



Wild Genetic Resource in Vegetable Improvement: Applications and Strategies

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Authors' contributions

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ABSTRACT

Domestication of feral types of the present-day cultivated types was in vogue in earlier days of farming followed by the selection and hybridization that led to many improved varieties. Expression of traits faded due to the development of homozygosity due to repeated interventions in the same set. Relying on wild relatives that are found to be the source of many target genes is found significant. While, this is not so easy, because of the barriers that reside in distant hybridization, consequences like failure in zygote and embryo formation, embryo death, linkage drags, and

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incompatibility within wild and cultivated groups, etc are need of concern. Strategies like bridge crosses, somatic hybridization, grafting, embryo rescue, etc., techniques are solely or in combination can combat these issues. Vegetables are the group of crops that are mainly cultivated for their high returns and quality aspects for the healthy food routine of the consumer. This review will spotlight the recent advances in vegetable improvement through their wild relatives and available strategies to overcome the barriers to the usage of wild relatives in vegetable improvement, meanwhile the conservation of these resources for the future utilization in vegetable improvement.

Keywords: Bridge cross; embryo rescue; polyploidy; somatic hybridization; vegetable grafting.

1. INTRODUCTION

As it is well established vegetables perform their prominent role in combating the insufficiency of the global food demand in terms of nutrition. Also, the vegetable consumption has been proved to be helpful in fighting against the several health issues, either as preventive or curative [1]. Hence, the vegetables are eulogized to be protective food. The varietal wealth of the vegetables is found to be unique in terms of their nutrition profiles. The present varieties and hybrids of vegetables not only nutritionally rich they also possess some of the important traits that are being resistant to some of the potential biotic (pest and diseases) and abiotic (climate and soil) factors hindering the production of vegetable produce to fulfil the demand of consuming community [2].

The diminishing numbers regarding vegetable cultivating area and thereby vegetable production figures are still below the expected ranges that are hindering the nutritional and alongside the economic stability of the nation. Which could be answered by the research innovations of developing an all-round vegetable variety that could be of high yielding, well adoptable, nutritionally rich and resistant to biotic and abiotic factors [3]. Although the development of an ideal variety of a crop in all senses, seems to be fictional, it is of utmost importance to be accomplished to fulfill consumer demand. The present-day cultivars at once, when they were considered to be novel were found to have all these desired characteristics. As reviewed by Bharathkumar et al [4] okra variety Pusa Sawani when released was found to be the one high-yielding variety due to its resistance towards Yellow Vein Mosaic Virus (YVMV) but with the time, the resistance of the variety found to be faded, the reasons may be the build-up of new viral strains against resistance mechanism, hindered genetic diversity in the existing germplasm due to selection pressure or any other demanding the search for a new source of

resistance [2]. Though there are many breeding strategies that are being employed in the development of the ideotype of the crop, none of the approaches are found solitarily sufficient in creating the gene that could be beneficial. As a solution, the most reliable treasure of these genes of interest lies in the feral types, these are the crops far related to the cultivated crops which were somewhere left behind in the wild in the course of domestication. The use of these wild relatives in the crop improvement is slow down due to some difficulties in the gene transfer to their cultivated farms. The common introgression difficulties are pre-syngamic (incompatibility, pollen tube death, etc.) and post-syngamic (embryo abortion, hybrid sterility, linkage drag, etc.). Many of the strategies have been designed to overcome the difficulties and surpass the challenges in utilization of the wild relative as a source of target alleles/ genes in the improvement of an ideal vegetable variety [5].

In this manuscript, the available wild source for useful traits (Table 1), and potential of the wild relatives in crop improvement, and the possible challenges to overcome with the potent strategies are been reviewed with special interest in vegetable crops. This could spotlight the possibilities of the utility of wild types in crop improvement.

2. CROP WILD RELATIVES IN VEGETABLE IMPROVEMENT

The domestication and the subsequent selection pressure (human and natural) towards their sustenance and suitability resulted in the attainment of homogeneity resulting in loss in diversity among cultivated forms, thereby the difference being non-significant in one or the other kind that hampered variability in germplasm, this may be referred as 'domestication bottleneck' [6]. This negative effect of domestication that led to loss of germplasm diversity can be surpassed by the use of crop wild relatives that are sources of

many useful traits in varietal development would bring the enhanced effect in terms of commercial yield, resistance to biotic hindrances, increment in the quality traits and also the wild gene introgression may also result in the improved adaptability to non-congenial abiotic factors [7].

