



ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2021; 10(10): 1422-1426
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www.thepharmajournal.com
Received: 17-07-2021
Accepted: 29-09-2021

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Review on genetics and breeding of sunflower (*Helianthus annuus*)

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Abstract

Sunflower (*Helianthus annuus* L.) is the world's second-most-important oilseed crop after soybean. It belongs to the Asteraceae/ Compositae family and has a high amount of unsaturated fatty acids and no cholesterol, so it provides a lot of oil with a good quality. Because of the understanding of mating systems and the possibilities of controlled hybridization within and among species, as well as the naturally existing variability in the species, great advances in sunflower genetics have been made. New methodological techniques, including as molecular mapping of crucial agronomical traits and the establishment of advanced-backcross and introgression lines, have provided significant tools for sunflower crop improvement and understanding domestication processes. The overall goal of sunflower breeding is to produce high-oil-yielding, drought-resistant lines with a single capitulum and a better root system. It is apparent that sunflower breeding results were based on traditional breeding-genetic procedures, and that no significant shift in the faster introduction of valuable features in cultivars has been made. It is undeniable that traditional breeding will not help to boost output in the future. As a result, tremendous progress in molecular genetics and the utilisation of molecular marker techniques is being made. As a result, using a combination of traditional breeding and modern plant biotechnology technologies, such as marker-Assisted Selection, for sunflower breeding could be beneficial.

Keywords: Sunflower, plants, *Helianthus annuus*

Introduction

Sunflowers are annual plants in the Asteraceae (synonym Compositae) family, which includes daisies and sunflowers. The plant's stem can grow up to 3 metres tall, and the inflorescence can reach a diameter of 30 cm. The sunflower inflorescence, commonly known as a capitulum, is a constricted raceme composed of multiple separate sessile florets that all share the same receptacle. It appears to be a single flower, similar to other Asteraceae. It is the second most significant oilseed crop in the world, after soybean, and comes from temperate North America. It contains a significant amount of unsaturated fatty acids and has no cholesterol, thus it provides a lot of oil with a good quality. It's a diploid crop with a 3000 Mb haploid genome and a diploid chromosomal number of $2n=34$ (Darvishzadeh *et al.*). Sunflowers have been successfully cultivated in a broad variety of locations around the world. It is a highly cross pollinated crop with great yield potential that may be adapted to a variety of environmental circumstances. It can be cultivated in all seasons and taken in a variety of inter and sequence cropping methods due to its photo insensitivity. Its acreage has expanded in both emerging and developed countries due to its modest requirements and good oil quality. It is currently grown on a global scale of 20.00 million hectares, with a yield of 30.00 million tonnes and a productivity of 1,500 kg ha⁻¹. Asia accounts for about 20% to 22% of the world's total sunflower acreage, contributing 18% of the total production. India is the leading country in the Asian sub-continent with an area of 0.29 million hectares and annual production of 0.21 million tonnes, and productivity of 738 kg ha⁻¹. Because of their increased seed yield capability as compared to other cross-pollinated species around the world, hybrids, particularly in sunflower, are becoming increasingly important. Hybrids are more stable, highly self-fertile, and have a higher yielding ability as well as greater uniformity at maturity. Drought and disease tolerance breeding has become increasingly important in the development of hybrid cultivars. The heterotic performance of a hybrid combination (showing superiority over their parents) is largely determined by the parents' combining ability. Heterotic hybrids were created by mating inbred CMS female and restorer lines with high GCA (General Combining Ability) and SCA (Specific Combining Ability) scores, according to the researchers. Higher GCA variation implies additive gene activity, which depicts the line's breeding value, but higher

SCA variance shows a stronger role for non-additive gene action, which is critical in heterosis breeding (Shabbir *et al.*). Production of sunflower can be increased by improving its genetic potential. Moreover, sunflower breeders aim to achieve the highest seed yield and oil content through the expression of heterosis. It has high yield potential, wider adaptability, drought resistance, and salt tolerance. For the selection of superior varieties/hybrids, genetic variability is necessary. In an efficient breeding program, selection of better parents for drought tolerant genotypes and the crosses to develop genetic variability is necessary for the identification of superior hybrids/ varieties.

