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Chapter

## Genetic Potential and Possible Improvement of *Sesamum indicum* L.

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#### Abstract

Sesame (Sesamum indicum L.) is one of the traditional oil seed crop widely cultivated in many countries. The top producers of sesame seeds are mainly Tanzania, Myanmar, India, China and Japan. Sesame oil contains high level of unsaturated fatty acids (80%) and low levels of saturated fatty acids (20%). The main fatty acids are palmitic, stearic, oleic, linoleic and trace amounts of linolenic fatty acids. Sesame seed contains 50–60% of high-quality oil rich in natural antioxidants such as sesamin, sesamolin, sesaminol and sesamol it enhances the stability and keeping quality of sesame oil. Sesame seeds have good sources of dietary fibre, fats, vitamins, minerals, proteins and rich in anti-oxidants. Polyunsaturated fatty acids in sesame will reduce the risk of high blood pressure, cardiac disorders and blood sugar levels. Sesame is believed to have been originated in India where maximum variability of genetic resources is available. High yielding varieties available to date have reached the yield plateau even with the advanced cultivation practices. The area under oilseed crops cultivation also reducing every year. Hence, there is an urgent need to increase the oil content and yield of Indian sesame varieties. Understanding the available germplasm and novel interventions to develop high yielding varieties warrant both molecular and phenotypic data which is meagre in case of sesame.

Keywords: sesame, germplasm, genetic diversity, improvement, oil content

#### 1. Introduction

Sesame is one of the imperative, oldest and underexploited oilseed crops in the world. Sesame seeds have different names in diverse locations such as ellu in Tamil, nuvvu in Telugu, til in Hindi, gingelly in English, and also with some other names such as sim sim, ajonjoli, benniseed and gergelim. In India, sesame is placed fifth in the list of edible oil crops after groundnut, rapeseed, mustard, sunflower and soybean [1]. Sesame is considered as a chief oilseed crop in the world due to its extraction process, good stability, and drought resistance [2]. Origin of *Sesamum indicum* is established by the existence of archaeological remnants seeing back to 5500 BC in the Harappa Valley of Indian subcontinent [3, 4].

#### 2. Botany of sesame

Sesemum belongs to Pedaliaceae family, which comprises 16 genus and 60 species. The number of species in sesame is not clear, though 40 species have been identified and 36 were mentioned in the Index Kewensis. In Africa, 18 species were available, 8 species were available in Indian-Srilanka region. All the wild species are prevalent in Africa. *Sesamum indicum*, *S. capense* Burm. (*S. alatum* Thonn.) and *S. schenkii* Aschers have a same somatic number 2n = 26. Other wild species such as *S. occidenale*, *S. radiatum* Schm & Thonn. has 2n = 64, *S. angolens* and *S. prostratum* 2n = 32, *S. laciniatum* 2n = 28. Nowadays, *Sesamum indicum*, ssp. Baumii, *S. malabaricum*, and *S. radiatum* are harvested and eaten rarely during food scarcity [4].

#### 2.1 Origin and Distribution

Sesame has a wide range of diversity and it was originated in Africa and spread early through West Asia, China and Japan. With the exclusion of *Sesamum prostratum* Retz, all the wild species are establish in Africa [5]. The inconsistency and the location of sesame in the economies of numerous African countries could further justify the African continent to be the ultimate centre of origin. However, Bedigian [6] established that the crop was first domesticated in India, citing morphological and cytogenetic affinities between sesame and the south Indian native *S. mulayanum* Nair, as well as archaeological evidence showed that it was refined at Harrapa in the Indus Valley between 2250 and 1750 BC. All these statements make it difficult to say with inevitability the precise origin of the crop. Due to its moderately low productivity sesame ranks only ninth among the top thirteen oilseed crops, which make up 90% of the world production of edible oil.

