

## Research Article

### CANNABIS SATIVA: AUTO FLOWERING AND HYBRID STRAINS

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#### ABSTRACT

This review paper highlights and discussed about Cannabis autoflowering and hybrid strains. Many Cannabis cultivars are obtained by crossing plants from commonly considered subspecies *Cannabis sativa*, and *Cannabis indica*, native of Indian origin are used to produce new varieties suitable for different uses, such as fiber, oil, medical drug, and recreational applications. There are more than 750 strains of recreational Cannabis, the most with colourful names. Some are strains of *Cannabis sativa* and *Cannabis ruderalis* subspecies are crossbred hybrids. The hybrid strains can be named in a variety of ways. Smell or lineage are common ways of naming. Afghan Kush, Hindu Kush, Green Kush, and Purple Kush are all pure *Cannabis indica* strains. *Cannabis ruderalis* contains very low levels of Δ<sup>9</sup>-tetrahydrocannabinol. *Cannabis ruderalis* flowers as a result of age, not light conditions, which is called autoflowering. It is principally used in hybrids, to enable the hybrid to have the autoflowering property. The locus responsible for the "autoflower" trait (Autoflower1), as well as a major effect flowering time locus, Early1, were mapped using bulked Segregant Analysis. Autoflowering plants offer the benefit of quicker harvest but they yield half as much flower per unit area as photoperiod-sensitive plants. It may be possible to complete two crop cycles per growing season using autoflowering plants. However, the second crop will be affected by the same problems that affect late-harvest photoperiod-sensitive plants. Another limitation of autoflowering plants is precocious flowering in response to transplant stress, which can significantly reduced yield potential.

**Keywords:** Autoflowering, Cannabis ruderalis, Hybrid strains, Indian Himalayan Region, Short-day plant, Δ<sup>9</sup>-tetrahydrocannabinol (Δ<sup>9</sup>-THC).

#### INTRODUCTION

*Cannabis sativa* was illegal during most of the 20th century, but has recently been decriminalized or even legalized in some countries (1-27). This reflects the fact that Medical *Cannabis sativa* (drug or marijuana) was illegal and needed to be rapidly distinguished from Industrial *Cannabis sativa* (fiber type or hemp) in the context of law enforcement (1-18). However, the plant became controversial owing to some psychoactive components, Δ<sup>9</sup>-tetrahydrocannabinol (Δ<sup>9</sup>-THC) that have adverse effects on human health (1-18). Medical *Cannabis sativa* (drug or marijuana) and Industrial *Cannabis sativa* fiber-Hemp type plants differ in biosynthesis, concentration, and composition of metabolites (1-26). Cannabis has been consumed for its psychoactive properties for over 10, 000 years, and its estimated global market value is US \$340 billion by 2030 (1-18, 26). To date, much of research on Cannabis has focused on distinguishing between marijuana (drug type cannabis) and hemp (fiber/seed-type cannabis), quantifying cannabinoids accumulation in plant tissues and elucidating Cannabinoid biosynthesis (1-18). The use of the Cannabis plant as a source of therapeutic compounds is gaining great importance since restrictions on its growth and use are gradually reduced throughout the world (1-18). This has intensified the Medical *Cannabis sativa* (drug or marijuana) production stimulated breeding activities aimed at developing new improved cultivars with precisely defined, and stable Cannabinoid profiles (1-27).

Cannabis is predominantly a photoperiod-sensitive, quantitative short-day plant (20-25). *Cannabis sativa* L. is an influential yet controversial agricultural plant species that today enjoys great interest with a very long and prominent history of recreational, medicinal, and industrial usages (1-26). This plant can be cultivated for the production of fibers (used to make different textiles), seeds (rich in unsaturated fatty acids for edible oils), and drugs from its female inflorescences that contain cannabinoids (compounds with psychotropic or psycho pharmaceutical effects) (1-25). Among these latter, the principal psychoactive constituent of Cannabis is Δ<sup>9</sup>-tetrahydrocannabinol (Δ<sup>9</sup>-THC), and the concentration of this metabolite is at the basis of the distinction between hemp and drug (marijuana) types, with hemp considered low in concentration, 0.3% or less THC content (non-psychoactive), and marijuana, on the other hand, containing up to 1 to 38% of Δ<sup>9</sup>-tetrahydrocannabinol (Δ<sup>9</sup>-THC) by dry weight (1-27).

