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BIOTECHNOLOGICAL INNOVATIONS FOR ENHANCING ORNAMENTAL CROP TRAITS

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ABSTRACT

Biotechnological innovations have transformed ornamental crop breeding by enabling precise genetic improvement of traits. Techniques such as Agrobacterium-mediated transformation and gene editing have enhanced flower color, fragrance, vase life, and stress tolerance. Notable examples include blue carnations, blue roses, and orange petunias created through pigment pathway modification. Manipulation of ethylene-related genes prolongs vase life, while stress-responsive genes improve resilience. These advancements boost aesthetic and market value while reducing chemical use, fostering sustainability and competitiveness in the global floriculture industry.

KEYWORDS: Fragrance, Flower color, Genetic engineering, Ornamental crops, Vase life

INTRODUCTION

Ornamental crops are integral to horticulture, valued for their beauty and their ability to enhance gardens, interiors, and events. Rising demand for a broader range of high-quality ornamental plants calls for innovative production and breeding strategies. Traditional breeding methods like hybridization and mutation induction, while foundational, often confront hurdles such as lengthy breeding cycles, unpredictable outcomes and limited precision in enhancing aesthetic and post-harvest traits. Biotechnology offers powerful tools to enhance characteristics like resilience, visual appeal, and sustainability in ornamental plants. The first genetically modified ornamentals, such as color-modified carnations and blue (lavender) roses, showcased tangible market success and consumer appeal. The iconic orange petunia, engineered by introducing a maize-derived gene, exemplifies how transgenesis can yield novel hues unattainable through cross-breeding

GENETIC ENGINEERING (RECOMBINANT DNA TECHNOLOGY)

This technique involves transferring genes between organisms to introduce new traits in the recipient plant. Crops developed this way are known as transgenic or genetically modified (GM) plants, and the transplanted gene is termed a “transgene.” Transgenic breeding refers to the genetic improvement of crops via these methods to enhance economically valuable traits.

GENE TRANSFORMATION METHODS

DIRECT GENE TRANSFER

1. Chemical Mediated Gene Transfer
2. Microinjection

Chemical mediated gene transfer: Chemicals like polyethylene glycol (PEG) and Dextran Sulphate induce DNA uptake into plant protoplasts. Calcium phosphate is also used.

Microinjection: Where the DNA is directly injected into plant protoplasts or cells (specifically into the nucleus or cytoplasm) using fine tipped (0.5 - 1.0 micrometer diameter) glass needle or micropipette.

INDIRECT GENE TRANSFER

In this method, the gene of interest is first inserted into the T-plasmid of *Agrobacterium*. Plant cells are then infected with this bacterial culture. Following infection, the transformed cells are regenerated using plant tissue culture techniques.

AGROBACTERIUM TUMEFACIENS: NATURE’S GENETIC ENGINEERING

Agrobacterium species are soil-dwelling Gram-negative bacteria that cause crown gall or hairy root disease in plants. The genes responsible for tumor formation reside on a Ti (tumor-inducing) plasmid within the bacterium. When *Agrobacterium* infects plant cells—typically at wound sites—it naturally transfers a segment of its Ti-plasmid (called T-DNA) into the plant’s genome. This unique ability earns it the title of “natural genetic engineer” of plants.

Two main *Agrobacterium* species are typically employed for plant transformation:

- *Agrobacterium tumefaciens*
- *Agrobacterium rhizogenes*

This technique can be used to:

- **Knock Out Unwanted Traits**: Unwanted characteristics, such as susceptibility to diseases or certain growth habits, can be eliminated, leading to the development of improved varieties.

- **Fine-Tune Expression of Traits**: Gene editing allows for the modulation of gene expression, which can enhance specific traits without introducing foreign DNA, making the modifications more acceptable to consumers.