In vegetable improvement there are many instances where these wild relatives are involved in crop improvement in all the mentioned aspects, the notable citations in the important vegetable crops are mentioned below with some noteworthy information that would support the use of wild relatives in vegetable improvement.

2.1 Yield Traits

In any crop improvement cycle the breeders certainly concentrate on the improvement of the yielding capacity of the resulting varieties. Many conventional breeding strategies like heterosis breeding between two high-yielding varieties or hybrids, and correlational studies to indirectly bring in the increment in one such complex trait i.e., yielding capacity by concentrating on corresponding traits etc., are found to give satisfactory results in many vegetable crops [41]. In this course, the use of wild relatives in interspecific hybridization programs also showed the considerable high-yielding potential in many vegetables as opined by Sun et al. [6] in the interspecific hybridization between *S. melangena* with the wild species *S. incanum* and *S. insanum* which are of primary gene pool resulted in the better fruit set along with good seed count in the fruit however, the fruits obtained by the cross with *S. sisymbriifolium* were found to be parthenocarpic in nature which of processing significance. The increment in the yield can also indirectly result from decreasing the unmarketable fruits in the brinjal crop as reported by Bhatt et al. [42] the distant hybridization of Pant Rituraj with *S. gilo* (wild brinjal). *Solanum viarum* crossed with *S. melangena* proved to increase the marketable yield with a very negligible incidence of fruit and shoot borer in brinjal confirmed through the RAPD (random amplified polymorphic DNA) markers. Bhatt et al. [42] crossed *Capsicum chinense*, *C. annuum* and *C. frutescens* among each other and confirmed the increment in yield both morphologically and through RAPD markers in chilli. As well established that Cole crops have their ogura CMS through their wild forms and this may be used in the improvement of cultivated farms Singh et al. [43] proved that the CMS lines of wild origin namely *Ogu402-6A*, *Ogu76-33A*,

and *Ogu76-4A* being helpful in an expression of earliness in cultivated farm upon introgression. Tan et al. [44] reported that the wild-originated gene *CsACS2* the encoder of a candidate gene (1-aminocyclopropane-1-carboxylic acid synthase) for monoecism (*m*) that is associated with fruit shape (elongated) and yielding attributes of cucumber. Yield increment through wild relatives is not only possible by hybridization, as reported by Kumar et al. [18] the wild *Solanum torvum* and *Solanum khasianum* upon their use as a rootstock in grafting with two improved varieties Pusa Shamala and Pusa Hybrid 6 of *solanum melangena* type exhibited superiority in yielding capacity. The grafts of brinjal cultivar *GJB-3* on *S. torvum* were profitable in yielding ability [42]. The grafting success varies with the various rootstock species, the graft of *Cucumis sativus* on *C. ficifolia* (fig leaf gourd) was found to be highly successful (70.23%) on the other hand the rootstock of *Legenaria siceraria* (bottle gourd) resulted in the least (63.91%) success [45]. Wild tomato species (*L. pimpinellifolium*) were found to give the highest number of seeds per plant, and fewer days for flowering and fruit setting [46].

2.2 Quality Enhancement

Though getting a high yield is one of the main objectives of all the breeding proposals, along with the higher quantity of produce it is noticed that the produce that contains the best quality will fetch a good price in the market alongside the quality product also plays an important role in consumer health. Hence, there are many conventional breeding approaches to improve the quality traits of vegetables [47]. However, all the efforts for quality elevation of the vegetable are somewhere facing a shortfall in the fulfillment of the objective. This may be since there is obviously less genetic diversity for nutritional traits in cultivated form where the usage of wild farms pronounces its importance [48]. The CMS lines *Ogu13-85-6A*, *Ogu309-2A*, and *Ogu2A* of wild origin can act as a parent for improving the head compactness in cabbage [43]. Among 18 introgression lines of somatic hybridisation between 'Korso' a wild type and *Brassica nigra*, introgression line 9 (*IL9*) was noticed to be with Potassium content: of 236 mg/100 g [49]. With the use of wild relative, the quality of the cucumber can be efficiently improved [50] as the wild progenitor species *Cucumis hardwickii* incorporated the small size to the fruits to ease the whole fruit canning and also the fruit vitamin A content can be improved by the β carotene