The major challenge facing the Cannabis industry is the need to develop new cultivars with desirable Cannabinoid profiles, high productivity, pest resistance, and overall vigour (1-26). Cannabis is considered as a facultative short-day plant (1-18). Growers use long photoperiods during propagation and vegetative growth phases and induce flowering using shorter photoperiods (1-25). Therefore, Cannabis may be considered under some circumstances as a day-neutral plant and provide a deeper understanding of Cannabis inflorescence development (1-18, 26, 27). Another challenge in breeding new Cannabis and hemp cultivars lies in the poor understanding of the phylogeographic structure and domestication of Cannabis (1-18). Day-length was found to be the most important factor influencing population structure (1-26). In case of hemp, yield

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and quality are largely determined by the cultivar, but environmental factors such as temperature and photoperiod also have strong influences on these parameters (1-18). Cannabis plants are susceptible to a variety of pathogens (fungal and bacterial) and insect pests that contribute significantly to yield losses (1-19). This is a particularly difficult challenge to address due to the nature of plant growing systems where natural predators do not exist, and the use of chemical control strategies is undesirable because of the residues left on flowers (1-18). Finally, propagation of vigorous, uniform plants remains a challenge for the Cannabis industry because this crop is dioecious and relies on cross fertilization for seed production (1-18).

## CANNABIS : AUTOFLOWERING

*Cannabis sativa* is usually considered as a short-day plant, flowering once night length reaches a critical threshold (20-25). The critical photoperiod varies by cultivar, but is typically between 12 and 14 h (21-25). Variations in flowering time within and across cultivars in outdoor grown populations have been previously identified, likely corresponding to genetic differences in this critical night length (20-25). Photoperiod-insensitive (day-neutral or autoflowering) cultivars of Cannabis are available, which initiate flowering based on maturity instead of day length (21). The autoflowering trait likely originated from plants growing in northern latitude locations where they experienced long days and a short growing season (20-25). It has been suggested that autoflowering Cannabis plants comprise a separate species or subspecies called *Cannabis ruderalis* or *Cannabis sativa* ssp. *ruderalis* (20-25). Autoflowering cultivars are used for grain production (1-25). However, there is also interest in them for flower production, because they mature significantly faster than photoperiod-sensitive cultivars (20-21-25). Outdoor flower production of photoperiod-sensitive plants requires 3.0 to 4.5 months from planting to harvest, depending on the cultivar and the planting location (21-25). At time of harvest, flower yield and quality may be adversely affected by frost and cold, rainy, or humid weather (21). As a result moisture related diseases, including *Fusarium* and *Botrytis*, are a problem for late-to-harvest crops (20-25).

**Autoflowering** plants offer the benefit of quicker harvest (45 to 75 d), but they yield half as much flower per unit area as photoperiod-sensitive plants (Coolong et al. 2023) (20-25). It may be possible to complete two crop cycles per growing season using autoflowering plants (2025). However, the second crop will be affected by the same problems that affect late-harvest photoperiod-sensitive plants (20-25). Another limitation of **autoflowering** plants is precocious flowering in response to transplant stress, which can significantly reduced yield potential (21-25). The development of high-yielding, early-flowering cultivars would benefit flower producers, especially in northern latitudes (21-25). Recently, triploid Cannabis cultivars, that exhibit significant reduction in fertility, have been developed as a solution for crop loss from seed set during outdoor flower production in areas experiencing pollen drift (21-25). The triploid Cannabis cultivar **Stem Cell CBG** was found to produce 98% less seed than its diploid counterpart, following exposure to feminized pollen which has lower viability than pollen from genetically male plants (21-25). If triploid Cannabis continues to demonstrate significantly reduced seed set compared with diploids when exposed to pollen, then triploid cultivars will be sought for flower production (20-25).