Floral biology

Sunflowers are tall annual or perennial plants which grows up to 300 cm in some species. They bear one or more wide, terminal capitula (flower heads), with bright yellow ray florets at the outside and yellowish disc florets inside. Some ornamental cultivars of *H. annuus* bear red-colored ray florets. During growth, sunflowers changes its direction towards the sun and then stops once they begin blooming. This movement of the head is called heliotropism. Sunflowers grow to face east by the time they reach maturity. Sunflowers move due to heliotropism, which occurs when the sunflower follows the sun and the opposite side of the stem begins to accumulate growth hormones, causing growth and redirecting the sunflower. In wild plants, the rough and hairy stem is branched in the upper part, but in domesticated cultivars, it is normally unbranched.

The petiole leaves are tough and only seldom sticky. The bottom leaves are usually simple, oval, and opposite. Technically, they're identified by the fact that the ray florets (when present) are sterile, and by the existence of a calyx with two awn-like scales that shed on the disc flowers (that is, simply detached and falling at maturity). Some species contain extra shorter scales within the calyx, while others don't have one at all. Another technical feature that classifies the genus more accurately, but necessitates the use of a magnifier to investigate, is the presence of a prominent, cellular appendage at the design's apex. Furthermore, a flower's florets are structured in an extremely natural spiral.

The perennial species that frame the majority of these within the genus show a lot of variation. Some plants have most or all of the large leaves during a rosette at the bottom of the plant and produce a flowering stalk with smaller leaves. The majority of the perennials have disc blooms with all yellow lobes, but a few have disc flowers with cherry-red lobes. *H. radula*, for example, has no ray blooms at all. The "seed" of the sunflower is actually an associate fruit - a monocarpellate (one carpel) and indehiscent (they don't open at maturity) fruit with one seed that almost fills the natural covering but doesn't adhere to it. It's an allogamic plant that need the presence of insects during flowering, particularly honeybees, for seed formation.

Cultivars of sunflower

- Giant types: Giant type range in size from 6 to 14 feet tall. maturing late, Enormous heads measuring 12 to 30 inches in diameter, with large seeds that are white, grey, or striped with black. The oil concentration is really low. As an example Mammoth Russian.
- Semi dwarf varieties Characteristics Medium height - 4 to 6 feet, growing early. Heads range in size from 7 to 9 inches in diameter. Smaller seeds that are black, grey,

or striped. It has a high oil content of 35%. E.g. Jupiter, Pole star.

- Dwarf types: 2 to 4½ feet tall. Early maturing. Head size 5½ - 6½ " diameter. Small seeds, high oil content 37%. E.g. Sunrise, Morden, Co1, Co2.

Genetics of sunflower

It's a diploid crop with a 3000 Mb haploid genome and a diploid chromosomal number of $2n=34$. New scientific techniques, such as molecular mapping of critical agronomical traits and the establishment of advanced-backcross and introgression lines, have provided useful tools for improving tomato crops and understanding domestication processes.

In contrast to findings in other plant species, where phenotypic variations generated by domestication are due to a smaller number of genes with a substantial influence [Deorge *et al* and Cai *et al*], genetic investigations of sunflower domestication revealed that there are a larger number of genes involved in domestication, with the majority of genes displaying mild or moderate phenotypic differences. Another difference between wild and cultivated sunflowers is the copy number of long terminal repeats (LTR) retrotransposons and splicing divergence.

Breeding of sunflower

In our changing world, with an exponentially rising population and significant environmental changes, breeding methods are crucial for enhancing food productivity. The advancement of molecular biology, and later "omics" sciences and bioinformatics, has provided significant prospects for improving the efficacy of traditional plant breeding efforts. Molecular and bioinformatics technologies can be integrated into traditional breeding schemes to examine huge numbers of traits and crosses rapidly during the early seedling stage, or they can be used to create novel breeding schemes and programmes that were previously unattainable. The structural and functional characteristics of plant genomes have been improved thanks to advances in genetics and genomics.

