mutagens, induced mutations in wheat have been discovered for both morphological and quantitative features (27). India's wheat production has grown rapidly to 96 million tonnes ever since the Green Revolution (28). However, the current breeding programs cannot increase production because they are not making the best use of genetic variety through recombination and selection. This circumstance demands the use of induced mutagenesis to produce fresh genetic variety. The major traits that should be improved in wheat are rust resistance, drought tolerance, short stature, early maturity, herbicide tolerance, and pre-harvest sprouting tolerance (Table 3). The most widely used genes in modern plant breeding are the semi-dwarf genes found in rice and wheat, which helped to spark a "green revolution" in the 1960s. The genes associated with the Green Revolution, namely *RhtB1b* and *Rht-D1b*, renowned for their capacity to reduce height, have been identified in over 95% of the wheat cultivars that have been introduced. In regions where the wheat crop is subject to terminal heat stress, early maturity is a desired characteristic. The flowering process in wheat is controlled by three gene sets: the photoperiod genes (Ppd), vernalization genes (Vrn), and earliness *per se* genes (eps), Early mutants were identified and confirmed in the M_3 generation of 2 cultivars, PBW677 and HD2967 (29).

Oat

A significant cereal grain grown all over the world is oat (*Avena sativa* L.). After maize, rice, wheat, barley, and sorghum, oat output is sixth in the world (30). The oat crop has previously been neglected in several ways and grown in places that are not ideal for growing wheat, barley, or maize. The popularity of oats has grown recently because oat grains are rich in unique galactolipids, unsaturated fatty acids, avenanthramides, and important dietary minerals including iron. Due to its grain's superior nutritional composition, which includes high-quality proteins, dietary fibers, carbohydrates, oil, minerals, and low starch content, oats are typically utilized as high-energy feed (31). Oats constitute a substantial source of high-quality proteins, with approximately 12% to 15% of cultivated oats being rich in protein. Oats with even greater protein levels would be a fantastic source of protein from the plant source. 230 mutant families with seed protein levels of 15% or greater, from a mutagenized oat population, were found in plants grown in greenhouses, 15 mutant lines with protein levels between 17 and 24% were chosen for future research (32). The identified elite mutants for various characteristics of oats are listed (Table 4).

Rye:

After wheat, rye (*Secale cereale* L.) is the second most employed grain for crafting bread. Rye can be grown in regions that are often unsuitable for other cereal crops because it is extremely winter-resistant and can survive in sandy soils with little fertility. Mostly, the rye planted worldwide is in cold temperate zones, but it may also be grown in semiarid areas close to deserts and at high altitudes (33). It is a significant crop in the Scandinavian nations, Germany, Poland, and Russia, where it is the main bread grain. Additionally, rye is utilized as animal feed and to make crispbread and alcohol. Genomics techniques were only recently created because Rye wasn't well known around the world and it was hard to genetically test a cross -pollinator with a genetic self-incompatibility mechanism (34). The identified elite mutants for various characters in Rye are listed (Table 5).