#### 2.2 Health benefits of sesame

Sesame seed oil is the most economical important product which is very stable in nature with good antioxidant properties and high PUFA content (**Table 1**) [7, 8]. Besides oil, seeds are also used in various culinary preparations. Sesame seed contains sesamin and sesamolin two lignans with medicinal properties. The term

Components of sesame seed	Quan	References		
	Sesame seed (mg g <sup>-1</sup> seed)	Sesame oil (mg g <sup>-1</sup> )		
Palmitic acid (16:1)	9.4%	14.45%	Hemalatha and	
Oleic acid (18:1)	39.1%	50.54%	Ghafoorunissa [7]	
Linoleic acid (18:2)	40%	45.50%		
Linolenic acid (18:3)	0.46%	0.85%		
Sesamin	8.80	6.20	Uzun et al. [8]	
Sesamolin	4.50	2.45		
Sesamol	1.20	_		
Sesaminol	1.40	0.01		

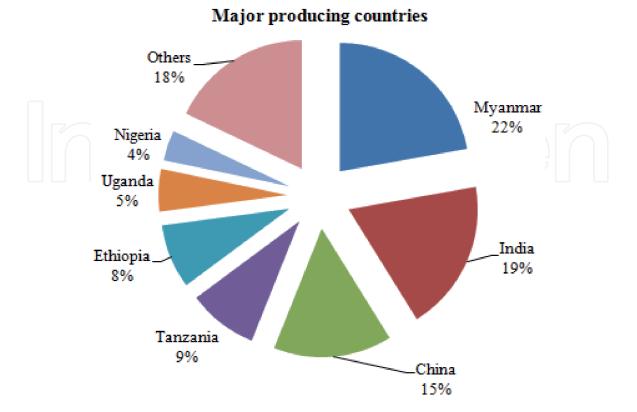
#### Table 1.

Bioactive components present in sesame seed and oil.

'Lignan' was coined by Howarth in 1948. It describes the group of dimeric phenyl propanoids that have therapeutic value. Sesamolin is converted to sesamol on roasting the seeds. Roasting is preferred in confectionary. The molecular structure of sesamol has phenolic and a benzodioxide group. It possesses antioxidant property and confers apoptotic effect in cancer cells. The pharmacological and health promoting effects of sesame seeds are anti-oxidant, anti- proliferative, anti-inflammatory, anti cholestrolemic, anti-hypertensive, lowering LDL, and guarding DNA mutants [9–12]. Sesame lignans also found to increase Vitamin E content in tissues which is also associated with aging process [13]. Besides seed oil and seeds, young leaves also found to have nutritional benefits and used in soup preparations in Africa [6].

#### 3. Sesame Production World Scenario

Sesame is an ancient oilseed crop valuable for export commodity in India. The major sesame producing countries are Myanmar, India, China, Tanzania, Ethiopia, Uganda, Nigeria and others (**Figure 1**). In 2018, 6,016,000 metric tonnes of sesame were grown world-wide on 11,743,000 hectares (ha) with an average harvest of 512 kg ha<sup>-1</sup>. Asia and Africa produced almost 97% of the world's source of sesame [14]. Globally sesame consumption is progressively raised due to consuming patterns and increasing health awareness of consumers. Consequently, the requirement of sesame seeds is higher at present. Sesame seed has numerous nutritional benefits such as minerals, fibre, protein and vitamins [15]. Tanzania is the highest sesame seed consuming country of about 21% (based on tonnes) followed by China (19%), Sudan (9%), Ethiopia, India, Myanmar and Nigeria (6% each) with approximately 74% of world's consumption [16].



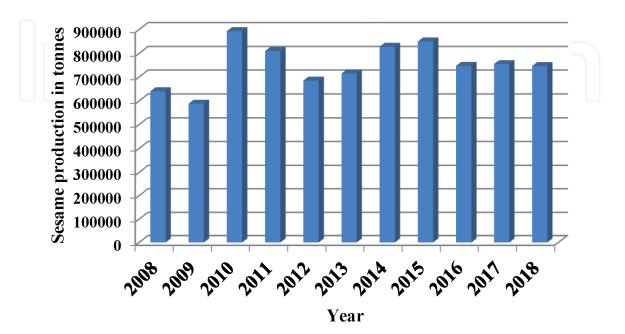
#### **Figure 1.** *Major sesame producing countries and its percentage of production. Source: FAOSTAT.*

