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Buckwheat Gene Pool: Potentialities and Drawbacks for Use in Crop Improvement Programmes

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ABSTRACT

Common buckwheat (*Fagopyrum esculentum* Moench), a diploid (2n = 16) annual crop plant, is an important crop in mountainous regions of India, China, Russia, Ukraine, Kazakhstan, parts of Eastern Europe, Canada, Japan, Korea and Nepal. 13S globulin, the main seed storage protein of common buckwheat, has a nutritionally balanced amino acid composition, providing an excellent supplement for cereal-grain diets. The plant is also a rich source of rutin, vitamins B_1, B_2, E and dietary proteins for gluten–sensitive individuals. The presence of protease inhibitors and antifungal proteins in grains of common buckwheat makes it an important genetic base for isolation of genes for such proteins for use in biotechnology industry. The genus *Fagopyrum* (Polygonaceae) consists of 19 species that are divided into two monophyletic groups *viz*. the *cymosum* group which includes *F. esculentum*, *F. tataricum*, and *F. homotropicum* and the *urophyllum* group which includes wild species that have small lustrous achenes completely covered with a persistent perianth. Even though buckwheat cultivation has been going on for a long time, the crop still has a long way to go in the direction of greater popularity and importance. Some of the inherent limitations in buckwheat like low and unstable yields, indeterminate growth habit, seed-shattering, lodging, presence of allergenic compounds, bitter taste and low shelf life of its flour restrict its popularity. Advancing the utilization of buckwheat would require an integrated approach involving MAS for genotypes showing quality traits, mutagenic approach aimed at elimination of immune-dominant allergenic proteins screening of the entire gene pool of buckwheat, including its wild species, for genes which could be used for improvement of the cultivated species through biotechnological approaches.

Keywords: allerginicity, antioxidants, buckwheat, *Fagopyrum* spp., functional foods, nutraceutical, nutritional enhancement, rutin, seed storage proteins

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INTRODUCTION

Plant genetic resources, representing the sum total of diversity accumulated through years of cultivation under domestication and natural selection, are considered as one of the most important gifts of nature to mankind. Despite a large diversity of such resources, mankind has depended on only a very limited number of crops to meet its domestic needs. Currently only 30 plant species are used to meet 95% of the world's food energy needs (FAO 1996). These crops are widely and intensively cultivated and have been selected from a large agrobiodiversity basket containing more than 7,000 food species (Wilson 1992), which is approximately 1/10 of the estimated number of edible species present in nature (Myers 1983). While the extensive breeding programmes initiated on conventional crops have fuelled their promotion and ensured their success across continents, the presence of a limited amount of germplasm of many other crops, which have not been subjected to domestication through selection processes, in gene banks and its poor representation in terms of genetic diversity (Padulosi 1999) represents a great challenge for the successful improvement and promotion of this group of species. Amongst the existing known plant resources, the International Plant Genetic Resources Institute (IPGRI) and Consultative Group on International Agriculture (CGIAR) have identified buckwheat (*Fagopyrum* spp.), grain amaranth (*Amaranthus* spp.) and *Chenopodium* spp. as important but underutilized nutraceutical crops which have tremendous potential for use in crop improvement programmes.

Even though buckwheat is considered to be a minor crop, it is an indispensable food in the temperate and hill regions of East Asia and Europe. Buckwheat is a multipurpose crop used for food, feed, medicine and manure (Li and Zhang 2001; Zeller 2001). Common buckwheat (*Fagopyrum esculentum* Moench), a diploid (2n = 16) annual plant, is widely cultivated in Asia, Europe, and America. Due to short growth span, capability to grow at high altitudes and the high quality protein content of its grains it is an important crop in mountainous regions of India, China, Russia, Ukraine, Kazakhstan, parts of Eastern Europe, Canada, Japan, Korea and Nepal. The plant is a rich source of Zn, Cu, Mn, Se, vitamin B_1, B_2, E and dietary proteins for gluten-sensitive individuals (Wei *et al*. 2003; Stibilj *et*