Table 1. Identified elite mutants for various characters in rice

Sl. No.	Parent	Mutant	Mutagen	Character	Gene	Author
1	Mahsuri	Mahsuri Mutant	EMS and Gamma Irradiation	Yield and disease resistance	-	(41)
2	IR8	CNM25	X rays	Long grain size	-	(42)
3	IR-8	CNM 31	X rays	Resistant to BLB, Early maturity	-	(42)
4	IR 8	CNM20	X rays	Early maturity, resistance to BLB and BPH	-	(43)
5	IR8	Au 1	Gamma rays	Early maturity	-	(43)
6	OC 1393	Biraj	X rays	Submergence tolerant	-	(44)
7	IR8	CNM 6 (Lakshmi)	X rays	Drought resistant	-	(44)
8	IR50	CRM 49	Sodium azide	Resistant to blast	-	(45)
9	Ptb 20	Dhanu	Gamma rays	Increased grain yield and tolerant to major pests and diseases	-	(46)
10	BPT-5204	Early samba	EMS	Stem blast tolerant	-	(46)
11	Oochikara	RH2	NAA (Auxin)	Root-hairless mutant	Single recessive gene	(47)
12	Basmati 370	CRM 2007 - 1	Gamma rays	Short duration, semi-dwarf	-	(48)
13	Muda	Muda 2	EMS and Gamma Irradiation	Yield and disease resistance	-	(49)
14	Manik	Manik817	EMS and Gamma Irradiation	Yield and disease resistance	-	(49)
15	IR6	Shadab	Ethyl methane sulphonate	High yield and fine grain quality	-	(50)
16	IR8	Shua-92	15 Gy fast neutrons	High yield and fine grain quality, salt tolerance	-	(50)
17	Jajai-77	Khushboo-95	200 Gy gamma rays	Short stature endowed with high-yield	-	(50)
18	IR8	Sarsha	150 Gy gamma rays	High-yielding variety with fine grain quality with tolerance to shattering and insect pests	-	(50)
19	Nipponbare	Narrow-leaf rolled mutant 1	-	Vigorously growing organs, such as roots, sheaths, and panicles	*nrl1*	(51)
20	Nipponbare	Narrow-leaf 9	T-DNA insertion	Reduced height and increased tillering	*nal9*	(52)
21	Japonica rice variety Kita-ake	Yellow Virescent	-	Chlorotic leaves	*vyl*	(53)
22	Indica cultivar 3037	sd37	-	Leaves, panicles, and seeds were smaller	*CYP96B4*	(54)
23	Nagina 22 HTM-N22	*N22*	EMS	Tolerance to a broad-spectrum herbicide, Imazethapyr	Single dominant gene (*AHAS*)	(55)
24	Jembar	Mustajab	Gamma Rays	Resistant to BLB and brown plant hopper	-	(56)
25	Improved White Ponni	Semidwarf early mutant	Gamma rays	Semi-dwarf, early maturing and high-yielding	*GA20Oxi_1 and GA20Oxi_2*	(57)
26	Jawaphool	CG Jawaphool Trombay	Gamma rays	Semi-dwarf, Mid early maturity	-	(58)
27	Improved White Ponni	IWPM9, IWPM20, IWPM1 and IWPM25	Gamma-irradiated	Earliness and semi-dwarfism	-	(21)
28	Simei no. 2	Zhefu 802	Gamma irradiation	Cold tolerance	-	(59)
29	Tarom Mahalli	Kian	Gamma rays	Drought tolerant	-	(60)
30	Safri 17	Vikram- TCR (Trombay Chhattisgarh Rice)	Gamma rays	Dwarf and high-yielding	-	(60)