#### 3.1 Sesame Production and export in India

In India, sesame is mostly cultivated in two seasons kharif (June–November) and rabi(November–April) and nearly 75% of annual production comes from kharif season. The major sesame growing states in India are Gujarat (1,16,200 ha),Uttar Pradesh (4,17,435 ha),Rajasthan (2,70,191 ha)and Madhya Pradesh (3,14,300 ha) together accounted for 85% of total acreage and other states served the remaining 15% percent [17]. The average productivity of sesame in India for the last years (2008–2018) are shown in Figure 2. In 2008, the production of sesame was around 640300 tonnes and it was increased in 2010 (893,000 tonnes) and it was gradually decreased in 2018 (746,000 tonnes). Global requirement for sesame has increased by nearly 80 percent. Sudan, India, Nigeria, Myanmar, Tanzania and China are the major exporters of sesame seeds and its products. China, Japan, South Korea, Turkey, Iran, Egypt, Germany and USA are the world's largest importers of sesame seed. Area covered, yield and production of sesame in world level was summarized in Table 2. India exported 312.62 lakh tonnes of 3920 crores value of sesame seed and oil in the year 2018–2019 [18]. The quantity, value and the share of sesame export for the year 2013 to 2019 was shown in Table 3.

#### 3.2 Production Technology

Fertile land with good irrigation and drainage facility is the most suitable land for sesame since it is sensitive to water stagnation. Fine tilth is suitable for sesame seed germination which can be obtained by couple of ploughings and few harrowing activities in any type of soil. A good field suitable for sesame cultivation should be free from weeds and levelled enough to avoid water stagnation. Seed rate used for a good crop stand is 4–5 kg/ha. Seed treatment with Thiram 3 g/kg or Thiram (1.5 g) + Bavistin (1.5 g) is prescribed to avoid seedborne pathogens. Line sowing is preferred for inter culture practices and high yield, when seed drills are used for sowing the seed rate can be reduced to 2.5 to 3 kg/ha. To avoid leaf spot diseases seed pre-treatment with 0.025% solution of Agrimycin-100 is suggested. The fertilizer recommendation for sesame is Sulphur 30 kg /ha in the form of gypsum+60:40:20 (N.P.K.) kg/ha. Sesame responds well to inorganic fertilizers and record higher



#### Figure 2.

Sesame productivity in India for the last ten years (2008–2018). Source: Food and Agriculture Organization Statistical Databases (FAOSTAT, 2019).

Sesame producing countries	Area Yield (kg ha <sup>-1</sup> ) ('000 ha)		Production ('000 MTha <sup>-1</sup> )	Percentage of World Production	
India	1730	431	746	12.40	
China	311	1393	433	7.20	
Myanmar	1463	525	769	12.78	
Sudan	3480	282	981	9.33	
Tanzania	800	701	561	14.56	
Nigeria	539	1063	573	9.52	
Ethiopia	415	726	301	5.01	
Uganda	210	667	140	2.33	

#### Table 2.

World status of sesame area, yield and production in 2018.

Year	Seed		Oil and fract	Total	
	Export Quantity (tonnes)	Rupees (crores)	Export Quantity (tonnes)	Rupees (crores)	(value)
2013–2014	2574.4109	3583.46	6.48973	87.45	3670.92
2014–2015	3756.5607	4717.77	7.07017	98.54	4816.30
2015–2016	3284.5572	3012.31	11.17834	77.63	3089.94
2016–2017	3073.2856	2695.84	12.59895	96.61	2792.45
2017–2018	3368.5038	2990.93	9.45222	140.22	3131.14
2018–2019	3119.8706	3761.93	9.22864	165.53	3927.46

#### Table 3.

Quantity of sesame seed and oil exported from India for the period 2013–2014 to 2018–2019.

seed yield. At the time of sowing half of the recommended nitrogen and full of P and K are used. Rest of the nitrogen is applied during flowering stage. Biofertilizer applications such as Azotobactor and phosphorus solubilizing bacteria have resulted in higher yield. In addition foliar spray of urea 2% at flowering and capsule formation resulted in higher yield. In addition to the recommended fertilizer dose, micronutrients zinc 20 kg/ha, iron 25 kg/ha and Farm Yard Manure (FYM) 2.5 t/ ha has resulted in maximum yield of sesame [19]. In situ moisture conservation can be accomplished by stirring the soil after each rain and soaking seeds for 8 h in thiourea (500 ppm). Kharif crop requires protective irrigation of 4–5 times depending on the soil type to overcome the moisture stress. Winter crop needs scheduled irrigation for 2–3 times. A good germination and crop establishment is observed when seeds are soaked prior to sowing named as seed priming [20].