promoter gene by crossing with *C. xishuangbannensis*. *Solanum galapogense* a wild relative to tomato found to be the source of β carotene [18]. *Solanum melaneense* a wild species is reported to contain starch content of 15.45 to 23%. The genome study of two wild relatives (*Ipomoea trifida* and *I. triloba*) of sweet potato confirmed their use in crop improvement with regard to increased carotenoid biosynthesis in the roots [51]. GABA (*γ-aminobutyric acid*) tomatoes are gaining importance in human health wild tomatoes are found to be a very efficient source of gene encoding to GABA [52]. Ghani et al. [46] noticed that *Solanum pimpinellifolium* could be used successfully for improving the quality traits like total soluble sugars (TSS) and lycopene content in fruits. As flavour of tomato or any fruit is the critical factor for its consideration by consumer, flavour can be altered by variation in the synthesis of sugar and amino acids, the wild tomato *S. pimpinellifolium* on QTL analysis was found to have genes helpful in flavour enhancement of tomato and also this wild type of tomato found to be the accelerating factor of lycopene synthesis [43].

2.3 Biotic Stress Resistance

Biotic stress factors like insect pests and diseases are one of the prominent constraints in food production that can be a direct cause of detrimental quality and decreased quantity of the production. [53]. The majority of vegetable crops are very succulent in nature and also rich in nutrients that make them vulnerable to the feeding of insect pests and infestation of serious pathogens respectively [54]. Though there are many improved varieties of vegetables that are bred through conventional breeding methods and majority of the resistance varieties have also already lost the resistance to these biotic stresses. This may be due to the reason that the pest (pathogen and insect) will adapt for the resistance reaction and adapt to feed or survive on the resistance cultivar itself eventually making the cultivar susceptible [55]. One classical instance is the Pusa Sawani of okra which is the result of hybridization between IC-1542 × Pusa Makhmali which was notified to be resistant to the infestation by aphids and consequently resistant to the YVMV infestation, but for now this variety have lost its resistance towards the virus. Another case is H24 (Hisar Anmol) of tomato which was evolved by the interspecific crossing of Sel7 (Hisar Arun) with the *Lycopersicon hirsutum* f. *glabratum* adopting modified pedigree

back cross method was reported to be resistant to TLCV but this resistance has been degraded and brought it to the class of moderately resistant and it is not surprising if after some years or growing seasons it attains the rank of susceptible. This degrading resistance reaction in the developed varieties may be due to numerous causes such as the evolution of virulence strains or insect life cycle, degradation in the purity of gene that is responsible for resistance or lack of maintenance breeding, etc. resulting loss in resistance, this finally demands the rediscovery of resistance genes in the wild farms and incorporate such genes in the cultivated types. As reviewed by Karim et al. [56] the chilli wild relative *Capsicum chinense* PBC932 was able to donate the resistance to the cultivated *C. annuum*. The leaf curl of chilli caused by begomovirus through white fly vector and the wild species *S. pseudocapsicum* was found to be asymptomatic to begomovirus [57]. *S. incanum* as a wild relative of brinjal is found to decrease the incidence of *Leucinodes orbonalis* (fruit and shoot borer) by 5.3-8.6% over the years [58]. The wide hybridizations between cultivated *Brassica napus* and wild *B. rapa* subsp. *sylvestris* lines were also found efficient in transmitting the resistance along with the linkage of a pair of SSR (simple sequence repeats) and IP (intron polymorphic) markers to club rot resistance in Cole crops by mapping the resistance gene on chromosome A03. IL6 was found to be resistant to clubroot race 4 among 18 introgression lines of somatic hybridization between 'Korso' a wild type and *Brassica nigra* [49]. In okra the biotic stress like the white fly being a very destructive pest due to its virus (Yellow Vein Mosaic) transmitting nature, the wild species *Abelmoschus caillei* has proven to be 66% efficient in being a source for white fly resistance [59] along with this species the other wild species namely *A. manihot* and *A. moschatus* are also efficient in confirming the resistance against fruit borer and hoppers. The wild resource can be undoubtedly referred as treasure of resistance genes, the wild types of potato *Solanum albornozii*, *S. andeanum*, *S. lesteri*, *S. longiconicum*, *S. morelliforme*, *S. stenophyllidium*, *S. mochiquense*, *S. cajamarquense*, and *S. huancabambense* are reported to be resistance source for late blight disease of potato [60] in addition to *S. cajamarquense* one more wild from *S. sogarandinum* also found promising in late blight resistance breeding of potato [61]. In carrot improvement, *Daucus capillifolius* is trusted to be the source for carrot fly resistance.