Table 2. Identified elite mutants for various characters in Barley

Sl. No.	Parent	Mutant	Mutagen	Gene	Trait/ Character	Author
1	Domen Mari	Kristina	Ems	-	High malting quality, lodging resistance, and resistance to straw breakage	(26)
2	Bonus barley	Pallas	X-rays	-	High yielding	(61)
3	RDB-1	RD-103	Neutrons	-	Dwarfness and erect leaf morphology	(62)
4	RDB-1	K-257	Neutrons	-	High yield, long ears	(43)
5	RDB-1	RD-137	Neutrons	-	High yield, less water requirement	(63)
6	RDB-1	Karan-15	Neutrons	-	Semi-dwarfness and yield	(44)
7	RDB-1	Rejkiran (RD-387)	Neutrons	-	Nematode resistant	(44)
8	Diva and Celb4	1017 and C-67-7	-	-	Seed protein	(64)
9	Haisa	Mutante 66 (M66)	X rays	*mlo*	Resistance to barley powdery mildew	(65)
10	Vollkorn	H3502	X rays	*mlo*	Resistance to barley powdery mildew	(65)
11	Malteria heda	MC20	Gamma rays	*mlo*	Resistance to barley powdery mildew	(65)
12	Foma	SRI	X rays	*mlo*	Resistance to barley powdery mildew	(65)
13	Carlsberg II	R5678	EMS	*mlo*	Resistance to barley powdery mildew	(65)
14	Carlsberg II	R6018	EMS	*mlo*	Resistance to barley powdery mildew	(65)
15	Carlsberg II	R7085	EMS	*mlo*	Resistance to barley powdery mildew	(65)
16	Carlsberg II	R7372	EMS	*mlo*	Resistance to barley powdery mildew	(65)
17	Diamant	SZ5139b	EMS	*mlo*	Resistance to barley powdery mildew	(65)
18	Foma	SR7	Gamma rays	*mlo*	Resistance to barley powdery mildew	(65)
19	73 M1	Stimul	NEU N-Nitroso- N-methyl urea	-	Early maturity	(66)
20	Sultan	Lysimax	-	*lys3a*	High-lysine	(67)
21	Arabi Aswad	Furat 3	Gamma rays	-	Resistant to lodging and drought	(46)
22	Diamant	Roxana	X-ray	-	Erectoid type	(46)
23	Maythorpe	Golden Promise	gamma-ray	*GPert*	Malting quality Salt tolerant	(68)
24	-	Dobrynia-3	NEU N-Nitroso- N-methyl urea	-	Tolerant to low temperatures	(66)
25	Diamant	Felicitas	Gamma rays	-	Erectoid type	(69)
26	-	Mari	X-ray	*Mat-a*	Early maturing and lodging resistance	(70)
27	-	semidwarf1	-	*sdw1*	Lodging	(71)
28	Sahin 91	Onder	Gamma rays	-	Lodging resistant	(72)
29	Valticky	Diamant	X-rays	*sdw*	Strongly tillering variety, with very good grain and malting quality, lodging resistance	(10)
30	Bulgarian winter feed barley variety	IZ Bori	sodium azide		Tolerant to low temperatures with very good resistance to powdery mildew, as well as to brown, black, and stem rust, with high grain yield (15–17%)	(10)

Table 3. Identified elite mutants for various characters in Wheat

Sl. No.	Parent	Mutant	Gene	Mutagen	Trait/ Character	Author
1	NP 799	NP 836	-	X rays	Higher yield	(73)
2	Sonora 64	Sharbati sonora	-	Gamma rays	Early maturity, amber grain color, high protein content	(74)
3	Lerma Rojo 64-A	Pusa Lerma	-	Gamma rays	Amber grain color, higher extensibility and elasticity of flour	(75)
4	New Thatch	NI -5643	-	Hybridization with two irradiated mutants	Early maturity, resistance to rust	(76)
5	*Triticum aestivum* L.	ND496-25	single recessive gene	Several mutagen-induced or spontaneous chlorina and virescent mutant	Inheritance of Three Chlorophyll-Deficient Mutants	(77)
6	Anza	Spinnakr	-	Fast neutrons	Resistance to lodging	(78)
7	*Triticum aestivum* L.	Chl b-less	-	-	Analyses of a series of allelic chlorina mutants of wheat that have partial blocks in chlorophyll synthesis	(79)
8	Wheat cv.	Ppd1	Individual genes	Sodium azide and EMS	Isolation and analysis of thermotolerant mutants	(80)
9	Durum wheat	Four independent mutants (139, 142, 196, 504)	-	-	An extended period of flag leaf photosynthetic competence is associated with the production of larger grains	(81)
10	*Triticum aestivum* L.	lpa1-type	single-gene	Ethyl methane sulfonate	Identification and Characterization of a Low Phytic Acid	(82)
11	Wheat	Ug99	*Sr31*	-	Durable resistance to wheat stem rust	(83)
12	Winter 6/92R149	Jingdong 23	-	Gamma rays	High yield	(84)
13	*Triticum* species	-	Several mutant genes	gamma radiation, high and low energy beams, ethyl methane sulfonate, EMS	enhancing nutrition and food production	(85)
14	WL-2265	Ganesha	-	Gamma rays	High yield and resistance to leaf rust	(86)
15	RAH -506	Darkhan -141	-	Gamma rays	Early maturity and high protein	(87)
16	Pasa	NARO Wheat 1	-	Gamma rays	Resistance to stem rust	(87)
17	Pasa	NARO Wheat 2	-	Gamma rays	Resistance to stem rust	(87)
18	Pasa	NARO Wheat 3	-	Gamma rays	Resistance to stem rust	(87)
19	Wheat	Ug99	Single or preferably multiple *R*-genes	NB-LRR	Recognition of the most essential chemical components of the plant pathogens should provide the greatest durability of resistance	(88)
20	L-880	Bingom-1	-	-	Salinity tolerant	(89)
21	Kalinova	Giant	-	Gamma rays	Drought tolerance	(90)
22	Favorika	Leana	-	Nitrosomethyl urea	earliness	(90)
23	Sonechko	Leroy		Gamma rays	Drought tolerant	(90)
24	Darkhan 95	Darkhan 172		Sodium azide	Resistant to loose smut	(56)
25	*Triticum aestivum* L.	XY22	Multigene pyramiding	Sodium dodecyl sulfate-polyacrylamide gel electrophoresis	High yield, high disease resistance, and high grain quality by the MAS method	(91)
26	Wheat	omt-A2 omt-B2	-	-	Analysis of the plant growth and metabolite	(92)
27	*Triticum aestivum* L.	CENH3	*TaGS5*, *TaGS3* and *TaCwi-A1*, *Psy1* (Phytoene synthase) gene and *Zds1*, *Cre3*, *Cre1*, *Cre* (Resistance)	Physical and chemical mutagens	Abiotic stresses at different growth stages	(93)
28	*P. tritici-repentis* and *S. nodorum*	ToxA-insensitive/ disease-susceptible mutant	dominant sensitivity genes-*Tsn1*	-	Plant disease resistance	(94)
29	*Triticum* species	NAM-B1/ GPC1 l	*TaVIT2*	-	Biofortification of iron and zinc	(95)