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al. 2004). Buckwheat leaves and flowers are a rich source of rutin, catechins and other polyphenols that are potential antioxidants (Luthar 1992; Oomah and Mazza 1996; Watanabe 1998). Buckwheat proteins have also been reported to have anticancer, hypoglycaemic and antihypertention properties (Kayashita *et al*. 1999; Kayashita *et al*. 1995a, 1995c, 1997, 1996b; Tomotake *et al*. 2000; Park and Obha 2004; Tomotake *et al*. 2006). As a green manure crop, buckwheat produces only modest biomass but offers rapid growth, improves soil and makes phosphorous more available. Quick, aggressive growth accounts for its success as a smother crop for suppressing weeds, particularly in late summer.

TAXONOMY AND PHYLOGENY

Taxonomic investigations on the genus *Fagopyrum* have made progress mainly through morphological studies. The wild species of buckwheat were first classified by Gross in 1913. However, many species which were included by Gross (1913) within the genus *Fagopyrum* are now classified in other genera in the family Polygonaceae. On the basis of embryo morphology and achene characters, Nakai (1926) insisted that *Fagopyrum* should be considered separately from other genera in the family Polygoneae. Furthermore, cytological studies revealed that all *Fagopyrum* species have the basic chromosome number 8 while the basic number is 10 for *Polygonum sensu stricto*, 11 for *Persicaria* and *Bistorta* (Munshi and Javeid 1986). While Gross (1913) has suggested that *Fagopyrum* was closely related to *Fallopia*, Ronse Decraene and Akeroyd (1988) were of the opinion that *Fagopyrum* was more closely related to *Persicaria*. Most studies have concluded that *Fagopyrum* lies at the basal position of the tribe Polygonaceae. The genus *Fagopyrum* consists of about 19 species of which only two species, namely, *F. esculentum* and *F. tartaricum* are cultivated. On the basis of morphological characters, the genus *Fagopyrum* has been divided into two phylogenetic groups *viz.* the *cymosum* group and the *urophyllum* group. The *cymosum* group comprises of the two cultivated species *F. esculentum* and *F. tartaricum* and other wild species like *F. cymosum*, *F. homotropicum*, *F. lineare*, *F. pilus*, etc. Plants belonging to this group have large lusterless achenes which are incompletely covered with a persistent perianth. The *urophyllum* group, on the other hand, comprises of *F. urophyllum* and other wild species which have small lustrous achenes which are completely covered with a persistent perianth (Ohnishi and Matsuoka 1996; Yasui and Ohnishi 1998a; Ohsako and Ohnishi 2000). The genus *Fagopyrum* is comprised of both perennial as well as annual species with diploid ($2n = 2x = 16$) as well as tetraploid ($2n = 4x = 32$) cytotypes. While the genus *Fagopyrum* is comprised of both self- as well as cross-pollinated species, the occurrence of dimorphic heterostyly renders some of the species self incompatible.

According to Ohnishi (1995), there are two hot spots *viz*. (i) northwest corner of Anion province of China and (ii) upper Min river valley of China, from where different species of the genus *Fagopyrum* have probably evolved. These regions are home to all the new species of *Fagopyrum* discovered by Ohnishi. Further, *F. cymosum*, the wild species of the *cymosum* group has been reported to occur in both the areas. This could imply that speciation in *Fagopyrum* might have taken place in these two areas independently (Ohnishi 1995). Ohnishi (1995) has postulated that a shift from a self-incompatible outbreeder to a self-compatible self-pollinator occurred independently in several branches of the phylogenetic tree. A shift from an outbreeding species to a selfing one also took place in *F. gracilipes*, where we still can find populations having both homostylous self-pollinating plants and heterostylous outbreeding plants. In the case of *F. gracilipes*, self-fertilization probably has arisen after duplication of the chromosome number (Ohnishi 1995). It would also appear that the species *F. tataricum*, a homostylous self breeder, originated from an ancestral heterostylous outbreeder. However, no such heterostylous outbreeder has been identified till date. With the exception of *F. gracilipes* and *F. cymosum* most of the wild species of *Fagopyrum*, including the newly discovered ones, have a narrow endemic distribution in southern China. While the tetraploid *F. gracilipes*, a mainly self-fertilizing weedy species, covers almost the whole of China, except Tibet the diploid *F. cymosum* is found in southern China only. On the other hand, the tetraploid *F. cymosum* has been shown to have its distribution westward to Nepal and North West Himalayas (Ohnishi 1993a).