Table 1. Wild relatives as a source of targeted traits in vegetable improvement

Vegetable crop	Wild Relative	Targeted Trait	References
Tomato (<i>Lycopersicon esculentum</i>)	<i>L. hirsutum</i> and <i>L. peruvianum</i>	Fungal resistance	[8]
	<i>L. peruvianum</i>	Nematode resistance	
		Source for <i>Tsw^δ</i> gene (Spotted wilt resistance)	[8]
	<i>Solanum habrochaites</i>	Leaf eating caterpillar resistance	
		Late blight resistance	[9,10]
		Source of Ty-2 gene (TYLCV)	
	<i>S. pinnellii</i>	Fruit ripening, high brix content and tolerant to drought	[11]
	<i>S. pimpinellifolium</i>	High soluble solids	[12]
	<i>S. chesmani</i> and <i>S. galapogense</i>	Salinity stress	[13]
	<i>S. Cheesmaniae</i>	Jointless gene (<i>j-2</i>), β carotene and thick skin	[14]
	<i>S. chemielewskii</i>	High sugar	[15]
<i>S. neorickii</i>	Bacterial wilt resistance	[16]	
<i>S. chilense</i>	Resistance to drought and virus	[13]	
Brinjal (<i>Solanum melangena</i>)	<i>S. khasianum</i> and <i>S. avivulares</i>	Solasodine rich	[17]
	<i>S. khasianum</i> and <i>S. viarum</i>	Resistance to shoot and fruit borer	[17]
	<i>S. xanthocarpan</i> and <i>S. torvum</i>	Resistance to phomopsis blight	[18]
	<i>S. sisymbriifolium</i> and <i>S. auriculatum</i>	Resistance to little leaf	[19]
	<i>S. macrocabon</i> and <i>S. gilo</i>	Tolerant to drought	[17]
	<i>S. sisymbriifolium</i> and <i>S.indicum</i>	Resistance to root knot nematode	[19]
	<i>S. integrifolium</i>	Fusarium wilt resistance	[17]
Chilli (<i>Capsicum annum</i>)	<i>C. frutescens</i>	Small highly pungent perennial nature	[20]
	<i>C. pubescens</i> and <i>C. microcrpum</i>	Powdery mildew resistance	[21]
Potato (<i>Solanum tuberosum</i>)	<i>S. demissum</i>	Resistance to late blight, leaf roll and potato virus X	[22]
	<i>S. chacoense</i>	Charcoal rot resistance	[23]
	<i>S. verrucosum</i> and <i>S. microdontum</i>	Resistance for late blight	[24]
	<i>S. raphanifolium</i>	Tolerant to cold induced sweetening	[25]
	<i>S. avilesii</i> , <i>S. terijense</i> , <i>S. chacoense</i> and <i>S. veturii</i> .	Source for <i>Rpi</i> gene (resistance to late blight)	[26]