Table 4. Identified elite mutants for various characters in Oat

Sl. No.	Parent	Mutant	Mutagen	Gene	Trait/Character	Author
1	Ryhti	Nasta	X-rays	-	Crossing with one mutant variety	(96)
2	Florad	Florida 500	Thermal neutrons	-	Improved agronomic type and resistance to crown rust	(97)
3	Alamo	Alamo-X	X-rays		Resistance to Victoria blight and Crown rust	(97)
4	Floriland	Florad	Thermal neutrons		Resistance to stem rust superior grain quality and straw stiffness	(97)
5	Florad	Florida 501	Thermal neutrons		Improved agronomic type and resistance to crown rust	(97)
6	Jo50-2395	Ryhti	X-rays		High yield and stiff straw	(97)
7	Florad	Bates	Thermal neutrons		Later maturity, shorter culm, Resistance to lodging higher yield resistance	(98)
8	Kransnodarskii 73	Zelenii			Large leaves, late maturity resistance to spring frosts	(99)
9	Florad	Bob	Thermal neutrons		High yield, resistance to crown rust, shorter clum	(98)
10	Ryhti	Puhti	X-rays		High yield and stiff straw	(62)
11	Orel	Belozernii	N-nitroso-N-methyl urea (NMU)		Resistance to lodging	(42)
12	Jo502395	Veli	-		High yield	(63)
13	OT207	Dolphin	-		Short culm length and high yield	(100)
14	OT207	Echindna	-		Short culm length and high yield	(100)
15	OT207	Kalgan	-		Character mid-season maturing dwarf oat with excellent straw strength	(101)
16	Selma	SIR-4	-		Early maturity and high adaptability	(101)
17	Florida 501	Ozark	Thermal neutrons		Resistance to low temperature and high yield potential	(102)
18	Irradiated monosomic substitution line	Centennial	Thermal neutrons		Resistance to crown rust	(103)
19	Irradiated monosomic substitution line	Horicon	Thermal neutrons		Resistance to crown rust	(103)
20	Irr4	Bay	Thermal neutrons		Resistance to crown rust and high-yield potential	(103)
21	Irr4	Belle	Thermal neutrons		Resistance to crown rust and high-yield potential	(103)
22	Irr4	Gem	Thermal neutrons		Resistance to crown rust and high-yield potential	(103)
23	O7207	ACRonald	Fast neutrons	*DW-6*	High yielding, white-hulled, and dwarf	(104)