Isozyme profiles, inter-specific crossability and *rbc*L gene sequence analysis in buckwheat have indicated a closer relationship within members of either *cymosum* or *urophyllum* groups than between members belonging to the two subgroups (Wang 1987; Ohnishi and Matsuoka 1996; Chen 1999a, 1999b). On the basis of morphology, *F. cymosum* appears to be more closely related to *F. esculentum* than to *F. tartaricum* (Steward 1930). However, based on RFLP analysis of cpDNA, Kishima *et al*. (1995) suggested that *F. cymosum* was more closely related to *F. tartaricum* than to *F. esculentum*. This view has been supported by Ohnishi and Matsuoka (1996), Yasui and Ohnishi (1998a, 1998b) and Rout and Chrungoo (2007). Until the discovery of *F. esculentum* spp. *ancestrale* and *F. tartaricum* spp. *potanini*, *F. cymosum* was considered to be the wild ancestor of *F. esculentum* and *F. tartaricum* (Ohnishi 1991, Ohnishi 1998a). Yasui and Ohnishi (1998a, 1998b) have postulated that *F. esculentum* spp. *ancestrale* differentiated from *F. cymosum* much earlier than *F. tartaricum* spp. *potanini*. Further, *F. homotropicum* is closely related to *F. esculentum* ssp. *ancestrale* and has probably evolved from *F. esculenttum* ssp. *ancestrale* following shifting of the breeding system from out-crossing to self-fertilization (Ohnishi and Matsuoka 1996; Yasui and Ohnishi 1998b).

BUCKWHEAT AS A NUTRACEUTICAL CROP

Buckwheat is a rich source of nutraceutical compounds including vitamins, antioxidants, proteins, dietary fibre, rutin and trace elements (Li and Zhang 2001; Fabjan *et al.* 2003). Buckwheat flour is one of the best sources of high-quality easily digestible proteins in the plant kingdom. Its 74% protein absorption rate makes it an excellent meat substitute as well as an excellent supplement for cereal-grain diets. The protein content of buckwheat grains has been reported to range from 12 to 18.9% (Steadman *et al.* 2001; Li and Zhang 2001; Wei *et al*. 2003; Krkošková and Mrázová 2005; Stempińska and Soral-Śmietana 2006). The main storage protein of buckwheat grains is 13S globulin (Aubrecht and Biacs 1999; Li and Zhang 2001) having hexameric structure typical of legumin type proteins (Aubrecht and Biacs 1999). The amino acid composition of buckwheat 13S globulin closely matches the FAO recommended values for a protein with nutritionally balanced amino acid composition (Rout *et al.* 1997; Radovic *et al.* 1999; Rout and Chrungoo 1999; Kato 2001). Radović *et al.* (1999) have also reported the presence of 8S vicilin-like proteins in grains of common buckwheat. The very low content of prolamins in the grains makes buckwheat flour a valuable ingredient in diets or food products for coeliac patients (Li and Zhang 2001). To develop dietary protein for utilization as an ingredient in the food industry, it is necessary to determine its physicochemical and functional properties. Investigations on buckwheat protein (BWP) carried out by Tomotoke *et al.* (2002) have revealed that the physicochemical properties of BWP were different from those of Soy protein isolate (SPI) or casein. Although the water holding capacity of BWP was lower than SPI it had a higher fat absorption capacity. They have suggested that BWP has a strong potential for application in food industry.