Further, some *Cannabis sativa* are photoperiod insensitive, colloquially referred to as "autoflowering" (20-25). This trait has been described as a simple recessive trait with major impacts on phenology and yield (20-25). In one of the study, the locus responsible for the "autoflower" trait (*Autoflower1*), as well as a major effect flowering time locus, *Early1*, were mapped using bulked

Segregant Analysis (20-25). *Autoflower1* confers photoperiod insensitivity in diverse *Cannabis sativa* germplasm, and segregates in a simple, recessive (Mendelian) manner (20-25). While *Autoflower1* is recessive with respect to photoperiod insensitivity, plants that were heterozygous for *Autoflower1* flowered approximately 2 weeks earlier than plants that were homozygous for *Autoflower1* under field conditions (20-25). This earlier flowering resulted in smaller plants with less total biomass, but may be useful for higher latitudes, as cultivars that are heterozygous for *Autoflower1* can produce very high yields in a shorter growing season (20-25). Beyond segregation for *Autoflower1*, several elite populations marketed as cultivars have been demonstrated as segregating 1:1 for a major-effect early flowering time phenotype (20-25). Breeder-friendly high-throughput molecular marker assays were subsequently developed for both loci (20-25).

Flowering time is important for all major market classes, and uniform flowering dates within a cultivar are essential for ease of harvest (20-25). Fiber hemp benefits from a long growing season, as harvest usually occurs around the flowering date, and early flowering results in a shorter vegetative growth phase to accumulate biomass (20-25). Grain hemp must flower early enough such that grain can be harvested before frost if growing in temperate latitudes, but precocious flowering can lead to severe yield penalties due to a lack of time to accumulate biomass that provides photosynthate for grain filling (20-25). For cannabinoid production, as with grain production, precocious flowering may result in reduced floral biomass yield, while plants that do not flower by the end of the season fail to accumulate high concentrations of cannabinoids (20-25). Additionally, Cannabinoid profiles change throughout the maturation of the inflorescence, making initiation of flowering an important factor in timing regulatory compliance testing and harvest (20-25). It has also been well established that some plants are photoperiod insensitive (day neutral), a trait proposed to have been introgressed from high-latitude populations, which have been classified by some as a putative species (1-25). This trait has been suggested to be inherited in a simple, recessive, Mendelian fashion (20-25). Different taxonomic classifications have also been proposed, including the putative species *Cannabis ruderalis*, which has been considered as the source of the "autoflower" trait in all *Cannabis sativa* populations (20-25).

There is demand for early-flowering Cannabis (*Cannabis sativa*) cultivars to hasten harvest and avoid late season detrimental weather conditions (21). Kurtz et al., (21) conducted a field study and greenhouse studies and evaluated the effect of gene dosage at the autoflowering locus on flowering timing for diploid and triploid hybrids between autoflowering and photoperiod-sensitive parents (21-25). According to this study, Autoflowering × photoperiod-sensitive hybrids were all photoperiod sensitive, but their critical photoperiods were longer than for homozygous photoperiod-sensitive plants, which resulted in earlier flowering (1-25). For triploid genotypes, decreasing dosage of the photoperiod-sensitive allele (A), from AAA to AAa to Aaa, reduced the time to flowering (1-25). Flowering timing for the diploid genotype Aa was intermediate between Aaa and AAa (21-25). These results provided the evidence of incomplete dominance of the A allele at the autoflowering locus (21-25). Plants of genotype Aaa flowered 32 to 40 days earlier in the field than genotypes of AA, 15 days earlier than genotype Aa, and were ready for harvest by the second week of August in Connecticut (21). Plants of Aaa were as tall as other diploid and triploid photoperiod-sensitive genotypes studied, which suggests that they have similar yield potential (21-25). The use of tetraploid autoflowering (aaaa) maternal plants in combination with diploid photoperiod-sensitive (AA) pollen parents to produce Aaa genotype seed is a reliable approach for developing early-flowering cultivars of Cannabis for flower production purposes (20-25).