Table 5. Identified elite mutants for various characters in Rye

Sl. No.	Parent	Mutant	Mutagen	Gene	Trait/Character	Author
1	Vjatka	HJA 6902	Gamma rays	-	Resistance to lodging	(76)
2	Vjatka	Hankkija's jussi	Gamma rays		Resistance to low temperature short stiff straw and good quality	(63)
3	Petkuser winterroggen stamm 267/70	Pollux	-	*PMS 2*	Shorter culm and resistance to lodging	(63)
4	Petkuser winterroggen stamm 267/70	Donar	ISO-PMS		Shorter culm, resistance to lodging, and sprouting	(63)
5	Secale cereale	Sr31 and Sr50	CC-NB-LRR		stem rust resistance	(105)

Integrative approaches in Mutation Breeding:

Mutation breeding involves 3 forms of mutagenesis. These are induced mutagenesis, in which mutations occur as a result of irradiation (gamma rays, X-rays, ion beam, etc.) or treatment with chemical mutagens; site-directed mutagenesis, which is the process of creating a mutation at a defined site in a DNA molecule and insertion mutagenesis, which is due to DNA insertions, either through genetic transformation and insertion of T-DNA or activation of transposable elements (35). Mutagenic plants or mutagenic seeds are developed and can be utilized directly as a commercial cultivar or as a parent to breed new commercial cultivars (36). Advanced mutation breeding methods and their outcomes are explained in (Fig 2). The M_2 and M_3 mutant populations can be used to map the altered genes using MutMap, MutMap$^+$, and MutMap Gap. The genes identified can be utilized in functional validation and characterization. The elite mutant identified and developed has been used for multi-omics research to study in-depth of the mutant.

Mutagenesis and Functional Validation:

Recently, the use of mutation breeding as a productive strategy for agricultural improvement has gained in popularity (6). The key outcomes of mutation breeding were quality traits like dwarfism, high yielding, early maturity, and tolerance to various biotic and abiotic stress; the resulting mutants are widely used for various purposes, such as improving both oligogenic and polygenic characters as well as morphological and physiological characters. They are employed to examine the genetic structure and provide functional validation. The direct use of mutations in the building of genetic maps in structural and functional genomics (Fig. 2) could quickly boost plant production and quality. The molecular methods for DNA fingerprinting and molecular mapping, including RAPD (Random Amplified Polymorphic DNA), AFLP (Amplified Fragment Length Polymorphisms), and STMS (Sequence-Tagged Microsatellite Sites), have made major contributions to the analysis and screening of mutants (6). Several rice genes have been functionally validated with the help of transgenics and EMS mutants (37). AKS-sd1, a derived cleaved amplified polymorphic sequence (dCAPS) marker, was created to validate the semi-dwarfing allele, sd1-bm (Bindli Mutant 34), This allele offers an option to the most used sd1-d in programs to improve rice (23). Two PCR markers were created to test for the presence of semi-dwarfing alleles in wheat the Rht-B1c and Rht-B1e. For Rht-B1c, a 256-bp product was amplified by PCR using the primer pair Rht-B1c-F1/Rht-B1c-R1 exclusively from lines bearing the Rht-B1c allele. For Rht-B1e, 2 primer sets were used: PCR with forward primer BF and reverse primer MR3 amplified a 228-bp fragment only in lines containing the Rht-B1e allele, while PCR with forward primer BF and reverse primer WR3 amplified wild-type sequences (38).

Application of Identified Mutant in Crop Improvement

Due to the limited amount of arable land present, the diminishing supply of water, and the fluctuating climatic conditions, the future of food production is gloomy. A decrease in arable land as a result of urbanization, salinization, biotic stress, drought, and desertification added to the issues. There are various methods for utilizing the heritable variations that are genetically encoded in existing crop plants for crop improvement programs (39). Such genetic alterations may take place experimentally under the influence of physical and chemical mutagens or may take place naturally at a very slow rate. Conventional mutation techniques have been employed frequently to enhance agricultural output, quality, disease, and insect resistance (Fig. 3) as well as the aesthetic appeal of flowers and ornamental plants. In most cases, mutation is the primary source of genetic variability, the raw material for natural selection-based evolution (40).