The specific characteristics of its flour make buckwheat the unsurpassed cholesterol-lowering food studied to date. Buckwheat flour is also known to reduce and stabilize blood sugar levels following meals – a key factor in preventing diabetes and obesity. The hypoglycemic effect of buckwheat flour has been attributed to the presence of the rare carbohydrate compounds called “Fagopyritols” (especially D-*chiro*-inositol), of which buckwheat is by far the richest food source yet discovered. Fagomin, a D-*chiro*-inositol from buckwheat has been shown to lower plasma glucose, improve glucose tolerance, and enhance insulin immunoreactivity in KK-A(y) mice (Bnouham *et al.* 2006). It has been suggested that fagomin-induced potentiation of insulin release may be a factor contributing towards the hypoglycaemic action of buckwheat extracts. Like the widely prescribed “ACE” hypertension drugs, rutin, the flavonoid glycoside present in buckwheat grains, inhibits the activity of angiotensin converting enzyme (ACE), thereby reducing hypertension (Huff and Carroll 1980; Sugiyama *et al.* 1985). The rutin content in buckwheat grains has been reported to range from 3-6 mg 100 mg^{-1} dry weight with *F. tataricum* having 2-4 times higher rutin content than *F. esculentum* (Kitabayashi *et al.* 1995; Oomah and Mazza 1996; Li and Zhang 2001; Chao *et al.* 2002). Not much information is, however, available about the levels of rutin in other species of buckwheat that have now been identified. As some of these species are now being crossed with common buckwheat and the remainder may be utilized as sources of specific traits, this information could be of high practical value. Kitabayashi *et al.* (1995) have evaluated the varietal differences and heritability of rutin content in the seed and leaf of 27 cultivars of common buckwheat. While the rutin content of grains in tetraploid cultivars from Japan ranged from 2.0-2.2 mg 100 mg^{-1} dry weight it ranged from 1.4 to 1.8 mg 100 mg^{-1} dry weight in the diploid cultivars. However, the leaf rutin content did not show any variation with ploidy level of the cultivars. The heritability coefficient for rutin content in the seed and the leaf has been estimated at 0.59 and 0.25, respectively. These results indicate that rutin content in the grains has relatively high heritability amongst different cultivars of buckwheat. Yan *et al.* (2004) also observed a significant difference on the variations of the rutin content among different genotypes as well as among different ecological environments. Buckwheat extracts have also been reported to lower blood cholesterol level, particularly that of LDL (low-density lipoprotein) and VLDL (very low-density lipoprotein) (Kayashita *et al.* 1995a; Misawo and Iwao 1996; Tomotake *et al.* 2002, 2006) and also suppress carcinomas mammary glands (Kayashita *et al.* 1999). While the hypocholesterolemic effect of buckwheat flour in humans has been linked to the presence of fibre-like substances in the flour (Kayashita *et al.* 1997; Tomotake *et al.* 2001), suppression of mammary carcinogenesis has been associated with the property of buckwheat to lower serum estradiol (Kayashita *et al.* 1999). Guo *et al.* (2007) have isolated a 31 kDa protein “TBWSP31”from grains of tartary buckwheat which shows potent antitumour activity in human breast cancer cell line Bcap37. Further, a 4 kDa antifungal peptide isolated from buckwheat endosperm tissues has been reported to inhibit proliferation of malignant hepatic cell line HepG2 and breast cancer cell line MCF7 (Leung 2007).

Buckwheat leaves and flowers are also a rich source of quercetin, catechins and other polyphenols that are potential antioxidants (Luthar 1992; Oomah and Mazza 1996; Watanabe 1998). Oomah *et al.* (1996) have suggested that the content of flavones and antioxidants in tissues of buckwheat depended on the geographical location could be an important factor leading to the differences in rutin and flavanoid content in buckwheat. Buckwheat grains also are a rich source of phenolic acids, including hydrobenzoic acids, syringic, *p*-hydroxy-benzoic, vanillic, and *p*-coumaric acids (Przybylski *et al.* 1998). Tartary buckwheat is known to have an advantage over cereals due to its high flavonoid content, particularly rutin, which is noted for its therapeutic effect for treatment of blood-vessel fragility with accompanied hypertension (Nikitchuk 2000). Buckwheat flowers are also a good source of honey. Honey obtained from buckwheat flowers has been shown to increase the antioxidative potential of human blood serum and protect the lipoproteins of blood serum against oxidative damage more effectively than saccharin analogues (Gheldof *et al.* 2003).