APPLICATION OF GENETIC ENGINEERING/ TRANSGENIC BREEDING

Genetic modification (GM) involves altering the genetic makeup of a plant to express desirable traits. In ornamental crops, GM can be used to:

- **Enhance Aesthetic Traits**: Genes responsible for flower color, size, and fragrance can be manipulated to produce new varieties with enhanced visual appeal. For instance, the introduction of anthocyanin biosynthesis genes can result in flowers with vibrant colors.

GENETIC ENGINEERING FOR FLOWER COLOUR

Flower color is one of the most important traits of ornamental plants influencing its commercial value. Flower color can also attract pollinators and protect floral organs. Several ornamental plants have been engineered for flower color modifications by targeting flavonoids, anthocyanins, carotenoids and betalains.

Flower color, critical to ornamental appeal and commercial value, is determined primarily by three pigment groups:

1. **Flavonoids** (notably anthocyanins)
   - Subclasses include cyanidin, delphinidin, pelargonidin, malvidin, peonidin, petunidin—responsible for red, blue, purple, and even black hues
2. **Carotenoids**
   - Responsible for yellow, orange and red hues.
3. **Betalains**
   - Naturally occurring in specific plant families, now engineered into others to broaden color options.

METHODS OF COLOR MODIFICATION

- **Gene introduction**: Adding genes like flavonoid 3′,5′-hydroxylase (F3′5′H) to enable delphinidin production in species like rose, chrysanthemum, and carnation—resulting in blue/violet hues.
- **Overexpression or suppression**:
  - ✓ Example: Expressing maize DFR A1 gene in petunia resulted in the first orange petunia (pelargonidin-based)
  - ✓ Silencing chalcone synthase (CHS) produces white flowers; suppressing F3′5′H shifts the pigment spectrum to reds/pinks

- **Betalain pathway engineering**: Introducing betalain-biosynthetic genes into torenia to generate reddish-purple flowers.
- **Carotenoid manipulation**: Editing carotenoid cleavage genes (e.g., CCD4) leads to cream/orange changes, as seen in morning glory and rapeseed.

Table1: Different Pigments in ornamental crops

Pigments	Colour	Example
1.Water soluble pigments		
a. Flavonoids	Scarlet	Scarlet pelargonium
i. Anthocyanins	Red and Magenta	Red rose
a) pelargonidin	Mauve, Purple and Blue	Blue delphinium
b) cyanidin	Deep red	Peony
c) delphinidin	Pink to mauve	Blue Streptocarpus
d) peonidin	Yellow	Pale yellow petunia
e) petunidin	Yellow	Yellow antirrhinum
f) malvidin	Cream to colourless	
ii. Chalcones	Ivory to deep yellow	
iii. Aurones		
iv. Flavones		
v. Flavonols		
b. Betalains	Reddish to violet	Flowers of cacti, *Amaranthus spp.*, bougainvillea
i. Betacyanins	Yellow to orange	
ii. Betaxanthins		
2. Lipid soluble pigments		
a. Chlorophyll	Bluish green	Green rose, Anthurium spathe
i. Chlorophyll a	Olive green	
ii. Chlorophyll b		
b. Carotenoid	Yellow	Marigold, daffodils, calendula
i. Xanthophyll		
ii. Carotene		

GENETIC ENGINEERING FOR BLUE CARNATION

Naturally carnation plants do not have the true blue colour flowers because lack of ‘Flavonoid 3', 5'-hydroxylase’ enzyme and delphinidin which are involved in flavonoid biosynthesis pathways which were responsible for blue colour pigment production. The scientists of Florigene Company were isolated the blue gene from the petunia in the year 1991 and they were got patented in the year 1992. Incorporation of the petunia gene into a carnation which was responsible for blue colour by that the carnation plant produces the blue colour flowers but this technique is not giving the good results in case of rose. Mauve coloured carnation variety that is 'moondust' developed by Florigene Company in the year of 1996. It was the 1st genetically modified variety of carnation which is in commercial market.