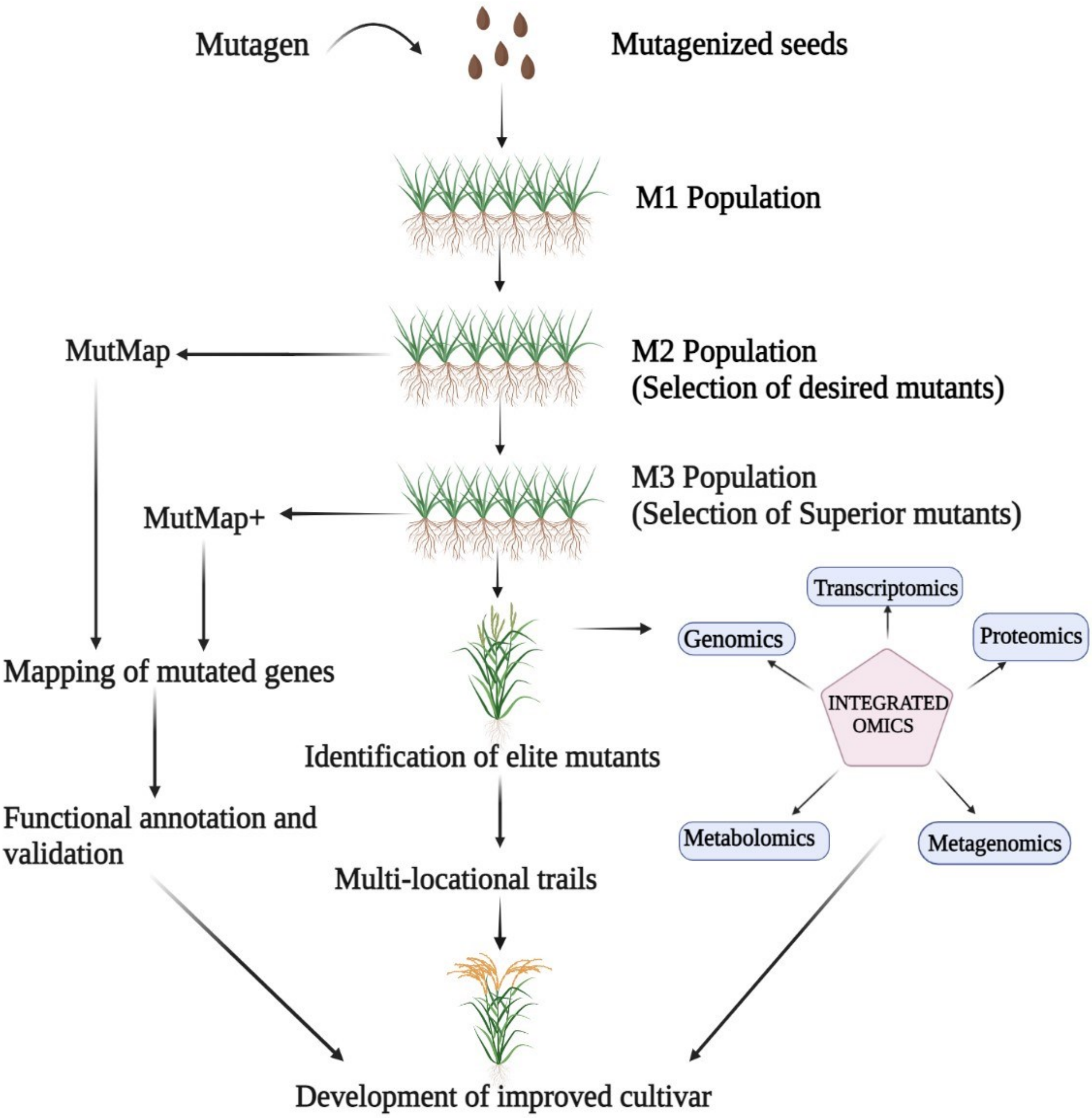

Fig. 2. Integrative approaches to mutation breeding and advanced multi-omic strategies to develop elite mutants with enhanced genetic gain.