Sunflower production used to be based on open-pollinated cultivars (Vear, 2016) [28]. Sunflower production shifted to hybrid breeding with the discovery of the first cytoplasmic male sterility (CMS) source and identification of matching restorative genes (Kinman, 1970) [16]. In 1972, the first commercial sunflower hybrid was released for production in the United States (Putt, 1978) [21]. Farmers were able to acquire larger seed and oil yields as well as increased uniformity by utilising heterosis for hybrid creation (Bohra *et al.*,2016) [3]. In recent days, both the public and private sectors have contributed to the development of a significant number of plant genetic resources in order to find markers for marker assisted selection (MAS) as well as the development of novel high-throughput technologies in sunflower. Global sunflower output is now projected to be worth \$20 billion per year.

Breeding objectives

Drought resistant nature with good root system:-

When it comes to drought tolerance breeding, the crop's root structure is quite significant. The effectiveness of moisture extraction from the soil is determined by root properties such as root biomass, maximum root length, and root volume. Sunflower genotypes with a deep and wide root system may extract water from a depth of 270 centimetres (Hilli, H.J 2021) [13]. Plant genotype, moisture or water availability, plant

population, and soil condition all have a role in root development and growth. An increase in root length, particularly during moisture stress, along with a reduction in lateral root length, can be regarded an adaptive measure for drought stress. Higher root growth was associated to increased drought resistance in these experiments, indicating that the creation of abiotic stress tolerant hybrids and lines is needed to increase production.

Resistance to diseases and pests

Breeding for resistance to the most harmful pests and illnesses is a typical breeding goal. If molecular markers for all known resistance genes are available, a considerable leap in exploitation of polygenic resistance might be made with the release of sunflower genome sequences. The development of disease resistant cultivars is perhaps one of the most important contributions of modern plant breeding to sunflower improvement.

Breeding for self fertile lines

Protoandry and self incompatibility mechanism operates in sunflower. Hence hand pollination is necessary. To avoid this, self fertile lines can be developed.

Branching architect

Branching has been extensively studied as one of the most significant modifications in flower architecture throughout domestication and breeding. If ancient endemic farmers had hand-picked this feature from among wild populations, this alteration in branching pattern could have occurred throughout or before the domestication procedure [Lentz *et al.*]. Branching is a fancy trait on that genetic background, according to studies of the genetic foundation of branching in crosses between wild and domesticated flowers [Burke *et al.*]. There are many publications about the number of genes controlling branching [Putt 1940, Putt 1964, Hocket *et al.*, Kovacik *et al.* and Gentzbittel *et al.*] ^[22, 23, 14].

This trait is particularly intriguing in helianthus, as branching could be a wild species-related trait that was lost in cultivated helianthus, only to be reintroduced from wild helianthus in restorer genotypes in order to lengthen the capitula variety and therefore guarantee prolonged pollen production and successful crossing between female (cytoplasmic male sterility-cms line) and male (cytoplasmic male sterility-cms line) a male and restoring helianthus male lines. The branching locus, B locus, was discovered on the upper portion of chromosome ten in helianthus, and it is recessive in branching genotypes [Bachlava *et al.*]. Many loci for domestication and post-domestication, such as oil content and achene weight, were discovered flanking B locus by the researchers. Given that branching may be a more advanced trait in crosses between wild and cultivated helianthus, as well as a significant domesticated trait in helianthus, several research have been carried out to identify QTLs linked to branching.

