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Article

Literature assessment and futuristic research approach on floriculture: Chrysanthemum

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Abstract: Immeasurable basic and applied information has been evolved on all important floricultural crops through large-scale worldwide research on interdisciplinary aspects. The quantum and quality of work done on Chrysanthemum, among all other ornamentals, are par excellence. Conscientious attempt has been made to collect the whole multidisciplinary experimental results achieved world over. Despite remarkable achievements in knowledge and technology, a major part of present experimental research on chrysanthemum is largely a routine repeat of work. Assessment of past and present work is now significant for preparing target-oriented future research resolutions. This will help to secure the favored results within a justifiable period.

Keywords: floriculture; chrysanthemum; phylogeny; cytology; tissue culture; mutation; characterization

1. Introduction

Commercial floriculture has developed into an ambitious business worldwide due to high-grade scientific technology-based expertise. The wide variations in flower shape, size, and color made the *Chrysanthemum* a very noteworthy ornamental crop for its unlimited use not only in the floriculture industry but also as a model plant for scientific research. The enormous volume of published work has been built up covering nearly all facets of *Chrysanthemum*. The present write-up encompasses almost the whole reported multitudinous research expedition on *Chrysanthemum* from starting to its present status quo. The author has made a comprehensive evaluation of multidisciplinary research work on *Chrysanthemum* which will benefit guide in formulating judicious future research road map plan with changing necessitates in floriculture. Some oversight may have dropped behind in conducting such a massive literature, regardless of attire attempts.

Chrysanthemum is a very favorable ornamental plant for multipurpose use throughout the world. Over time, remarkable basic and applied knowledge has been assembled on various aspects. Literature survey intimate that the work enriched the literature on the following topics-species and cultivars, evolution, systematics, germplasm status and their usages, phylogeny, geographical journey, medicinal use, propagation, nursery management, post-harvest, techno-economics, conventional breeding, induced mutagenesis, new varieties, cytogenetic, tissue culture, characterization, management of disease, etc.

Considering the magnitude of socio-economic importance and quantum of work accumulated over the period, it is very much needed to review the history of accumulated work done on *Chrysanthemum*. The review will administer a factual document on the past work, the proliferation of rise and fall, actuality and constraint of successes, and the futuristic work approach on *Chrysanthemum*. It is very meaningful to visualize the necessity for practicality studies after such a long period.

Over the decades there has been an incredible leap up in multidisciplinary scientific research around the world on all socioeconomic parameters of *Chrysanthemum*. To estimate its impact at this stage, a thorough evaluation of important contributions during the entire period is crucial. Substantial numbers of accessible published works were examined reasonably to conceive this report. No attempt has been made to elaborate on each reference.

The author made conscientious efforts to prepare a trustable updated diary covering all research results about *Chrysanthemum*. All published subjects have been spotlighted exactly after careful assessment on an experiment-to-experiment basis. The entire work has been classified into different aspects after a wise examination of the enormous literature. The most appropriate and inevitable references have been presented in the review and the complete 'Reference File' is restored which one can get on request (either from author or editorial office). Results of each survey literature were very confidently examined and mostly the routine results have been indicated briefly indicating only the subject. Results of interesting significant subjects have been elaborated considering their importance in future research.

2. Scrutiny of literature

The present write-up has been prepared entirely based on published available references. About 992 references were explored over the period to prepare the review. Explored research segments have been distinguished into the following ten classes based on work components-1. Botany/Geographical distribution/Origin/species/domestication; 2. Cultivation/growth/physiology/location trial; 3. Tissue Culture; 4. Post-harvest; 5. Disease; 6. Cytology/Genetics; 7. Biochemical/Pigments; 8. Improvement; 9. Characterization; 10. Dehydration. Whole papers published in each class have been highlighted in **Figure 1**. The publications on medicinal uses of chrysanthemum are voluminous and highly impressive. Separate article on this topic is in progress.