#### 4. Sesame crop improvement in Tamil Nadu

Tamil Nadu, the South most state of India harbours several land races and wild sesame varieties. Tamil Nadu Agricultural University is actively involved in genetic improvement of sesame. **Table 4** summarises the varieties released by this University. The following section deals with a recent morphological evaluation of its germplasm for selection for further use. Sesame crop improvement by crossing

Crop/	Year	Parentage	Duration	Yield	(kg/ha)	Special features	
Variety	ofrelease		Days	Rainfed	Irrigated		
TMV 1	1939	Mass selection from Palani (local)	85	300	600	Erect, fairly bushy with moderate branching, 4 loculed reddish brown to black seeds. Oil 50%	
TMV 2	1942	Nagpur white x Sattur	80	300	—	Open, moderate branching 6–8 loculed, cylindrical big sized capsules dark brown to black seed Suitable for cold weather 52% oil.	
TMV 3	1943	South Arcot variety x Malabar Variety	80	350	700	Bushy with profuse branching 4 loculed, dark brown to black seed 51% oil.	
KRR1	1967	Pureline selection From Karur Paramathy	120	450	—	Bushy with profuse branching, 4 loculed, brown seeds, oil 52%.	
KRR 2	1970	Karur local x Bombay white	110		_	Bushy with profuse branching4 loculed, oil 52%, dull white seeds.	
TMV 4	1977	Pureline selection Sattur (local)	85		700	Bushy with profuse branching, 4 loculed, brown seeds, 51% oil.	
TMV 5	1978	Pureline selection from Srivaikuntam local	80	400	750	Erect with moderate branching, 4 loculed, brown seeds, 51% oil.	
TMV 6	1980	Selection from Andhra local	85	_	750	Erect with moderate branching, 4 loculed, brown seeds, 54% oil.	
CO 1	1983	(TMV 3 x Si 1878) x Si 1878	85–90	600	900	Bushy plant, 4 loculed, black warty seeds, 51% oil content Notification No: 596(E)/13.08.1984	
Paiyur 1	1990	Si2511 x Si 2314	90		644	Resistant to powdery mildew, 4 loculed, bushy suited for irrigated condition, black seed, oil 50	
SVPR 1	1992	Selection from "Western ghat white"	80		800	White seeded, 4 loculed, high yielding variety suitable for irrigated tracts of Tamil Nadu, oil content 50%	
VRI 1	1995	Pureline selection from Tirukkattupalli local	75	_	700	Short duration crop, 4 loculed, suited specially for rice fallows, oil content 51%	
VRISV2	2005	US9003 x TMV6	80–85	706	726	Moderately resistant to shoot webber, 4 loculed, high oil content (51.9%)	
TMV (Sv) 7	2009	Si 250 x ES 22	85–90	750	820	High yield, 4 loculed, tolerant to root rot disease, Lustrous brown testa, oil content 50%	
VRI 3	2017	SVPR 1 x TKG 87	75–80	995	1055	Moderately resistant to phyllody and root rot diseases White seed 50.1 per cent oil content.	
ource: https:/	//sites.google.co	m/a/tnau.ac.in/cpbg/oilseeds/sesame-	-varieties				
<b>ble 4.</b> etails of sesa	ame varieties	released from TNAU, Coimbatore,	Tamil Nadu,	India.			

to exploit the hybrid vigour needs identification of diverse germplasm both by morphological and genetic markers. A diversity analysis was performed on the germplasm of sesame to evaluate yield and associated traits. Fourteen qualitative and six biometric traits were evaluated phenotypically on 270 sesame lines from TNAU using the NBPGR descriptors.

