We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,300

Open access books available

172,000

International authors and editors

190M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

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

Muthulakshmi Chellamuthu, Selvi Subramanian
and Manonmani Swaminathan

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, sesamol, 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. occidentale*, *S. radiatum* Schm & Thonn. has 2n = 64, *S. angolens* and *S. prostratum* 2n = 32, *S. laciniatum* 2n = 28. Nowadays, *Sesamum indicum* is cultivated mainly however, a few other species: *S. angustifolium*, *S. calycinum*, 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	Quantity		References
	Sesame seed (mg g ⁻¹ seed)	Sesame oil (mg g ⁻¹)	
Palmitic acid (16:1)	9.4%	14.45%	Hemalatha and Ghafoorunissa [7]
Oleic acid (18:1)	39.1%	50.54%	
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. Sesamol 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⁻¹. 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 ('000 ha)	Yield (kg ha ⁻¹)	Production ('000 MTha ⁻¹)	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

Source: Food and Agriculture Organization Statistical Databases (FAOSTAT), 2020.

Table 2.
World status of sesame area, yield and production in 2018.

Year	Seed		Oil and fractions		Total (value)
	Export Quantity (tonnes)	Rupees (crores)	Export Quantity (tonnes)	Rupees (crores)	
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

Source: Directorate General of Commercial Intelligence and Statistics, (Kolkata).

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/ Variety	Year of release	Parentage	Duration Days	Yield (kg/ha)		Special features
				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 seeds. 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 branching 4 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.

Source: <https://sites.google.com/a/tnau.ac.in/cpbg/oilseeds/sesame-varieties>

Table 4.
Details of sesame 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 α -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, stearyl-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 β -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 was 80% 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 sesamol 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.

Acknowledgements

The authors thank Department of Biotechnology, PSG College of Technology for providing infrastructural facilities and we thank Tamil Nadu Agricultural University for providing sesame seed varieties for our research purpose.

Conflict of interest

The authors declare no conflict of interest.

IntechOpen

Author details

Muthulakshmi Chellamuthu¹, Selvi Subramanian^{1*} and Manonmani Swaminathan²

¹ Department of Biotechnology, PSG College of Technology,
Coimbatore, Tamil Nadu, India

² Department of Plant Genetic Resources, Tamil Nadu Agricultural University,
Coimbatore, Tamil Nadu, India

*Address all correspondence to: selvi.bio@psgtech.ac.in

IntechOpen

© 2020 The Author(s). Licensee IntechOpen. This chapter is distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/3.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