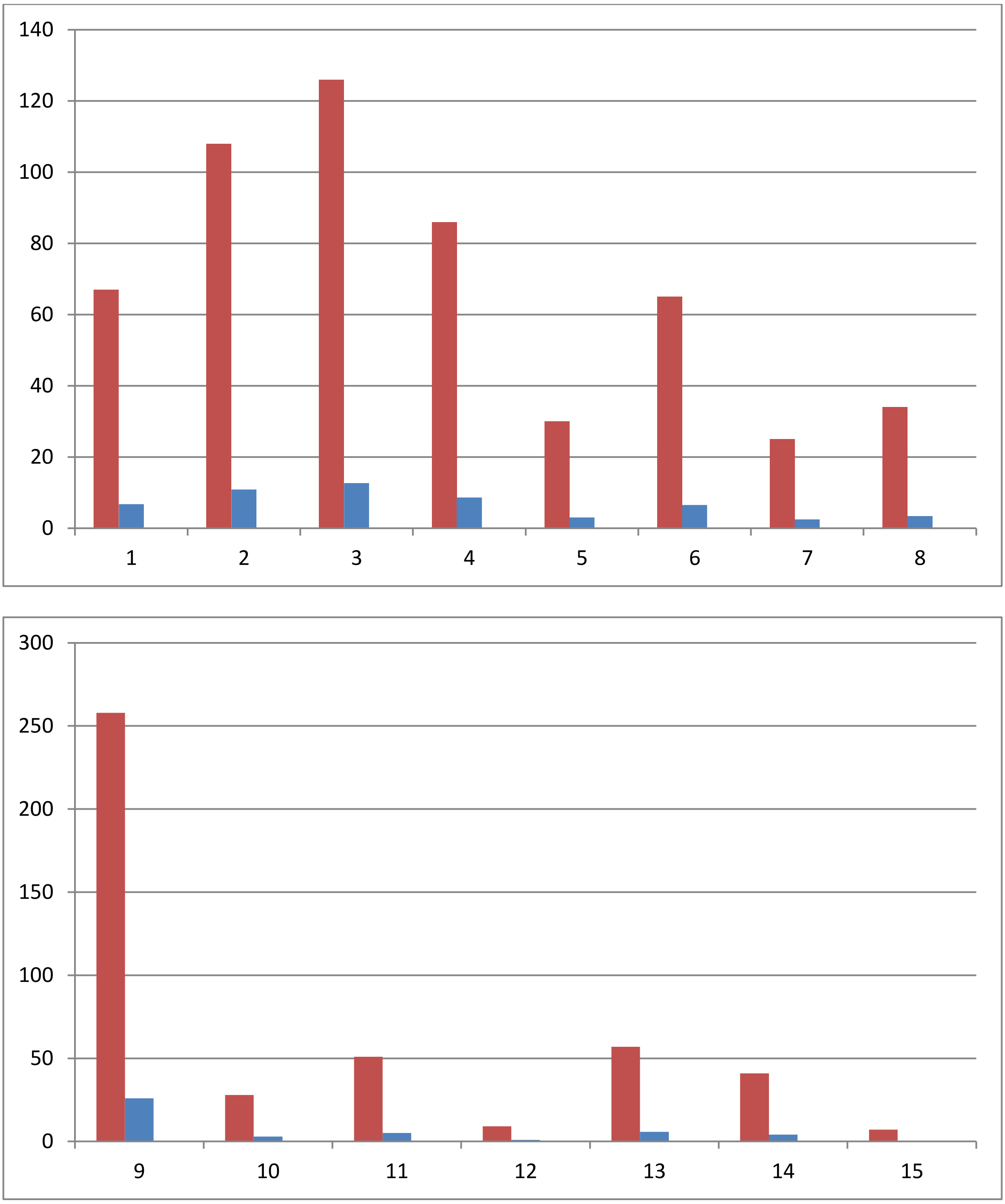


Figure 1. showing number of articles and their percentage values topicwise-1. (2.1. Botany/Geographical distribution/Origin/species/domestication); 2. (2.2. Cultivation/growth/physiology/location trial); 3. (2.3. Tissue Culture); 4. (2.4. Post-harvest; 5. (2.5. Disease); 6. (2.6. Cytology/Genetics); 7&8. (2.7. Biochemical/Pigments - 7. Biochemical 8. Pigments); 9. (2.8.i. Mutation); 10. (2.8.ii. Selection/Hybridization/Genetics);11. (2.8. iii. Biotechnology); 12. (2.8. iv. Somatic Embryognesis); 13. (2.9. Characterization); 14. (2.9. i. Molecular analysis); 15. (2.9. Dehydration).