The morphologic characters stem and leaf hairiness, branching, plant growth habit and type exhibited variations. Whereas the six quantitative characters had significant diversity similar to the finding of Abate et al. [20]. The flower colours were white with pink shade suggesting its origin and several of them were shattering type which is prevalent in Indian germplasm [21]. Parental selection was performed based on their mean performance for traits such as days to 50% flowering, days to maturity and plant height. For characters, primary branches, capsules and seed yield per plant grand mean + SE was good. Early maturing phenotypes are the choice in recent years to evade pest and diseases and to accommodate more crops per year. Short duration genotypes, RSS-379-2, SI-3296, GUN-3-NL-1, G-10, NIC-8317, NIC-8283, IC-131651 and SI-2143 with 93–95 days for maturity were selected since they matured early and performed well than the control TMV7. Similarly dwarf genotypes which resist to lodging were preferred. The following genotypes recorded <85 cm height SI-2144, SI-440, JLSC-96, SI-1712, SI-345, SI-395 and SI-2143. Highest number of branches of 10 was observed in ORM-7, SI-395, KMS-4343, SI-533, NIC-1610 and SI-2143. High number of primary branches, high capsule number and yield were recorded together in genotypes, SI-395, NIC-1610 and SI-2143. Yield obtained was 23.09 g, 21.5 g and 21 g in SI-395, NIC-1610 and SI-2143 respectively. SVPR variety recorded a slightly lower yield of 18.8 g. A positive correlation of primary branches, number of capsules and yield was observed similar to the results reported by Ozcinar et al. [22]. Genotypes SI-395 and SI-2143 were high yielding and dwarf genotypes with a negative direct effect between height and yield as indicated by Agarwal et al. [23, 24]. Variability measures in terms of genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) help us to evaluate the contribution of genetic and environmental factors. The phenotypic character, days to 50% flowering showed a moderate GCV and PCV as reported by Iqbal et al. [1], Parmeshwarappa et al. [25] and Sumathi and Muralidharan [26]. However, the same trait recorded highest heritability and genetic advance and similar observations were also made by Kiruthika et al. [27]. In contrast, the days to maturity recorded low GCV and PCV with high heritability and low genetic advance as that of Hika et al. [28] and Sourey et al. [29] respectively. Plant height exhibited moderate GCV and PCV with high heritability and GA in this study as observed by earlier studies by Parmeshwarappa et al. [25].

Genetic divergence by Mahalanobis D2 analysis [27] in these 270-sesame clustered them into 16 groups and 10 of them are single monogenotypic suggesting the diverse nature of germplasm. Genotypes of different geographical origin also have clustered together similar observation recorded by Tripathi et al. [30]. Analysis of  $\alpha$ -linolenic acid, sesamin and sesamol content from Tamil Nadu sesame germplasm collection was reported by Chellamuthu et al. [31] enabling the choice of varieties for medicinal purposes.

#### 5. Sesame Indian Scenario

Varietal Development: Eighty-three varieties have been developed for different agroecological situations. Seed yield of these varieties range from 800 to 1000 kg/ ha, days to maturity 80–95 and oil content 48–52%. State wise recommended varieties are shown in **Table 5**.

State	Varieties
Gujarat	Gujarat Til-1, Gujarat Til-2, Gujarat Til-10, Gujarat Til-3
Madhya Pradesh	TKG-21, TKG-22, TKG-55, JTS-8, PKDS-11, PKDS-8, PKDS-12, TKG-306, TKG-308
Chattisgarh	TKG-21, TKG-22, Uma, RT-54, TKG-55, JTS-8
Rajasthan	RT-46, RT-54, RT-103, RT-125, RT-127, RT-346, RT- 351,
Maharashtra	Phule Til-1, Tapi, Padma, AKT-64, AKT-101, PKV-NT-11,JLT-408
Uttar Pradesh	T-12, T-13, T-78, Sekhar, Pragati, Tarun
Tamil Nadu	TMV-3, TMV-4, TMV-5, TMV-6, CO-1, TSS-6, Paiyur-1, VRI-1,
West Bengal	Savitri, Rama
Orissa	Uma, Usha, Nirmala, Prachi, Amrit
Andhra Pradesh	Madhavi, Rajeshwari, Varaha, Gautama, Swetha, Chandana, Hima, Sarada
Kerala	Kayamkulam-1, Thilak, Thilathara, Thilarani
Karnataka	DS-1, DSS-9
Punjab	Punjab Til-1, TC-25, TC-289
Bihar	B-67, Krishna
Haryana	Haryana Til-1
Himachal Pradesh	Brijeshwari

Table 5.