Methods of sunflower breeding

Mass selection

Mass selection is the process of selecting a number of phenotypically superior plants, seeds from the field population, harvesting and bulking their yield in order to sow the following year, and repeating this procedure until desired traits are obtained. This can be accomplished by simply cultivating the desired variety in the field and removing the

undesirable variety. The remaining plants (probably the good ones) are bulked up when they reach maturity. Tagging only the best plants in the field and harvesting them in bulk at maturity is one adaptation. The main difference between the two procedures is the degree to which selection is intense. Modern plant breeding usually employs mass selection to preserve the characteristics of established varieties [Frankel *et al.*]. In this method phenotypically superior plant are selected and harvested from a population and their seed are mixed to continue new variety modern and is useful for characters which are highly heritable. E.g. Plant height, disease resistant.

Hybridization

Currently, there is a greater focus on developing the sunflower hybrid that was first launched in India in 1980. For hybrid seed production, cytoplasmic genetic male sterile lines are accessible in isolated fields, and the BHS-1 (CMS-234*RHA-274) hybrid seed production programme is used.

Heterosis breeding

In Russia, inbred lines were developed and crossed in order to harness heterosis as early as 1920. In wild types and obsolete cultivars, cytoplasmic genetic male sterility was discovered in 1970. This method is now widely employed in the development of hybrids. Eg: BSH-1

Biotechnology in sunflower breeding

Most future breeding efforts to leverage the heterogeneity in the cultivated and wild relatives of sunflower are projected to rely on traditional breeding techniques. Due to the length of time required for breeding, traditional improvement methods are time-consuming and inconvenient, and there is a problem with selecting adequate breeding criteria. As a result, establishing simple and efficient regeneration systems is a necessary precondition for using cell and tissue culture for genetic enhancement (genetically transformed plants for commercial applications).

Only few works have been done on this crop related to genetic engineering. The development of transgenic sunflowers with increased fatty acid oil content i.e Seed oil composition analysis showed significant decrease in stearic acid content in the seeds obtained from transgenic plants in one of the research work carried out. Some progenies shown less than 10% saturated fatty acid content whereas other plants had high palmitic acid content with reduced stearic acid content (Rousselin *et al.*). Hydroxymethylglutaryl-CoA (Hmgr-CoA) and Erwinia uredovora phytoene desaturase (Crtl) genes were introduced into sunflower to obtain potential increase in oil quality (Dagustu *et al.*).

A progress remains being created in expeditiously reworking flower crops however stable transformation of flower plants is simply time consuming in generating homozygous lines and in regeneration of tissue. Also, flower features a long life cycle and transient expression of genes is an alternate technique in elucidating molecular mechanisms like operating of promoters, regulation of sequence, subcellular localization of proteins, protein stability, protein-protein interactions and small RNA function. Wild helianthus species give larger contribution as an rich supply of genes in crop improvement to motivate economic viability in cultivates species as major oilseed crop (Seiler and Fredrick). As having a narrow background in domesticated sunflowers with deficient genes, discovery of distinctive genes from wild helianthus plants is indispensable and is still underway. This might facilitate in

developing transgenic sunflowers with desired traits from wild population.

Transgenic technology holds imperative role in flower breeding and exerts robust guarantees to extend yield, oil content, insect/fungal resistance, stress tolerance and production of biopharmaceutical proteins. Albeit having improved techniques in flower, additional economical transformation protocol must be explored for generating hyperbolic success rates in getting transgenic sunflowers additionally as look for candidate genes with elite traits in developing transgenic crops will stay apparent. Traits that are being studied in flower for environment release is rare. Sunflower is known to have a high exposure to gene flow ultimately generating continuous variability. Strict environmental monitoring is inevitable to preclude undesired outcomes.