2.1. Botany/Geographical distribution/Origin/species/domestication

The work report categorized under this class covers salient features of *Chrysanthemum*, early history, phylogeny, introduction to old books, market potential, species, origin and worldwide distribution, domestication, etc. In addition, it explored all about phytometabolites, chemodiversity, fluorescent in situ hybridization, single-copy nuclear gene, and chloroplast DNA sequences to reveal phylogenetic relationships, etc. Knowledge generated on all these issues is highly praiseworthy. Available 67 papers (RF 1-67) were scrutinized for this column.

The *Chrysanthemum* (family Compositae) comprises a complex polyploidy form grouping from 2× to 22×, and several aneuploids. A huge number of flowers is arranged on a flattened axis as a compact capitulum inflorescence-like floral head that looks like a single bloom. Two floral forms (ray florets and disc florets) are present in the bloom. The beauty of *the Chrysanthemum* flower head is mainly due to the large, charming, colorful, and wide range of shapes of ray florets. Female organ is present only in ray florets and both male and female germinal components are present in the disc florets. The bloom category is based on the respective number of florets and their shape and direction of growth [1–3].

Doubtably, the genus *Chrysanthemum* represents 200 species of which autumn autumn-blooming perennial *Chrysanthemum morifolium* Ramat is most notable. The name of *Chrysanthemum morifolium* has been changed to *Dendranthema grandiflora* [4–6] but most commonly it is known as *C. morifolium*.

Chrysanthemum primarily belongs to the northern hemisphere mainly in Europe and Asia. Its origin in China is also justified by many. Recently, author [7] systematically examined all relevant accessible records on the history of the geographical movement of chrysanthemums in different countries like China, Japan, France, the USA, and India. It covered the description of early varieties, the development of desirable commercial varieties, and many other aspects related to genetic diversification. The review indicates that all present-day glorious cultivars have been developed as a result of a network of involvement of elemental species in interspecific crosses, selection through open pollination, extensive intervarietal hybridization, spontaneous and artificial mutation techniques, etc. Literature indicates involvements of following elemental species to create the present distinction of *Chrysanthemum*-*C. boreale*, *C. carinatum* (tricoloured), *C. coronarium* (yellow and white blooms), *C. cinerariifolium*, *C. coccineum* (white, pink and red), *C. frutescens* (white and soft yellow flowers), *C. indicum* (yellow), *C. japonicum*, *C. maximum* (white and yellowish), *C. ornatum*, *C. satsumense, C. sibiricum, C. sinense* (white), *C. coccineum* (white, pink, or red), *C. Cinerariifolium*, *C. parthenium* (white or pale yellow), *C. balsamita* (yellowish with some white rays), *C. mayimum* (white perennials), *C. nipponicum* (white daisies*), C. rubellum* (pink to rose red), *C. cliginoseem* (white large daisies), *C. zawadskii* (rose-pink daisies), *C. alpinum* (daisies of glossy white), *C. arcticum* (white or pink), *C. mawii* (pink daisies, white with pink-reverse), *C. weyrich* (pink), *C. carinatum* (tricolour, purple or reddish rings with yellow and white base), *C. segetum* (corn chrysanthemum or Corn Marigold, deep yellow or whitish), *C. frutescens* (white or soft yellow daisies), *C. indicum* (yellow), *C. "hortorum"* (not a valid species), *C. sibiricum* (single flowers, white aging to carmine pink). Contribution to diversify genetic resources of some more wild species have been described—*C. oreastrum*, *C. hypargyrum*, *C. zawadskii*, *C. chanetii*, *C. naktongense*, *C. mongolicum*, *C. argyrophyllum*, *C. rhombifolium*, *C. vestitum*, *C. dichrum*, *C. glabriusculum*, *C. lavandulifolium*, *C. foliaceum*, *C. nankingense*, *C. potentilloides* and *C. maximowiczii* [2,8–15].