List of sesame varieties available in India.

#### 6. Molecular markers of sesame

Molecular marker technologies have been exploited for sesame genotyping and breeding. The first class of molecular markers including random amplified polymorphic DNA (RAPD) and amplified fragment length polymorphism (AFLP) were planned and employed for genetic diversity reports [32]. The second class of markers include simple sequence repeats (SSR) types such as Inter-Simple Sequence Repeats (ISSR), Expressed Sequence Tags-SSR(EST-SSR), cDNA-SSR, Genome sequence – SSR, Chloroplast-SSR [33–36]. There are more than 7000 validated and 1, 00,000 non-validated SSR markers were listed and acquired for sesame research. These markers were used for genetic and association mapping, molecular breeding and genetic diversity studies of sesame. Recently, next generation sequencing (NGS) technology, the third class of molecular markers were initiated. SNPs are very valuable genetic markers than other conventional markers because they are most plentiful and steady form of genetic variation in genome. Restriction site-associated DNA sequencing (RADseq), Specific length amplified fragment sequencing (SLAF-seq), RNA-seq, Whole genome sequencing (WGS), Genotyping by sequencing (GBS), insertions/deletions (Indels) has also been stated in sesame [37–40].

#### 6.1 Genome Resources

The nuclear genome of sesame containing of 54.5 Gb of high quality data from the cultivar "Zhongzhi No.13" through Illumina sequencing [41]. The draft genome including of 27,148 genes dispersed on 16 linkage groups (LG) with 274 Mb of size. This genome consist of contig N50 of 52.2Kb and a scaffold with 2.1 Mb has been recently improved to reach 13 pseudo chromosomes, 94.3% of the

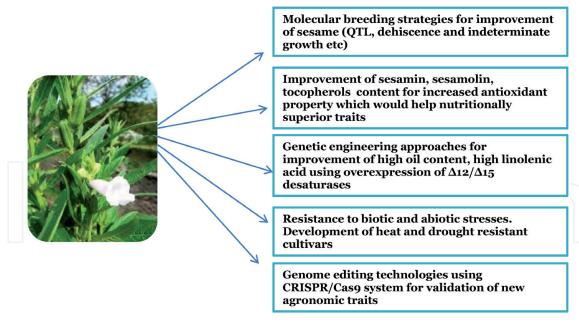
estimated genome size and 97.2% of predicted gene models [42]. Another genome sequencing project was started parallel under Sesame Genome Working Group (SGWG). They assembled the variety "Yuwhi 11" a genome size of 293.7 Mb out of 354 Mb estimated in sesame and predicted the function of 23,713 genes. Recently, two new genome sequences from landraces "Baizhima" and "Mishuozhima" have been announced [43]. In addition, a team from National Bureau of Plant Genetic resources, India ensued in the genome sequencing of India variety "Swetha". Nearly 1000 sesame accessions were re-sequenced to provide genome-wide information [44–46]. Gene cloning and molecular breeding, genome wide association studies (GWAS), genome variation and evolution studies are possible nowadays [47, 48]. Novel breeding methods like genomic selection (GS) could be performed for crop improvement in sesame [49].

#### 6.2 Transcriptome Assembly

The first transcriptome summarizing began with 3328 ESTs obtained from cDNA library of 5–25 days old immature sesame seeds. These reports bring out the metabolic pathways implicated in lignan biosynthesis including sesamin and sesamolin [50]. On the other hand, sesame productivity is severely influenced by different biotic and abiotic stresses; studies have been pointed to find out some potential genes to convey stress tolerance in root tissues to waterlogging stress in sesame [51]. Another significant abiotic stress spoiling sesame productivity is drought stress, for that gene expression changes were examined in two sesame genotypes (tolerant and sensitive) through Illumina Hiseq 4000 sequencing platform [52]. RNA-seq study was applied for resistant and susceptible sesame cultivars inoculated with *Fusarium oxysporum* to shed light on molecular mechanism of sesame resistance to Fusarium wilt. It is one of the major diseases in sesame accounting to a yield loss of 15–30% [40].