BUCKWHEAT AS A SOURCE OF NOVEL GENES

Buckwheat is not only a promising crop in itself but also offers a wide range of desirable qualities for the improvement of other crops as well as holds possibility for making important contribution to the biotechnological industry. The crop calls for further research to maximize its potentials and popularity so that the potentialities of this crop may be tapped to the maximum. The presence of allelopathic chemicals, protease inhibitors, antifungal peptides, aspartate proteases, antiglycaemic, hypocholesterolmic, and anti-cancer compounds in buckwheat makes it an important genetic base for isolation of genes for such proteins for use in biotechnology industry. The large collection of buckwheat germplasm available in different repositories across the globe could be the source of genes for various quality traits present in buckwheat. The variability in buckwheat germplasm can be seen as a source of novel genes for the improvement of other crops. Rout *et al.* (1997) have isolated a 26 kDa basic subunit of the 13S globulin which has more than 6% lysine and about 2% methionine. The gene coding for this protein could be a valuable source for improving the nutritional imbalances in cereals which are generally deficient in lysine. Full length cDNA and genes for legumin-like storage polypeptides from buckwheat have already been cloned (Bharali and Chrungoo 2003; Samardzic *et al.* 2004). Belozersky *et al.* (1995) and Park *et al.* (1997) have isolated two distinct proteinase inhibitors, BWI-1 and BWI-2a, from buckwheat grains. While BWI-1 has, on the basis of amino acid sequence similarity, been identified as a member of the proteinase inhibitor I family, BWI-2a has been reported to be a novel inhibitor with similarity to a plant storage protein vicilin (Park *et al.* 1997).

Cytotoxicity assays of BWPI, a proteinase inhibitor from buckwheat grains, on HL-60 cell strain of acute myelocytic leukemia, have revealed significant repression of the proliferating of HL-60 cells, and lower cytotoxicity on normal cells with (IC_{50}) of 0.29 and 1.01 μg l^{-1} for the myelocytic leukemia cells and normal cells, respectively (Yang *et al.* 2007). Similarly, Zhang *et al.* (2007) has demonstrated the efficacy of recombinant buckwheat trypsin inhibitor (rBTI) in inhibiting growth rate of cancer cells without any effects on normal human peripheral blood mononuclear cells (PBMCs). These results indicate that buckwheat protease inhibitors could have potential applications in treatment of cancer.

Buckwheat BWI is known to efficiently inhibit serine proteases that are the main proteolytic enzymes in many microorganisms and insect pests, and it also inhibits the growth and germination of pathogenic microflora (Dunaevsky *et al.* 1997). It showed inhibition against the mycelia growth of *Alterneria alternata* and *Fusarium oxysporum* and proteases of animal and bacterial origin (Dunaevsky *et al.* 1995). Recently, Khadeeva *et al.* (2009) synthesized an artificial gene corresponding to the amino acid sequence of BW-1 isolated from buckwheat. *In vitro* translation of these gene yielded recombinant product, whose molecular mass corresponded with protease inhibitor, BW-1a. This synthetic gene was transferred into tobacco and potato in order to increase the resistance of these plants against biotic stress. The transformed tobacco and potato plants exhibited microbial activity in biotest. Transformed tobacco lines showed strong resistance to white flies (*Trialeurodes vaporariorum*) as compared to the untransformed plants. The use of this gene may be extended to other crops as well. The high metal binding capacity of buckwheat makes it an ideal candidate to look for metallothionins and their genes for use in phyto-remediation processes (Maksimovic *et al.* 2004).