Vegetable crop	Wild Relative	Targeted Trait	References
Okra (<i>Abelmoshus esculentus</i>)	<i>A. tuberculatus</i>	Symptomless resistance to YVMV	[27]
	<i>A. manihot</i> var. <i>manihot</i>	Immune to YVMV	[28]
	<i>A. pungens</i>	True resistance to YVMV	[28]
	<i>A. tuberculatus</i> and <i>A. caillei</i>	Tolerant to shoot and fruit borer	[27]
Cole crops (<i>Brassica</i> spp.)	<i>B. carinata</i>	Source of male sterility and powdery mildew resistance	[29]
	<i>B. nigra</i>	Chromosome B4 (resistance to <i>Leptosphaeria maculans</i> [blackleg])	[30]
	<i>B. hirta</i>	Resistance to leaf blight of cabbage	[31]
	<i>B. fruticulosa</i>	Male sterility to <i>B. juncea</i> , <i>B. oleracea</i> and <i>B. napus</i> .	[32]
	<i>B. napus</i>	Low euricic and low glucosinolate content	[33]
Sugar beet (<i>Beta vulgaris</i>)	<i>B. procumbens</i> , <i>B. webbiana</i> and <i>B. pettellaris</i>	Cyst nematode resistance	[34]
	<i>B. vulgaris</i> L. ssp. <i>maritima</i>	Cyst nematode resistance	[35]
Carrot (<i>Daucus carota</i>)	<i>D. carota</i> subsp. <i>carota</i> , <i>D. carota</i> subsp. <i>Capillifolius</i> and <i>D. carota</i> subsp. <i>Gummifer</i>	Abiotic stress (salt, drought and flood) tolerant	[36]
Cowpea (<i>Vigna unguiculata</i>)	<i>V. luteola</i> , <i>V. marina</i> and <i>V. vexillat</i>	Salinity tolerance	[37]
	<i>V. radiate</i> var. <i>sublobata</i>	Yellow mosaic virus	[38]
	<i>V. unguiculata</i> group <i>sesquipedalis</i>	Temperature and salinity stress tolerance	[39]
Pea (<i>Pisum sativum</i>)	<i>Lathyrus</i> spp.	Broomrape weed resistance	[40]

2.4 Climate Resilience and Adaptation

The successful crop can be raised with availability of the suitable condition, this could have become the challenge or otherwise [62]. It is noticed that the highly improved cultivation technology consisting of integrated nutrient management, well-protected structure, high yielding varietal seed inputs though applied the failure of the suitable climatic condition (areal or soil) leads to the partial or complete failure of the crop [63]. The vegetable crops are especially found to be very sensitive to unfavourable growing conditions and also noted to be stage selective towards climatic factors for instance the tomato crop requires a specific temperature for lycopene production and maintenance as the temperature of 20-24 °C is found to be optimum but the temperature below 10 °C during the night and more than 30°C will lead for dropping down of its production and ultimately the hotter surroundings with 40°C will full breakdown the lycopene and makes the appearance inferior [64]. The wild relatives are trusted to be the final resort for the resistant genes for these adverse climate conditions as these wild relatives themselves evolved in the challenging climates. Hence, the incorporation of the wild *capsicum xyloglucan* endotrans-glucosylase/hydrolase (*CaXTH3*) gene into cultivated and reported tolerance towards salinity and drought. Wild types of okra (*A. tetraphyllus*) are found to be efficient in incorporating the drought-tolerant gene into cultivated farms [65]. Suma et al. [66] confirmed the importance of the use of wild species (*Abelmoschus pungens* var. *mizoramensis*, *A. enbeepeegeearensis*, *A. caillei*, *A. tetraphyllus* and *A. angulosus* var. *grandiflorus*) for introgressing the adoptability traits for adverse growing situations. The continuously changing climate is found to affect the crop growing areas there by impacting the shift in crop cultivation to the non-agriculture side Fumia et al. [67] performed a simulation study and inferred that the potato wild relatives (*Solanum gracilifrons* and *Solanum salasianum*) would be the source of genes to fight against changing climatic specifications. The wild potato species *S. chacoense* and *S. commersonii* reported to impart the high temperature tolerance to the introgressed crop *S. melameanum* is found to be the source for freezing tolerant genes that can withstand -5°C [68]. With other crops carrots are also susceptible to biotic stress to certain extent, the wild relative of carrots i.e., *Daucus carota* L. subsp. *Capillifolius* can be used as a donor for drought and heat sustenance [36]. The

starchy root crop cassava (*Manihot esculenta*) is found to be compatible with its progenitor species *M. esculenta* ssp. *flabellifolia* rendering it to be successfully grown in water logging areas. *Lactuca serriola* the wild relative of the lettuce reported to be useful in crop improvement against salinity and drought since, the wild genes that are confirmed with SNP (single nucleotide polymorphic) markers to have six QTLs that are associated with the deposition of mineral ions on roots of crop rendering adaptability [69]. Tomatoes are found to be very sensitive to the herbicides sprayed, this can be solved by incorporating the gene that is prominent in the biosynthesis of 7-epizingiberene the farm of sesquiterpene, *S. habrochaites* the wild farm of tomato is found very eminent in making cultivated tomatoes herbivore resistant and this wild species can also efficiently improve the quality traits of tomatoes like improved antioxidant activity, improved DPPH (2,2-Diphenyl-1-picrylhydrazyl) radical scavenging activity combined with higher phenols and flavonoids [70].