#### 7. Conventional Breeding methods

Conventional breeding approaches mainly involve the existence of wild relatives, elite cultivars, and landraces to enable the assortment of superior lines for quality enhancement (Figure 3). Genetic diversity studies can be carried out by several methods such as biochemical, morphological and molecular markers. Genetic differences examined using morphological markers is also an essential tool among sesame genotypes. A few investigations dependent on morphological markers have shown the presence of high genetic diversity in sesame populations [21, 49]. The high level of genetic diversity prevalent among the 58 Indian collections is probably indicative of the nativity of this crop species [50]. As part of broadening the genetic base of sesame (Sesamum indicum L.) in India through germplasm enhancement, National Bureau of Plant Genetic Resources has made initiatives [51]. A selection was made of 24 of the most diverse and unadapted parental lines, including one accession of the wild species S. mulayanum, and these were intercrossed in various combinations to maximize genetic diversity and to develop locally adapted pools of genetic resources. Genetic analyses on sesame crosses have shown the presence of additive, dominance and epistatic gene interactions for yield and its components. Molecular marker techniques such as random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), simple sequence repeats (SSR), inter simple sequence repeats (ISSR) are extensively used for genetic diversity studies. AFLP markers showed a low level of genetic diversity (0.14–0.21) among 36 sesame germplasm collections [53]. Characterization of Indian sesame



**Figure 3.** *Future perspectives for improvement of sesame.* 

varieties was performed using SSR and ISSR markers. Results indicated that the varieties were clustered independently of their geographical locations [52]. Single nucleotide polymorphism (SNP) plays an imperative role in genotyping and these are the most abundant molecular markers which are widely dispersed throughout the genomes with a variable distribution among species [54]. Expressed sequence tags (EST) from *Sesamum indicum* and *Arabidopsis thaliana* resulted in similar and different gene expression profiles during seed development and 41,248 ESTs for developing seeds of sesame cDNA library has been generated [55].

#### 7.1 Genetic Engineering approaches for sesame improvement

For oil quality improvement fatty acid composition and enzymes involved in metabolism are very important, so for our gain like to increase or decrease the quantity of particular fatty acid through genetic engineering we can reduce or increase the expression of endogenous enzymes by various means. It proved that genes for membrane-bound fatty acid modifying enzymes not only from plants but also from bacterial, animal, yeast have been shown to function in transgenic plants. The enzymes such as fatty acid synthase, thioesterases, elongases, desaturases, stearoyl-ACP desaturase,  $\Delta$ 12-desaturase,  $\Delta$ 15- Desaturase, acyltransferases and Hydroxylases are important in fatty acid manipulation. Suppression of the oleate  $\Delta$ 12-desaturase gene (which normally converts 18:1 to 18:2) in soybean, sunflower, cotton and canola has resulted in the production of oils with a high oleic acid content, which have greater oxidative stability and improved performance in high temperature cooking application [56]. Yadav et al. [57] reported for the first-time successful recovery of fertile transgenic plants of sesame with transformation frequency of 1.01%. From cotyledon explants inoculated with *A. tumefaciens* carrying a binary vector pCAMBIA 2301 that contains a neomycin phosphotransferase gene (nptII) and a  $\beta$ -glucuronidase (GUS) gene (uidA) interrupted with an intron. The most efficient gene transformation protocol using de-embryonated cotyledon of sesame (cultivar VRI-1) was reported by Chowdhury et al. [58]. Shoot regeneration from cotyledons was reported recently in Indian sesame genotypes [59]. Enhancement of omega 3 fatty acid content of sesame using *Fusarium moniliforme*  $\Delta 12/\Delta 15$  bifunctional

desaturase gene through genetic engineering approach (Unpublished data).Yeast is an excellent model for lipid biosynthesis related studies. Functional characterization of DGAT and PDAT genes of sesame using yeast H1246 oil synthesis deficient mutant and their oil accumulation were analysed [60]. Co-expression analysis of DGAT1 and PDAT1 genes with omega 3 desaturase genes were characterized in the yeast expression system for oil quality enhancement (Unpublished data). These are some of the strategies for improvement of sesame through genetic engineering.