POWO [16] ('Plants of the World Online'-it was launched in March 2017) have reported some more valid/accepted species but their contribution in breeding new form not yet worked out—*Chrysanthemum aphrodite*, *C. arcticum*, *C. argyrophyllum*, *C. arisanense*, *C. chalchinqolicum*, *C. chanetii*, *C. arassum*, *C. cuneifolium*, *C. daucifolium*, C. dichrum, *C. foliaceum*, *C. glabriusculum*, *C. horaimontanum*, *C. hypargyreum*, *C. indicum* L., *C. integrifolium*, *C. japonese*, *C. konoanum*, *C. lavandulifolium*, *C. leucanthum*, *C. longibracteaum*, *C. maximoviczii*, *C. miyatojimense*, *C.* x *morifolium* (Ramat.)., *C. morii*, *C. naktongense*, *C. ogawae*, *C. okiense*, *C. oreastrum*, *C. ornatum*, *C. parvifolium*, *C. potentilloides*, *C. rhombifolium*, *C.* x *rubellum*, *C.* x *shimotomaii*, *C. sinuatum*, *C. vestitum*., *C. yantaiense*, *C. yoshinaganthum*, *C. zawadskii*, *C. zhuozishanense*.

2.2. Cultivation/growth/physiology/location trial

These are the most popular topic of interest by all. Therefore, the quantum of work done by different scientists is maximum. Series of 108 articles (RF 1-108) explored and the topics have been analyzed and results highlighted—Photoperiodic effect on seed germination, response to paclobutrazol residues on surfaces in sub irrigation; effect of planting dates on growth and flowering; effect of paclobutrazol on growth and flowering; influence of treating cuttings with various root formation stimulants; relation between nitrogen and soluble carbohydrate concentrations and subsequent rooting; influence of drugs on the intensity of root formation; influence of temperature on growth and development; effect of nitrogen, phosphorus and potassium levels on growth and flower yield; efficacy study of selected PGPRs on growth and flowering; effect of VA-mycorrhizae fungi application in chrysanthemum rooting and overall performance; effect of paclobutrazol, ethrel and B-9 on stem elongation and flowering characteristics; effects of irrigation method and controlled release fertilizer rate on leachate and growth parameters; studies on nutrient requirement; influence of substrate and plant growth regulators on the rooting; effect of plant growth regulators; effect of fertilizer, sodium chloride and foliar spray of benzyl adenine, cytozyme and potassium on growth and flowering quality; integrated nutrient management: N, P and K fertilizer studies; effect of pinching, disbudding and foliar spray; influence of air temperature on yield and phytochemical content; effect of chilling and gibberellin spray; role of uniconazole treatment in elongation of cuttings; effects of plant densities, pinching and growth regulators; role of MH-a substitute for pinching; adventitious roots development and root system architecture cuttings; effect of different planting dates in polyhouse and open field; comparing feasibility of various propagation methods; evaluation of different cultivars for growth and yield; effect of micronutrients; production and post-production quality of potted chrysanthemums; field evaluation of conventional vs. micropropagated plants; effects of saline irrigation; salt tolerance identification; effect of change in temperature during long nights on flowers; influence of artificial long-day treatment on expression of blooming period; studies on photoperiodic response; effect of supplementary illumination; use of short interval of light during night in delay blooming; assessment of standard *Chrysanthemum* cultivars under off-season cultivation; effect of modified photoperiod on the growth and flowering;

evaluation of cultivars for their flowering, flower quality and yield under different agro climatic zones; testing of off-season flower production and year round cultivation; study on fresh and exhausted mushroom compost substrates in greenhouse conditions; precision blooming; application of wireless power transmission led lighting system in propagation; wireless light-emitting diode system for micropropagation etc. A complete package of knowledge on agro-technology and techno-economics and multi-locational agronomical trials of germplasm and new varieties have been well standardized [17–23].