DRAWBACKS AND APPROACHES FOR IMPROVEMENT

Even though buckwheat cultivation has been going on for long time, the crop still has a long way to go in the direction of greater popularity and importance. Some of the inherent limitations in buckwheat like low and unstable yields, indeterminate growth habit, seed-shattering, lodging, presence of allergenic compounds, bitter taste and low shelf life of its flour restrict its popularity. The main cause for the decreesing popularity of buckwheat is low yield and its allergenicity. Two important factors responsible for low productivity of buckwheat are self-incompatibility caused from dimorphic heterostylism, incomplete sexual organs (especially in female), failure of fertilization and seed collapse in post zygotic stage (Adachi 1991). Only 10 to 20% of the flowers are reported to set seeds (Obendorf 1991). Adding to these problems are the high losses of grains at harvest and thrashing as a result of shattering (Fesenko 1986; Alekseeva and Malikov 1992) and logging (Fesenko 1986) and indeterminate growth habbit (Tahir and Farooq 1991b).

The susceptibility of common buckwheat to frost is another major bottleneck in expanding the areas of cultivation especially at high altitudes. Although screening of accessions has shown very little variability in tolerance to frost damage, this aspect is now being addressed through interspecific hybridization. The higher capacity of *F. tataricum* and *F. homotropicum* to tolerate cold and frosts is being evaluated to determine if the trait can be transferred between species. If the character can be transferred then a major constraint to the production of this crop could be altered or removed.

The major bottle neck in buckwheat breeding is its heterostylous incompatibility. Many efforts have been made to produce self-pollinating buckwheat for ease in genetic manipulation and evaluation. Samimy *et al.* (1996) attempted hybridizing *F. esculentum* with *F. tartaricum*, which is a self-compatible species with homomorphic flowers, but the resulting progenies were sterile. A major breakthrough was achieved by successful interspecific hybridization between *F. esculentum* and *F. homotropicum* in which the self-compatibility trait was introduced from *F. homotropicum* into *F. esculentum* (Campbell 1995). This successful inter-specific cross between *F. esculentum* and *F. homotopicum* at diploid level has opened new possibility for improvement of common buckwheat. The development of self pollinating buckwheat allowed some program to rapidly develop characteristics that affect yield in different ways. Unfortunately, *F. homotropicum* has severe seed-shattering, leading to considerable losses in grain yield. Little is known about the inheritance of seed shattering in buckwheat because both cultivated buckwheat species *F. esculentum* and *F. tartaricum* are resistant to shattering. A major crossing program has revealed that *F. esculentum* has recessive alleles at two or three loci which affect the abscission layer development in the pedicel and consequent shattering (Matsui *et al.* 2004; Wang *et al.* 2005). Studies undertaken by Wang *et al.* (2005) indicated the occurrence of recessive epistasis type of gene interaction, with two alleles at each of the three loci controlling shattering in buckwheat. Homozygous recessive genotypes at minimum of one locus produce the non-shattering phenotype, while genotype with dominant allele at each of the three loci produces the shattering phenotype. Thus, non-shattering genotypes could be produced by crossing between two shattering parents or by selfing plants that are heterozygous at different loci.

The scarcity of photosynthetic products in kernel is also reported to be one of the main factors that cause low yield in buckwheat (Yang *et al.* 1998). Buckwheat flowering is profuse and the photosynthetic capabilities of the plant do not meet the requirements to properly fill all the seeds. If the yielding ability of common buckwheat were to double, the plants only require 24% of the flowers now being produced. The nutrients that are now being expended on the production of the 76% of flowers it does not need, can then be redirected into filling more seeds. Therefore, the present emphasis in the self-pollinating breeding program is a reduction in flower number. The yield potential of the self-pollinating buckwheat would then be determined by the photosynthetic capability of the plants (Woo *et al.* 2006).