3. STRATEGIES TO USE WILD-FARMS

Though the wild relatives are found to be of the greater significance in vegetable improvement they are considered to be the last resort for the crop improvement [71]. The main reason for this is the barriers present in the way of using the wild farms in crop improvement. The distant hybridization is found to be hindered due to pre-syngamic and post-syngamic difficulties which are recorded to be incompatibility (genetic and genomic) and barrier (ploidy level, crossability, and hybrid sterility) [72]. The failure in the success of wide hybridization involving wild farms may be due to factors that the lack of fertilization, inability of the pollen tube to reach the embryo sac, failure in the formation of the zygote, and death of the zygote after formation. To overcome this many ways are found to be efficient in getting wide hybridization successful. Among many of the techniques breeding strategies like protoplast fusion, embryo rescue through embryo culture, in-vitro methodologies (anther culture, pollen culture, and haploid rising), bridge mating, and grafting are found to be efficient [73].

3.1 Bridge Cross

Wide hybridization faces the prominent problem of failure in crosses between the wild and cultivated farm, this may be due to the change in

ploidy levels of both the parents in hybridization since many of the wild farms are found to be polyploids in nature this can be overcome by a strategy of employing the bridge crosses [74]. Manzur et al. [75] overcame the cross incompatibility between *cucurbita* species by employing the *cucurbita lundelliana* as a bridge species for the transfer of bush habit from *C. pepo* to *C. moschata* and *C. maxima* and these interspecific hybrids were also found to be multiple disease resistant. Parry et al. [76] noted the use of *Capsicum frutescens* as a bridging species in the cross of *C. annuum* and *C. baccatum* for transfer of anthracnose resistance. The cultivated rape seed can be made *sclerotinia* resistant by transferring resistance genes from wild *Brassica incana* by the use of *Brassica napus* as a bridging species [77]. The usage of this bridge crosses demands in-depth knowledge of the intermediate species that are compatible with both wild and cultivated forms facilitating the transfer of a targeted genes from wild to cultivated relatives, in this regard there is a need to study the available genetic resources both cultivated and wild farms and their crossability and this can be achieved by the molecular analysis using a wide range of molecular marker for diversity analysis.

3.2 Somatic Hybridization/ Protoplast Fusion

Many of the wild farms are also found to be not crossable with the cultivated ones, this can be dealt with the somatic hybridization or protoplast fusion, where the excised protoplast of both the parents are made to fuse outside the seed parent [78]. Protoplast fusion between *Solanum melangena* and *Solanum torvum* that showed resistance to the *verticillium* wilt. Somatic hybrids resulting from the symmetric fusion of two protoplast from the wild species found to be of great importance in the crop improvement, hybridized the protoplasts of *Solanum integrifolium* and *S. sanitwongsei* employing the electro fusion method and the resulting hybrids were of targeted trait viz., yield and quality and were of $2n=48$ indicating symmetric hybridization [79]. Somatic hybridization is also helpful in the regeneration of the intergeneric species, *Alternaria brassicae* the devastating disease of the brassica family does not have any resistant source in its species or genera. However, the somatic hybridization between the *Brassica oleracea* and *Camelina sativa* (false flax) found to be imparting resistance to *alternari*. Not only in the crop improvement but this somatic hybridization also has its part in the creation of

the new species like pomato, this was done with the intension of getting tomato fruits and potato tubers in a single plant but the results were opposite to the breeder's expectations [80]. The resistance against the *Phytophthora infestans* from the wild *Solanum michoacanum* can be transferred to cultivated diploid potatoes through somatic fusion [81].