MOONSERIES OF FLORIGENE CARNATION

Table 2: Genes involved in pigment synthesis of different flower crop

Enzymes and Genes	Flower crop
Chalcone synthase (Chs)	Antirrhinum, Chrysanthemum, Carnation and Rose
Flavonoid 3'5' hydroxlase (F3'5'h)	Antirrhinum, Aster, Chrysanthemum and Orchid
Flavonol synthase (Fls)	Petunia and Rose
Flavone synthase (Fns)	Antirrhinum and Gerbera
Dihydroflavonol-4-Reductase (Dfr)	Antirrhinum, Aster, Carnation, Orchid and Petunia

EVOLVING OF BLUE ROSES THROUGH ENGINEERING OF THE ROSE FLAVONOID BIOSYNTHETIC PATHWAY (Katsumoto et al., 2007)

In roses, the native DFR gene efficiently makes red pigment but poorly produces blue pigment, making true blue roses difficult to breed. Using RNA interference (RNAi), scientists first silenced the rose DFR gene to block red pigment production. Next, they inserted a delphinidin gene from pansy (Viola sp.) to enable the blue pigment pathway. Finally, an iris DFR gene was introduced into *Rosa hybrida*, resulting in high delphinidin accumulation and a novel bluish flower colour. **Suntory** created the first, true blue rose named **"APPLAUSE"** in 2007

"APPLAUSE"

GENETIC ENGINEERING FOR FRAGRANCE

Fragrance is a key quality trait in ornamental plants, contributing to their commercial appeal and ecological roles such as attracting pollinators and deterring pests. Many modern ornamentals have lost their natural scent due to intensive breeding focused on color and form. Genetic engineering provides a precise approach to restore or enhance fragrance by manipulating biosynthetic pathways responsible for volatile compound production. Floral scents are mainly derived from three groups: terpenoids, phenylpropanoids, and aromatic amino acid derivatives. Genes encoding enzymes involved in these pathways can be introduced or overexpressed to boost fragrance levels.

For example, linalool synthase (LIS) from *Clarkia breweri* produces linalool, a pleasant floral scent. Other genes like IEMT, BEAT, and SAMT from *Clarkia* enhance methyl eugenol, benzyl acetate, and methyl salicylate, respectively. In roses, OOMT genes regulate the synthesis of key aromatic compounds, while in lilies, LiTPS genes have been engineered to accumulate linalool, myrcene, and β-ocimene. Genetic modifications can also fine-tune the timing and intensity of fragrance release to suit market preferences.

(Sheng et al.,2021) reported that the RrNUDX1 gene plays a role in enhancing the fragrance of petunia flowers, and they could lay an important foundation for the homeotic transformation of RrNUDX1 in R. rugosa for cultivating new *R. rugosa* varieties of high-yield and quality essential oil. (Zhang et al., 2020) inserted LiTPSs in tobacco plants and relative expression levels of 3 LiTPSs in flowers at different flowering stages and leaves was obtained by PCR. Results showed that the flowers of LiTPS2 transgenic lines accumulated linalool, myrcene and β-ocimene, which was consistent with the main scent components of oriental lily hybrids and the leaves of LiTPS2 transgenic lines released linalool as well.

Table 3: Genes involved in fragrance synthesis

Name of gene or gene product	Source	Product of the reaction catalyzed by encoded enzyme
LIS	*Clarkia breweri*	Linalool
IEMT	*Clarkia breweri*	Methyl eugenol and isomethyleugenol
BEAT	*Clarkia breweri*	Benzyl acetate
SAMT	*Clarkia. breweri*	Methyl salicylate
BEBT	*Clarkia breweri*	Benzyl benzoate
BPBT	*Petunia x hybrida*	Benzyl benzoate and phenylethyl benzoate
BAMT	*Antirrhinum majus*	Methyl benzoate
Myrcene synthase	*Antirrhinum majus*	Myrcene
OOMT1 and OOMT2	*Rosa. hybrida*	3methoxy,5hydroxy toluene and 3,5 Dimethoxytoluene and 1,3,5 trimethoxybezene
POMT	*Rosa chinensis*	3,5dihydroxyanisole

GENETIC ENGINEERING FOR LONGER VASE LIFE

In cut flowers, long vase life is a critical trait as flowers must survive several weeks before reaching the market. Yellowing of leaves in ornamental plants, associated with senescence, have a negative attribute decreasing plant attractiveness, quality and vase life. Ethylene is a plant hormone, which plays a major role in the senescence.