#### 7.2 CRISPR/Cas9 Applications in Plants

The accumulation of complete genome sequencing data has enabled the targeted genome editing strategy using CRISPR/Cas9 system for oil improvement in many oil seed crops. In allotetraploid *Brassica napus*, the efficiency of the CRISPR/Cas9 mutation was examined for 12 paralogous genes, BnaA9.RGA, BnaC9.RGA, BnaA6. RGA, BnaC7.RGA, BnaA2.DA2.1, BnaA2.DA2.2, BnaC6.DA2, BnaC5.DA1, BnaA6. DA1, BnaA9.FUL, BnaC2.FUL, and BnaC7.FUL. They determined the specificity and heritability of the CRISPR/Cas9 mutants. The result showed that the targeted mutation in the T0 generation was stably inherited into the progeny and the mutation frequency ranged from 27.6% to 96.6% and no off-target mutation was identified. FAD2-2 is a desaturase gene which uses the substrate oleic acid and converts it to linoleic acid. In soybean this gene was mutated using CRISPR-Cas9 system in order to improve the seed oil. The result showed that the mutation efficiency was 21% and the content of oleic acid was increased to 65.58% from 17.34% and the level of linoleic acid was reduced to 16.08% from 59.54% [61]. Camelina sativa is considered as one of the most important sources of cooking and industrial oil. The total oil content of Camelina is found to be 32-40%. Increase in oleic acid and decrease in poly unsaturated fatty acids such as linolenic acid and linoleic acid contents can provide better suited oil for many industrial purposes and mainly biofuels. A research group in United States attempted the CRISPR/Cas9 gene-editing system to increase the oleic acid content and decrease both linolenic acid and linoleic acid content by knocking out the FAD2 gene in Camelina. The allohexaploid Camelina genome contains a total of six FAD2 genes. They designed sgRNA constructs to knockout both allelic copies of FAD2 genes in Camelina. The result showed that seeds had over 50% vs. 16% oleic acid and less than 15% polyunsaturated fatty acids in T4 generation. This work provided the proof of concept that the FAD2 genes in oil seed plants can be successfully edited using the CRISPR/Cas9 system to yield plants capable of producing commercially valuable oils [62]. In bread wheat Triticum aestivum, two genes, inositol oxygenase (inox) and phytoene desaturase (pds), were targeted using CRISPR/Cas9 gene-editing system. Two sgRNA constructs were used to target each gene. The efficient production of insertions and deletions were observed in wheat cell suspension cultures with each of two sgRNA constructs. When the two sgRNA genes were placed together in a single expression cassette, the gene fragment between the two target sites was deleted. This study demonstrated that creation of gene knockout and gene fragment deletions in hexaploid wheat were also possible using CRISPR/Cas9 [63]. In rice, FAD2-1 gene was mutated using CRISPR/Cas9 to produce high oleic acid and low linoleic acid in bran oil. The results showed that the content of oleic acid was increased twice the wild type which was80% vs. 32% [64]. There are many constraints for molecular and biotechnological approaches in developing elite varieties in sesame. Besides, seldom available transgenic plants and approaches are not well received by public. These are some of the strategies will help the researchers to generate superior sesame traits through CRISPR/Cas9 based targeted editing and mutation breeding.

#### 8. Prospects of Sesame improvement

- 1. Development of large number of varieties suitable for our agro climatic conditions.
- 2. Improvement of value added products in sesame will enhance the economic value in the world market.
- 3. Development of sesame plants with high lignans, tocopherol, and omega 3 fatty acid content will help to reduce the risk of cancer, diabetes, cardiovascular problems.
- 4. Conventional breeding methods, advancement in next generation sequencing will help to develop tools for genetic improvement of sesame.
- 5. Enhancement of oil quality through CRISPR/Cas9 to generate superior varieties in sesame.

#### 9. Conclusion

Nowadays vegetable oil demand was increasing globally and oil consumption was expected to be doubled in 2030. There is lot of room to improve the sesame varieties for yield, oil content and quality. Besides oil, other lignans such as sesamin and sesamolin contents in Indian varieties add unique flavor and value to the sesame oil. Sesame is used as a promising target oilseed for biofuel applications, pharmaceutical etc.

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#### **Conflict of interest**

The authors declare no conflict of interest.

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