References

- [1] Iqbal A, Pati PK, Akhtar R, Begum T, Dasgupta T. Diversity in Sesame Accessions. *International Journal of Agriculture, Environment and Biotechnology*. 2018;**11**(5):725-731
- [2] Bedigian D, Van der Maesen LJ. Slimy leaves and oily seeds: Distribution and use of *Sesamum* spp. and *Ceratotheca sesamoides* (Pedaliaceae) in Africa. In *Plant Resources of Tropical Africa: Proceedings of the First PROTA International Workshop, Nairobi, Kenya, 23-25 september 2002/GH Schmelzer and EA Omino 2003* (pp. 271-273).
- [3] Weiss EA. *Oilseed crops*. Blackwell Science; 2000
- [4] Ashri A, Singh RJ. Sesame (*Sesamum indicum* L.). *Genetics resources chromosome Engineering and crop improvement, Oilseed crop*. 2007;**4**:231-89.
- [5] Pursglove JW. *Tropical crops: Dicotyledons*. Longman Group, London, Third Edition, London, 1977: 719p.
- [6] Bedigian D. History and lore of sesame in Southwest Asia. *Economic botany*. 2004 Sep 1;**58**(3):329-53.
- [7] Hemalatha S. Lignans and tocopherols in Indian sesame cultivars. *Journal of the American Oil Chemists' Society*. 2004 May 1;**81**(5):467. DOI: 10.1007/s11746-004-0924-5
- [8] Uzun B, Arslan Ç, Furat Ş. Variation in fatty acid compositions, oil content and oil yield in a germplasm collection of sesame (*Sesamum indicum* L.). *Journal of the American Oil Chemists' Society*. 2008 Dec 1;**85**(12):1135-1142. DOI: 10.1007/s11746-008-1304-0
- [9] Shahidi F, Liyana-Pathirana CM, Wall DS. Antioxidant activity of white and black sesame seeds and their hull fractions. *Food Chemistry*. 2006 Jan 1;**99**(3):478-83.doi: 10.1016/j.foodchem.2005.08.009
- [10] Yokota T, Matsuzaki Y, Koyama M, Hitomi T, Kawanaka M, Enoki-Konishi M, et al. Sesamin, a lignan of sesame, down-regulates cyclin D1 protein expression in human tumor cells. *Cancer science*. 2007 Sep;**98**(9):1447-1453. DOI: 10.1111/j.1349-7006.2007.00560
- [11] Hsu DZ, Su SB, Chien SP, Chiang PJ, Li YH, Lo YJ, et al. Effect of sesame oil on oxidative-stress-associated renal injury in endotoxemic rats: involvement of nitric oxide and proinflammatory cytokines. *Shock*. 2005 Sep 1;**24**(3):276-280. DOI: 10.1097/01.shk.0000172366.73881.c7
- [12] Penalvo JL, Heinonen SM, Aura AM, Adlercreutz H. Dietary sesamin is converted to enterolactone in humans. *The Journal of nutrition*. 2005 May 1;**135**(5):1056-1062 <https://doi.org/10.1093/jn/135.5.1056>
- [13] Wei X, Liu K, Zhang Y, Feng Q, Wang L, Zhao Y, Li D, Zhao Q, Zhu X, Zhu X, Li W. Genetic discovery for oil production and quality in sesame. *Nature communications*. 2015 Oct 19;**6**:8609. DOI: 10.1038/ncomms9609 |.
- [14] Food and Agriculture Organization (FAO). FAOSTAT statistical database of the United Nation Food and Agriculture Organization (FAO) statistical division 2019. Available online: <http://faostat.fao.org/>
- [15] Morris JB. Food, industrial, nutraceutical, and pharmaceutical uses of sesame genetic resources. *Trends in new crops and new uses*. 2002:153-156
- [16] Rahman A, Bhattarai S, Akbar D, Thomson M, Trotter T, Timilsina S.

Australia C. MARKET ANALYSIS OF SESAME SEED.

[17] KHARIF-2019 SURVEY OF SESAME CROP. http://www.iopepc.org/misc/2019_20

[18] Directorate General of Commercial Intelligence and Statistics (Kolkata), Ministry of Commerce and Industry, GOI.

[19] Jawahar Lal, J., H. H. Kumaraswamy, A. R. G. Ranganatha. 2015. Quality seed production technology in sesame. Technical report, <https://www.researchgate.net/publication/311776080>

[20] Abate M, Mekbib F, Ayana A, Nigussie M. Genetic variability and association of traits in mid-altitude sesame (*Sesamum indicum* L.) germplasm of Ethiopia. Journal of Experimental Agriculture International. 2015 Aug 4:1-4. <https://doi.org/10.9734/AJEA/2015/18483>

[21] Akbar F, Rabbani MA, Shinwari ZK, Khan SJ. Genetic divergence in sesame (*Sesamum indicum* L.) landraces based on qualitative and quantitative traits. Pak. J. Bot. 2011 Dec 1;43(6):2737-44.

[22] Ozcinar Bilmez A, Sogut T. Analysis of sesame (*Sesamum indicum* L.) accessions collected from different parts of Turkey based on qualitative and quantitative traits. International Journal of Crop Breeding and Genetics. 2017;3(1):45-51

[23] Agrawal MM, Singh S, Macwan S, Sasidharan N, Wawge MN. Genetic variability analyses for economic traits in sesame (*Sesamum indicum* L.) germplasm. Electronic Journal of Plant Breeding. 2018;9(3):1221-1225. DOI: 10.5958/0975-928X.2018.00150.3

[24] Agrawal MM, Singh S, Macwan S, Sasidharan N, Wawge MN. Genetic variability analyses for economic

traits in sesame (*Sesamum indicum* L.) germplasm. Electronic Journal of Plant Breeding. 2018;9(3):1221-1225. DOI: 10.5958/0975-928X.2018.00150.3