Common buckwheat is known to contain highly allergenic hypersensitive proteins (Smith 1909; Nakamura and Yamaguchi 1975). The major allergenic protein characterized from buckwheat is a 24 kDa protein which has been shown to bind to patients' IgEs at a higher frequency and with stronger activity than other proteins from the grains (Kondo *et al.* 1993; Nair and Adachi 1999; Park *et al.* 2000; Yoshimasu *et al.* 2000; Kondo *et al.* 2001; Tanaka *et al.* 2002). Mutation breeding for negative immunoreactivity or identification of lines lacking the major allergenic proteins could be an important step leading to greater utilization of buckwheat. Identification of two species of *Fagopyrum viz. F. lineare* and *F. urophyllum* that lack a 22 kDa allergenic protein by Nair and Adachi (2002) indicates the possibility of identification of other species/cultivars lacking such allergenic proteins. Nair and Adachi (2002) have highlighted the possibility of development of hypoallergenic cultivars from autogamous common buckwheat by development of a null mutant for the gene(s) encoding the 22 and 24 kDa allergenic proteins. The allergenic proteins could be modified by regulating the biosynthetic pathways for altered expression or by using gene silencing techniques. Similar approach has been used successfully to develop hypoallergenic peanuts (Dodo *et al.* 2007). Le *et al.* (2006a, 2006b) have used RNAi strategy to silence the allergens Lyc e 1.01 and Lyc e 1.02, two highly similar isoforms of tomato profilin. While RNAi technology may be a promising tool for developing hypoallergenic buckwheat, the threat exists if the major allergenic proteins are essential requirement for some normal plant function. This has been amply demonstrated in the case of profilin, which is an important pan-allergen for humans and also a protein that is essential for plant cell function. On the other hand, the successful production of developmentally normal Ara h 2-deficient transgenic peanuts and ns-LTP- deficient transgenic tomatoes shows that many food allergens can be reduced or eliminated without any associated yield or growth penalty.

FUTURE PROSPECTS

The potential of buckwheat utilization is known to be high and diverse. All the aboveground parts of buckwheat can be used in different industries. Buckwheat, therefore, can be cultivated as a multipurpose, fully utilized crop. One of the major constraints in enhancing the buckwheat cultivation is the low seed set due to high rate of abortion and consequently low grain yield. However, despite having been identified as a major problem in buckwheat, the causative factors underlying seed abortion are not fully understood. While *F. tataricum* and *F. homotropicum* do not show any seed abortion, the absence of genetic variability amongst different populations of common buckwheat for seed abortion indicates that breeding programmes aimed at developing varieties with reduced seed abortion would have to focus on mutational/interspecific breeding approach. Factors affecting breeding efforts in buckwheat include the phenomenon of self incompatibility, apomixis, sterility, low seed set and shattering. Breeding barriers in buckwheat have been identified as a major bottleneck in crop improvement programmes for the genus. These phenomena pose a challenge to national research capabilities in increasing buckwheat yields in a sustainable manner. In this context the use of molecular approaches including in vitro culture techniques, development of QTL maps and construction of detailed linkage maps would assume significance for initiating crop improvement programmes in buckwheat. The isolation of a full length clone for the lysine rich seed storage protein of buckwheat is an important contribution in the direction of utilizing the quality traits of this crop for improvement of other conventional crops through biotechnological approaches.

DNA marker systems have been developed in the genus *Fagopyrum* to assist germplasm management, selection within the breeding pool or gene introgression from wild species, and for disease diagnosis. The identification of self compatible species *F. homotropicum* has opened up the possibility of interspecific transfer of traits within the genus. Efforts also needed to be made towards development of hypoallergenic cultivars of buckwheat with reduced levels of antinutritional factors so as to improve the nutritional quality of seed proteins of buckwheat.

Since gene flow between wild and domesticated populations is likely to play an important role in evolution and domestication process, distribution mapping and characterization of wild progenitors of domesticated crops assumes great significance. Although a great deal of information is available on the origin, distribution of the various species of buckwheat as well as interspecific diversity within the genus *Fagopyrum*, several issues on intraspecific differentiation remain to be resolved. Further information on distribution of the wild progenitors and their diversity is scanty. The origin and intraspecific differentiation of *F. gracilipes*, a tetraploid self-fertilizing species, have not yet been clarified. Intra- and inter-specific differentiation of three recently discovered species, *F. pleioramosum*, *F. macrocarpum*, and *F. callianthum* is also an unsolved problem. Hence, there is a need to lay greater emphasis on collection and characterization of wild species of the genus from diverse geographic regions. This will not only facilitate their use in breeding programs but will allow a clearer understanding of the site of origin and the differentiation in the genus *Fagopyrum*.

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