- ✓ The vase life of flowers can be altered by manipulating the biosynthesis of ethylene.
- ✓ The enzymes ACC synthase and ACC oxidase encoded by acs gene and aco genes.
- ✓ Transgenic flowers having antisense aco gene or sense acs gene have been produced and exhibited longer vase-life.

Transgenic carnations and petunias with antisense aco genes have shown significantly extended vase life. Such innovations enhance postharvest quality, reduce waste and improve the economic value of ornamental crops globally.

Table 4: Gene for improvement of vase life in flowers

Traits	Candidate Gene and Pathway
ACC synthase	Inhibition with reduced ethylene production
ACC oxidase	Inhibition with reduced ethylene production
ACC deaminase	Over expression with reduced ethylene production
Etr 1	Expression of a defective gene with reduced ethylene production
ERS	Expression of a mutated gene with reduced ethylene production

GENETIC ENGINEERING FOR BIOTIC AND ABIOTIC STRESSES

- Biotic stress occurs in plants due to damage done by plant pathogens or pests (bacteria, fungi, virus, insects, nematodes,) during production, storage, distribution and end-consumer use. Plant growth and yield are strongly influenced by biotic stress. By introducing genes that provide resistance to specific pathogens, GM can help reduce reliance on chemical pesticides, making cultivation more sustainable
- Abiotic stresses such as drought, salinity, cold and heat etc. These stresses can affect plant growth and productivity.

AREB/ABF TFs, *AP2/ERF*, NAC TFs, bZIP TFs, *MYC*, and *MYB* have been implicated in drought stress response. CBF genes encode *AP2/ERF* (*APETALA2*/Ethylene-Responsive Factor) transcription factors to initiate cold stress responses in plants. Genes such as Na+/H+ antiporter genes (*AtNHX1*, *AtSOS1*), *DREB1A*, *SOS1*, *ICE1*, *WRKY4*, *NAC1*, *SOS3*, etc. have been identified for imparting salt tolerance in flower crops. Heat shock transcription factors (HSFs) mediate the expression of heat shock protein, where several genes have been shown to impart heat tolerance such as; HSP70, FaHsfA2c, etc. (Sharma et al 2023). (Li et al., 2018) studied that chrysanthemum CmHSFA4 gene positively regulates salt stress tolerance in transgenic chrysanthemum. CmHSFA4 overexpressing plants showed less stress damage compared with WT chrysanthemum plants. (Hong et al., 2009) reported that expression of the *Arabidopsis DREB1A* gene in transgenic chrysanthemum enhances tolerance to low temperature.

CONCLUSION

Biotechnology and genetic engineering have revolutionized the ornamental crop industry by enabling precise improvement of traits that were difficult to achieve through conventional breeding. Genetic modification has facilitated the development of novel flower colors, enhanced fragrance, extended vase life and improved tolerance to biotic and abiotic stresses. Techniques such as Agrobacterium-mediated transformation and direct gene transfer allow targeted insertion or silencing of genes, producing transgenic varieties with greater market appeal and sustainability. Successful examples include blue carnations, blue roses, and orange petunias, showcasing how manipulation of pigment biosynthesis pathways can create entirely new hues. Similarly, genes regulating ethylene production have been engineered to prolong vase life, while transcription factors and stress-related genes enhance environmental resilience. These innovations highlight biotechnology's immense potential in expanding ornamental diversity, meeting consumer demand and reducing dependence on chemical inputs, paving the way for a sustainable and competitive floriculture industry.

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