Genetic Maps and Sunflower Genome Sequence

The RAPD markers were first used to create maps (Rieseberg *et al.*, 1993) [24]. At a subsequent stage, maps were created in different crosses of grown sunflower utilising non-PCR based RFLP markers. Due to firms participating in the production of the sunflower map, these maps were published several years later than RLFP maps, for example, in wheat, maize, barley, rice, and oilseed rape (Hu, 2010) [15]. AFLP markers were later added to the maps. The number of haploid chromosomes in sunflower was represented by 17 linkage groups in the majority of the linkage maps generated. Following these maps were genetic maps based on SSR markers. The first composite genetic SSR map covered 1423 cM and included 278 single-locus SSR markers as well as 379 additional markers (public and proprietary). This map that nowadays serves as reference genetic map for sunflower was then further saturated with additional SSR markers exploring three new mapping populations (Yu *et al.*, 2003) [30]. In between more than 2,000 SSR have been derived from genomic sequences (gSSR) and EST (EST-SSR) and are now available for mapping and genotyping Chapman *et al.*, 2008; Heesacker *et al.*, 2008) [6, 12]. Existing helianthus maps were more enriched by these gSSRs, EST-SSRs, INDELs, TRAPs markers. These SSR markers (sequences and primers offered through NCBI) represent awfully valuable tool as they permit the localization of genes on individual linkage groups also as on the recently revealed helianthus ordering sequence of HanXRQ. regarding three gigabases (Gb) representing 80% of the total ordering size were assembled and represent a very useful tool for all totally different research analysis programs that aim at the advance of helianthus hybrids. Finally, the step toward high-density maps was created attainable by help of SNP-based markers, beginning with Lai *et al.* (2005) [18] who derived SNPs from an EST database (as part of the Compositae Genome Project) and used them for mapping. An Infinium Beadchip including 9,480 SNPs based on transcriptome data was developed by Bachlava *et al.* (2009) [1] to obtain four high-density genetic maps.

Marker Development by Linkage Mapping

Linkage maps that have been developed provide an excellent foundation for locating and mapping simple hereditary traits. Most downy mildew resistance genes, which provide resistance to the oomycete *Plasmopara halstedii*, have been discovered to be dominantly inherited, making molecular mapping very simple. The discovery of closely related markers is also a useful starting point for map-based cloning.

Puccinia helianthi causes the rust disease. This fungus, which is mostly spread in North America, Argentina, South Africa, and Australia, can cause significant damage and yield reduction in infected fields. Although genetic management of the disease is possible, resistance is only short-lived due to the rapid creation of new races through sexual or asexual reproduction. As a result, a lot of effort has gone into finding rust resistance genes and incorporating them into commercial lines and hybrids with the goal of pyramiding resistance genes to generate long-term resistance.

Association Mapping

In sunflower, only one of the association mapping studies so far was performed genome-wide (Mandel *et al.*, 2013) [20], all others were candidate gene based Genome-wide association mapping was performed using 5,359 SNP marker from the Illumina Infinium Beadchip. Flowering period, branching, and heterotic groups all have associations that were investigated. Although there was a lot of variation in LD across the genome, substantial marker-trait relationships were discovered. The genome-wide changes in the LD profile could be due to disease resistance selection as well as initial domestication (Mandel *et al.*, 2013) [20]. This first screen was followed by a more detailed, refined association mapping approach based on candidate genes for branching. Shoot branching was differentiated in no branching, apical, mid-apical, mid, mid-basal, basal branching as well as whole plant branching or other phenotype.

Summary and conclusion

Sunflower is a cross-pollinated diploid species with seventeen pairs of chromosomes (2n=34) and is a cross-pollinated diploid species with seventeen pairs of chromosomes. It belongs to the asteraceae family and contains a lot of unsaturated fatty acids and no cholesterol, thus it has a lot of oil benefits and a good quality. The cultivars with good root system which is highly desirable for the crop especially during stress conditions, which aid in extracting moisture from deeper layers of the soil. Breeding for the lines with restorer lines are also much useful during hybridisation programme. The cultivars with high oil yielding and with single capitulum per plant are more desirable for successful yield levels.

It was discovered that current sunflower breeding achievements are based on traditional breeding-genetic procedures, and that no significant changes in the rapid introduction of valuable traits into cultivars have been produced. It is reasonable to believe that conventional breeding will not allow for future productivity increases. The significant progress in molecular genetics and use of molecular marker techniques are established. Therefore, the combined application of traditional breeding and contemporary plant biotechnology methods including selection based on molecular markers marker-Assisted Selection might be valuable tools for breeding.

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