## CANNABIS HYBRID STRAINS

Cannabis is a genus of annual flowering plant. Cannabis is often divided into 3 species—*Cannabis sativa*, *Cannabis indica*, and *Cannabis ruderalis* (3). Both *Cannabis sativa* and *Cannabis indica* are the native of Indian origin found wild in Indian Himalayan Region and other parts of Asia, China, Nepal, Bhutan, Pakistan, Afghanistan and Iran (1-25). *Cannabis sativa* can grow up to 5–18 feet or more, and often has a few branches. It is a tall thin-leaved plant, which flowers under certain light conditions (3). Flowering is initiated when darkness exceeds 11 h per day (3). *Cannabis indica* typically grows 2–4 feet tall and is compactly branched (3). *Cannabis indica* is more broad-leaved than *Cannabis sativa* (3). It is most commonly shorter and has more leaves and buds, giving it a more bushy appearance (3). The buds tend to be wider. It typically grows 2–4 feet tall, and is compactly branched. It contains very low THC, which is rarely grown by itself (3). These subspecies differ in phenotype and chemotype, and the main characteristics according to which they are commonly distinguished are size, leaf shape, terpene accumulation, the quantity and chemistry of cannabinoids produced and earliness of flowering (2). A great amount of interest from breeders is focused on the determination of the subspecies “composition” of the parental lines used in crosses and that of the obtained offspring. It is important to consider the origin and phylogeny of a line or cultivar to better plan breeding strategies and guarantee a higher level of traceability (2). Whether for medical or recreational use, costumers are increasingly interested in tracing the origins of the products they use (2).

*Cannabis ruderalis* contains very low levels of  $\Delta^9$ -tetrahydrocannabinol so is rarely grown by itself (3). *Cannabis ruderalis* may have originated from southern Russia (3). *Cannabis ruderalis* flowers as a result of age, not light conditions, which is called autoflowering (3). It is principally used in hybrids, to enable the hybrid to have the autoflowering property (3). There are more than 750 strains of Cannabis, often with colourful names (3). Some are strains of *Cannabis sativa* and *Cannabis ruderalis* subspecies. Many are crossbred hybrids (3). The hybrid strains can be named in a variety of ways (3). Smell or lineage are common ways of naming (3). There are only a few rules about how the strains are named, and the most strains' names do not follow the rules (1-20). Each species can be interbreed with the other species, leading to hybridization (3). It is important to note that there is ongoing hybridization, to create new strains and select desirable characteristics (3). One of the characteristics frequently chosen is THC content (1-25). While there is some concern with regard to the high THC content of modern hybrids, there is probably no added therapeutic effectiveness (3). There are more than 750 strains of recreational Cannabis, most with colourful names (3). In some countries, the recreational forms of Cannabis can be used for medical purposes (3). There are a couple of examples where there does seem to be some sort of naming convention. Kush typically indicates either pure *Cannabis indica* or a *Cannabis indica* hybrid (3). Afghan Kush, Hindu Kush, Green Kush, and Purple Kush are all pure *Cannabis indica* strains (4). Blueberry Kush and Golden Jamaican Kush are hybrids based on *Cannabis indica* (1-25). For *Cannabis sativa*, there are similar examples using the words Diesel and Haze, with some strains being pure *Cannabis sativa*, and some strains being hybrids of *Cannabis sativa*. Diesel is named for its smell (1-25).