[25] Parameshwarappa SG, Palakshappa MG, Salimath PM, Parameshwarappa KG. Studies on genetic variability and character association in germplasm collection of sesame (*Sesamum indicum* L.). Karnataka Journal of Agricultural Sciences. 2009;22(2):252-254

[26] Sumathi P, Muralidharan V. Analysis of genetic variability, association and path analysis in the hybrids of sesame (*Sesamum indicum* L.). Tropical Agricultural Research and Extension. 2011 Jun;22:13(3)

[27] Kiruthika S, Narayanan SL, Parameswari C, Mini ML, Arunachalam P. Genetic variability studies for yield and yield components in sesame (*Sesamum indicum* L.). Electronic Journal of Plant Breeding. 2018;9(4):1529-37.

[28] Hika G, Geleta N, Jaleta Z. Genetic variability, heritability and genetic advance for the phenotypic traits in sesame (*Sesamum indicum* L.) populations from Ethiopia. Science, Technology and Arts Research Journal. 2015;4(1):20-26

[29] El Soury HF, El Bashir G, Ginaro MK. Phenotypic and genotypic coefficients of variation and other growth attributes in sesame genotype under rain-fed conditions. Advances in Agr. and Agr. Sci. 2016;2:079-084

[30] Tripathi AN, Bisen RA, Ahirwal RP, Paroha S, Sahu R, Ranganatha AR. Study on genetic divergence in sesame (*Sesamum indicum* L.) germplasm based on morphological and quality traits. The bioscan. 2013 Oct 29;8(4):1387-1391

[31] Chellamuthu M, Sekar P, Subramanian S. Evaluation of sesame

(*Sesamum indicum* L.) germplasm collection of Tamil Nadu for-linolenic acid, sesamin and sesamol content. African Journal of Biotechnology. 2017 Jun 7;16(23):1308-1313. <https://doi.org/10.5897/AJB2016.15688>

[32] Sehr EM, Okello-Anyanga W, Hasel-Hohl K, Burg A, Gaubitzer S, Rubaihayo PR, Okori P, Vollmann J, Gibson P, Fluch S. Assessment of genetic diversity amongst Ugandan sesame (*Sesamum indicum* L.) landraces based on agromorphological traits and genetic markers. Journal of crop science and biotechnology. 2016 Mar 1;19(1):117-24. J. Crop Sci. Biotechnol. 19, 117-129. doi: 10.1007/s12892-015-0105-x.

[33] Wu K, Liu H, Yang M, Tao Y, Ma H, Wu W, Zuo Y, Zhao Y. High-density genetic map construction and QTLs analysis of grain yield-related traits in Sesame (*Sesamum indicum* L.) based on RAD-Seq technology. BMC plant biology. 2014 Dec 1;14(1):274. doi: 10.1186/s12870-014-0274-7

[34] Yu J, Dossa K, Wang L, Zhang Y, Wei X, Liao B, et al. PMDBase: a database for studying microsatellite DNA and marker development in plants. Nucleic Acids Research. 2017 Jan 4;45(D1):D1046-D1053. DOI: 10.1093/nar/gkw906

[35] Wang L, Xia Q, Zhang Y, Zhu X, Zhu X, Li D, et al. Updated sesame genome assembly and fine mapping of plant height and seed coat color QTLs using a new high-density genetic map. BMC genomics. 2016 Dec 1;17(1):31. DOI: 10.1186/s12864-015-2316-4

[36] Zhang H, Miao H, Wang L, Qu L, Liu H, Wang Q, et al. Genome sequencing of the important oilseed crop *Sesamum indicum* L. Genome biology. 2013 Jan 1;14(1):401. DOI: 10.1186/gb-2013-14-1-401

[37] Wei W, Zhang Y, Lü H, Li D, Wang L, Zhang X. Association Analysis

for Quality Traits in a Diverse Panel of Chinese Sesame (*Sesamum indicum* L.) Germplasm. Journal of integrative plant biology. 2013 Aug;55(8):745-758. DOI: 10.1111/jipb.1204

[38] Uncu AO, Frary A, Karlovsky P, Doganlar S. High-throughput single nucleotide polymorphism (SNP) identification and mapping in the sesame (*Sesamum indicum* L.) genome with genotyping by sequencing (GBS) analysis. Molecular breeding. 2016 Dec 1;36(12):173. doi: 10.1007/s11032-016-0604-6