3.3 Embryo Rescue

If at all the hybridization is achieved through bridge crosses, manipulating the pollinating organs (pistil and stigma) of wild and cultivating farms to facilitate hybridization [43], there exist challenges regarding the formation and survival of the embryo. This can be dealt with the procedures like embryo culture which is also referred to as embryo rescue as it saves the denaturing embryo through in-vitro procedures. The wild relatives of tomato like others also showed the embryo degeneration after 25 days of fertilization in between *Solanum lycopersicum*, and *S. peruvianum*, this is abated by the embryo rescue of the cross on MS (Murashige and Skoog's) media and the hybridity being confirmed using RAPD markers [82]. Though the success of embryo rescue is well documented in many interspecific crosses, this strategy also depends on some specification like age of rescued embryo, shape of the embryo, etc., in interspecific hybridization of *Brassica rapa* × *B. oleracea* rectified that the embryo rescue after pollination at cotyledonary stage gave better results, Zaman and Parihar [83] also proved the embryo culture of the cross between the cultivated and wild forms led to improvement of okra varieties resistant to YVMV diseases.

3.4 Ploidy Alteration

The variation in ploidy levels of the wild and cultivated crops also is regarded as a threat to crossability, this demands techniques that can alter the ploidy level of the candidate species in distant hybridization, the chemical is widely used in the alteration of ploidy levels is colchicine ($C_{22}H_{27}O_6N$) [84]. The colchicine treatment at 0.1% for F_1 of the cross *Abelmoschus esculentus* and *A. manihot* subsp. *Tetraphyllus* was found to be efficient in overcoming the hybrid sterility in the cross [85].

3.5 Grafting

Although all the above-mentioned strategies are found to have proved their efficiency in

overcoming the challenges that are in the way of distant hybridization that involve gametes of the parents some are found still challenging in terms of tediousness and special requirements [86]. One of the strategies that is comparatively easier and more effective is the grafting, where the cultivated farms are grafted on the rootstocks of wild relatives [87]. To achieve the highest yield, quality, and growth of brinjal cultivars the grafting over the wild rootstocks of *Solanum khasianum* and *Solanum torvum* were found to be suitable [18]. Performance of the grafted globe artichoke on the wild cordon and found to have resistance for *verticillium* wilt. Most of the wild farms of brinjal were found to impart the quality and resistance traits on grafting which were of incompatible reactions on hybridization [88]. Thangamani et al. [39] also evidenced that the usage of wild relatives gave a good result compared to distant hybridization in making cultivated form resistant to disease and pest and of superior quality in cucumber.

4. LIMITATIONS OF THE STRATEGIES

Though the applicability of all the above-mentioned strategies is proved in the vegetable improvement, these also persists some limitations or drawbacks that could be the limiting factors to world wide applications of these modern breeding methods. As in bridge crosses the main limiting issue is the discovery of the bridging species with not to minimum intervening unwanted linkages upon which requires more breeding cycles to break these linkage drags [89]. The strategies which are regarded to be most accurate in the results viz., protoplast fusion and embryo rescue demands the intense technical skills and specialized working condition that may be of financial concerns to the small-scale projects [90]. The method of ploidy alteration in the wide hybridization with anti-mitotic agents demands the more caution with respect to the dose, part used and time of application, this information is still insufficient in many vegetable crops [91]. Grafting poses the limitation in its application due to the incompatibility (immediate or later stage) that hinders the stable performance of the inter-grafts [92]. However, relying on the prior established information on wide hybridization and employing the suitable breeding strategies singly or combined may yields less risks and more fruitful results.

5. CONCLUSION

Wild relatives are found to be very useful in vegetable improvement regarding yielding

capacity, enhanced quality, and improved resistance towards biotic and abiotic stress. Usage of these wild relatives in the improvement of vegetable crops also showed a considerable increment in the targeted traits. The effective usage of these wild types in vegetable improvement demands some basic requirements like knowledge about the existing wild diversity of the concerned vegetable crop, knowledge of cross-compatibility and incompatibility reaction in between cultivated and their wild relatives, some negative issues linked with the wild farms of vegetable crops as some wild forms are also found to associated with high toxic compounds that may be resulting in inedibility of the produce and barriers or problems in the usage of wild types in vegetable improvement, available strategies and their application in course of developing improved varieties, etc., before using any wild farms in the crop improvement there should be an account of wild genetic diversity and also this demands the efforts for conserving the wild farms of cultivated vegetable types. The efficiency of the wild relative in the improvement of cultivated crop diversity is all the way proven, however due to overexploitation of these wild farms by humans is challenging their existence as a consequence of genetic drift, etc., factors, hence there is an urgent need to conserve these wild farms.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declares that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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