Cannabis exists in many varieties and sub-species (1-25). Among people using Cannabis for pain management, OG Shark, Afghani, and Skywalker are among the most popular strains (4). All strains of Cannabis derived from the Cannabaceae family of plants (1-25). Some experts consider that *Cannabis indica* and *Cannabis sativa* are the two main subspecies. Many producers crossbreed

Cannabis plants to develop new strains with specific characteristics (1-25). Experts suggested that there more than 750 hybrid strains as the Trusted Source of Cannabis (4). Kush- Pure *Cannabis indica* or *Cannabis indica* hybrid (4). Afghan Kush, Hindu Kush, Green Kush, Purple Kush- Pure Cannabis indica. Further, Blueberry Kush, Golden Jamaican Kush-Cannabis indica hybrid (4). Finally, Diesel Haze-Pure *Cannabis sativa* or hybrid (4).

The Cannabis inflorescences are comprised of individual flowers clustered together in a raceme and are harvested after 7–8 weeks of development in the flowering phase of production (27). They are dried under specific temperature and relative humidity conditions for 4–5 days to achieve a moisture content of 10–12% prior to packaging and distribution to the medicinal and recreational markets (27). Depending on its therapeutic effects, researchers noted that some participants preferred certain hybrid strains over others (4, 26, 27). For example, the top three Cannabis strains for insomnia or sleep disorder are, Lemon sour diesel, OG shark and Skywalker (4). For mental health conditions, such as anxiety and depression, the commonly used hybrid strains are, Jack Herer, Island sweet skunk, and White widow (4, 26). Potential uses of these cannabinoids can include treating some of the health disorders are pain, nausea, cancer, appetite loss and eating disorders, epilepsy, spinal cord injury, glaucoma, multiple sclerosis, Tourette syndrome, anxiety, post-traumatic stress disorder (PTSD), irritable bowel syndrome (IBS), and sleep problems (4, 26). Further, people typically use CBD to treat the, inflammatory bowel disease, seizures, depression, inflammation, psychosis or mental disorders, and migraine (4, 26). THC has potential uses in treating some of the health disorders are glaucoma, rheumatoid arthritis (RA), muscle spasticity, low appetite, insomnia. People may use Medical Cannabis sativa (drug type or marijuana) either to treat pain anxiety, and nausea (4, 26).

## CONCLUSION

*Cannabis sativa* L., a member of the family Cannabaceae, is cultivated worldwide for production of fiber and seed (as hemp) and for its medicinal and psychotropic effects found in high THC-containing genotypes. *Cannabis ruderalis* has the property of autoflowering and flowers appear as a result of plant age rather than light conditions. It is principally used in hybrids, to enable the hybrid to have the autoflowering property. The locus responsible for the “autoflower” trait (*Autoflower1*), as well as a major effect flowering time locus, *Early1*, were mapped using bulked Segregant Analysis. The plants are smaller than *Cannabis sativa* and have adapted to colder temperatures than *Cannabis sativa*. It does not typically grow to be more than 2 feet tall and is unbranched. It has been suggested that autoflowering Cannabis plants comprise a separate species or subspecies called *Cannabis ruderalis*. There are many different genotypes of *C. sativa* (also referred to as chemotypes) that accumulate THC to different extents, ranging from 0 to 0.3% (hemp) to over 1 to 38% (expressed in relation to dry weight of the harvested female inflorescences or “buds”) in Medical *Cannabis sativa* (drug or marijuana). These phytochemicals, in particular  $\Delta^9$ -tetrahydrocannabinol (THC) and Cannabidiol (CBD), are synthesized from precursor molecules of phenolics and terpenes by specific enzymes and accumulated within stalked glandular heads of trichomes. These trichomes are found primarily on bract tissues within female inflorescences. There are more than 750 strains of recreational Cannabis, most with colourful names. Some are strains of *Cannabis sativa* and *Cannabis ruderalis* subspecies are crossbred hybrids (3). The hybrid strains can be named in a variety of ways (3). Smell or lineage are common ways of naming.

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