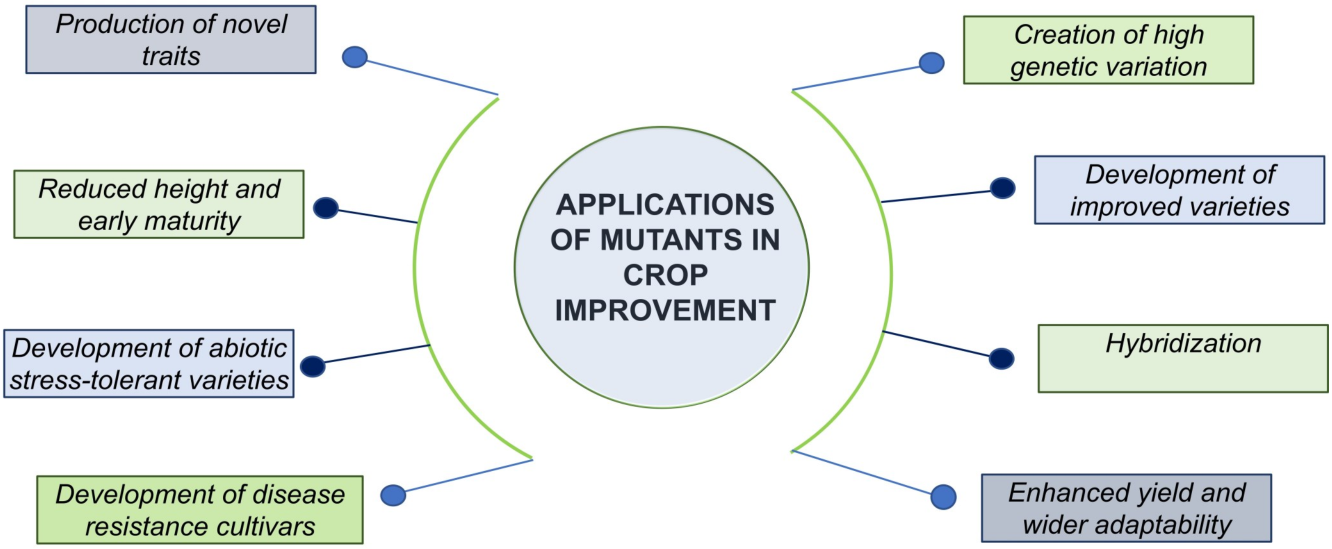

Fig. 3. Generalized applications of developed mutants in the breeding program and crop improvement.

Conclusion

Over the last few years, mutation breeding has gained popularity and has been adopted by many countries to improve characters. It enhances various qualitative and quantitative crop plant characteristics and has been successfully employed in a variety of cereals, grain legumes, oil seeds, vegetables, fruits, medicinal plants, ornamental plants, and fodder crops. In this review, mutants identified in cereals were emphasized. These elite mutant varieties produced by mutation breeding can be used in hybridization programs for the introgression of superior genes in the different elite backgrounds using marker-assisted selection in the crop species. This will enhance the genetic gain by designing superior crop varieties. The next step in mutation breeding is multi-omics studies, where the best mutants will be used for integrated omics research (genomics, transcriptomics, proteomics, metabolomics, and metagenomics) and functional characterization, as well as for creating superior crop varieties and enhancing crop quality.

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Authors' contributions

AJ, NED, and AJ have equally contributed to write an article. AJ, NED, and AJ searched, collected, wrote, and edited the manuscript. RS conceived the idea of writing the review and designed the content. IP, LP, DN, and NF analyzed the manuscript and provided regular assistance to revise and finalize it. RS and AP critically reviewed and edited the same. All authors have read and approved to publish the manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None.

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