[39] Wang L, Yu S, Tong C, Zhao Y, Liu Y, Song C, et al. Genome sequencing of the high oil crop sesame provides insight into oil biosynthesis. Genome biology. 2014 Feb;15(2):1-3. DOI: 10.1186/gb2014-15-2-r39

[40] Wei X, Zhu X, Yu J, Wang L, Zhang Y, Li D, Zhou R, Zhang X. Identification of sesame genomic variations from genome comparison of landrace and variety. Frontiers in plant science. 2016 Aug 3;7:1169. doi: 10.3389/fpls.2016.01169

[41] Zhang H, Miao H, Li C, Wei L, Duan Y, Ma Q, et al. Ultra-dense SNP genetic map construction and identification of SiDt gene controlling the determinate growth habit in *Sesamum indicum* L. Scientific reports. 2016 Aug 16;6:31556. DOI: 10.1038/srep31556

[42] Uncu AÖ, Gultekin V, Allmer J, Frary A, Doganlar S. Genomic simple sequence repeat markers reveal patterns of genetic relatedness and diversity in sesame. The Plant Genome. 2015 Jul;8(2):1-2. DOI: 10.3835/plantgenome2014.11.0087

[43] Wei X, Liu K, Zhang Y, Feng Q, Wang L, Zhao Y, et al. Genetic discovery for oil production and quality in sesame. Nature communications. 2015 Oct 19;6:8609. DOI: 10.1038/ncomms9609

- [44] Dossa K, Wei X, Zhang Y, Fonceka D, Yang W, Diouf D, et al. Analysis of genetic diversity and population structure of sesame accessions from Africa and Asia as major centers of its cultivation. *Genes*. 2016 Apr;7(4):14. DOI: 10.3390/genes7040014
- [45] Yu J, Wang L, Guo H, Liao B, King G, Zhang X. Genome evolutionary dynamics followed by diversifying selection explains the complexity of the *Sesamum indicum* genome. *BMC genomics*. 2017 Dec;18(1):1-4. DOI: 10.1186/s12864-017-3599-4
- [46] Dossa K, Li D, Wang L, Zheng X, Yu J, Wei X, Fonceka D, Diouf D, Liao B, Cisse N, Zhang X. Dynamic transcriptome landscape of sesame (*Sesamum indicum* L.) under progressive drought and after rewatering. *Genomics data*. 2017 Mar 1;11:122-124. doi: 10.1016/j.gdata.2017.01.003.
- [47] Suh MC, Kim MJ, Hur CG, Bae JM, Park YI, Chung CH, et al. Comparative analysis of expressed sequence tags from *Sesamum indicum* and *Arabidopsis thaliana* developing seeds. *Plant molecular biology*. 2003 Aug 1;52(6):1107-1123. DOI: 10.1023/B:PLAN.0000004304.22770.e9
- [48] Wang L, Zhang Y, Li P, Wang X, Zhang W, Wei W, Zhang X. HPLC analysis of seed sesamin and sesamolin variation in a sesame germplasm collection in China. *Journal of the American Oil Chemists' Society*. 2012 Jun 1;89(6):1011-20. doi: 10.1007/s11746011-2005-7
- [49] Arriel NH, Di Mauro AO, Arriel EF, Unêda-Trevisoli SH, Costa MM, Bárbaro IM, et al. Genetic divergence in sesame based on morphological and agronomic traits. *Crop Breeding and Applied Biotechnology*. 2007 Sep;1:253-261 <http://dx.doi.org/10.12702/1984-7033.v07n03a04>
- [50] Bhat KV, Babrekar PP, Lakhanpaul S. Study of genetic diversity in Indian and exotic sesame (*Sesamum indicum* L.) germplasm using random amplified polymorphic DNA (RAPD) markers. *Euphytica*. 1999 Oct 1;110(1):21-34
- [51] Bisht IS, Mahajan RK, Loknathan TR, Agrawal RC. Diversity in Indian sesame collection and stratification of germplasm accessions in different diversity groups. *Genetic Resources and Crop Evolution*. 1998 Aug 1;45(4):325-335 <http://dx.doi.org/10.1023/A:1008652420477>
- [52] Kumar V, Sharma SN. Comparative potential of phenotypic, ISSR and SSR markers for characterization of sesame (*Sesamum indicum* L.) varieties from India. *Journal of Crop Science and Biotechnology*. 2011 Sep 1;14(3):163-171 <http://dx.doi.org/10.1007/s12892-010-0102-z>
- [53] Laurentin HE, Karlovsky P. Genetic relationship and diversity in a sesame (*Sesamum indicum* L.) germplasm collection using amplified fragment length polymorphism (AFLP). *BMC genetics*. 2006 Dec 1;7(1):10. <http://dx.doi.org/10.1186/1471-2156-7-10>
- [54] Gupta PK, Varshney RK, Sharma PC, Ramesh B. Molecular markers and their applications in wheat breeding. *Plant breeding*. 1999 Oct;118(5):369-390 <http://dx.doi.org/10.1046/j.1439-0523.1999.00401.x>
- [55] Ke T, Dong C, Mao H, Zhao Y, Chen H, Liu H, et al. Analysis of expression sequence tags from a full-length-enriched cDNA library of developing sesame seeds (*Sesamum indicum*). *BMC plant biology*. 2011 Dec 1;11(1):180 <http://dx.doi.org/10.1186/1471-2229-11-180>
- [56] Metzger JO, Bornscheuer U. Lipids as renewable resources: current state of chemical and biotechnological

conversion and diversification. *Applied microbiology and biotechnology*. 2006 Jun 1;**71**(1):13-22

[57] Yadav M, Chaudhary D, Sainger M, Jaiwal PK. *Agrobacterium tumefaciens*-mediated genetic transformation of sesame (*Sesamum indicum* L.). *Plant Cell, Tissue and Organ Culture (PCTOC)*. 2010 Dec 1;**103**(3):377-386. doi:10.1007/s11240-010-9791-8

[58] Chowdhury S, Basu A, Kundu S. A new high-frequency *Agrobacterium*-mediated transformation technique for *Sesamum indicum* L. using de-embryonated cotyledon as explant. *Protoplasma*. 2014 Sep 1;**251**(5):1175-1190 <https://doi.org/10.1007/s00709-014-0625-0>

[59] Zimik M, Arumugam N. Induction of shoot regeneration in cotyledon explants of the oilseed crop *Sesamum indicum* L. *Journal of Genetic Engineering and Biotechnology*. 2017 Dec 1;**15**(2):303-308 <https://doi.org/10.1016/j.jgeb.2017.07.006>

[60] Chellamuthu M, Kumaresan K, Subramanian S, Muthumanickam H. Functional Analysis of Sesame Diacylglycerol Acyltransferase and Phospholipid: Diacylglycerol Acyltransferase Genes Using In Silico and In Vitro Approaches. *Plant Molecular Biology Reporter*. 2019 Jun 15;**37**(3):146-156 <https://doi.org/10.1007/s11105-019-01144-7>

[61] al Amin N, Ahmad N, Wu N, Pu X, Ma T, Du Y, Bo X, Wang N, Sharif R, Wang P. CRISPR-Cas9 mediated targeted disruption of FAD2-2 microsomal omega-6 desaturase in soybean (*Glycine max*. L). *BMC biotechnology*. 2019 Dec;**19**(1):1-0. <https://doi.org/10.1186/s12896-019-0501-2>

[62] Jiang WZ, Henry IM, Lynagh PG, Comai L, Cahoon EB, Weeks DP. Significant enhancement of fatty acid composition in seeds of the

allohexaploid, *Camelina sativa*, using CRISPR/Cas9 gene editing. *Plant biotechnology journal*. 2017 May;**15**(5):648-657 <https://doi.org/10.1111/pbi.12663>

[63] Upadhyay SK, Kumar J, Alok A, Tuli R. RNA-guided genome editing for target gene mutations in wheat. *G3: Genes, Genomes, Genetics*. 2013 Dec 1;**3**(12):2233-2238 <https://doi.org/10.1534/g3.113.008847>

[64] Abe K, Araki E, Suzuki Y, Toki S, Saika H. Production of high oleic/low linoleic rice by genome editing. *Plant Physiology and Biochemistry*. 2018 Oct 1;**131**:58-62 <https://doi.org/10.1016/j.plaphy.2018.04.033>