2.3. Tissue culture

Screening of 126 references (RF 1-126) indicates that a wide range of parameters were chosen for tissue culture work. Explants tested for standardization of *in vitro* culture were-leaf disc, meristem apices, petal segments, petal epidermis, shoot tips of the periclinal chimera, ray and disc florets, ovules and ovaries, capitulum, etc.

Objective and results of tissue culture reported on following aspects—Effect of culture media and growth regulators on *in vitro* propagation; rapid multiplication; optimization of factors affecting efficient shoot regeneration; efficiency of interaction between cytokinins and auxins; influence of auxin and growth regulators concentration on *in vitro* rooting; effect of season of collection of explants on micropropagation; assessment of cultivars based on direct shoot regeneration rates; effect of kinetin on the elongation of adventitious shoots regenerated *in vitro*; proliferation rate of axillary shoot; effect of agar concentration and pH levels on *in vitro* propagation; improvement of micropropagation system based on Pluronic F-68 supplmented media; new methodology for mass-production of micropropagated plants using microponic system in plant factory; standardization for low cost micropropagation; growth and photosynthetic characteristics of plantlets as affected by PH and EC of the nutrient solution in microponic culture; clonal fidelity of cultivars after long term micropropagation; effect of some of coconut water concentration in artificial media; , influence of genotypes in micropropagation; , adventitious bud and embryoid formation; callus culture and ability to synthesise pyrethrins; *in vitro* culture medium sterilization by chemicals and essential oils without autoclaving; standardization of protoplast-to-plant system; large scale production of plantlets using microponics with the supplement of silver nanoparticles under Light-Emitting Diodes; silver and gold nanoparticles impact on *in vitro* adventitious organogenesis; application of silver and copper nanocolloids in disinfection of explants; silver nanoparticles as the sterilant in large-scale micropropagation; standardization of an easy screening method to evaluate the non-branching characteristics in the juvenile plantlet stage; somaclonal variation in protoplast regenerants; assessment of somaclonal variation using RAPD and morphological assessment, repository of germplasm; establishing a hybrid artificial intelligence procedure for high-throughput modeling and maximizing tissue culture operation; utilization of bioreactor system for extensive production; development of *in vitro* plants through alginate-encapsulated shoot tips; primary and secondary somatic embryogenesis; evaluation of tissue culture technique in mutagenesis and genetic transformation; performance of NAA, 2iP, BAP and TDZ on callus multiplication; development of salt resistance strains etc. [24–33].

2.4. Post-harvest

Chrysanthemum is a very important floriculture crop for cut flowers and pot plants. Postharvest management is a very important research target in ornamental plants and scientists were stimulated to handle postharvest operations of *Chrysanthemum*. As per the description of 86 articles (RF 1-86), the research objectives mainly concentrated on cut flowers and pot plants with special reference to growth and flowering, increasing the flower longevity/vase life, foliage discoloration, flower opening, etc. A wide range of agents were applied and tested their efficiency to achieve the target results. Results accumulated on effects, changes and role of temperature, stem length, stomatal characteristics, desiccation response, carbohydrate content, storage treatments, holding solutions, pulsing, packaging, water relations and oxidative stress, membrane permeability, sucrose and 8-hydroxyquinoline citrate, cytokinin, cobalt chloride, silicon, calcium, nitrogen, phosphorus, magnesium, antioxidant enzymes and lipid peroxidation, ethylene, methanol, microparticles, growth regulators, 1 methyl cyclopropene, antitranspirants, B-9, 1-MCP, ABA, GA3, CO_2, benzyl amino purine, lipoxygenase and superoxide dismutase, ethylene and benzyl adenine, fertilizers, commercial floral preservatives, gamma radiation etc. [34–38].

2.5. Disease

A necessary strategy was adopted for the management of diseases. In-depth knowledge and different specialized techniques have been evolved to manage specific diseases. Targeted disease and technical procedure described in 30 papers (RF-30) followed are—standardization of tissue culture to develop virus free propagules; application of soil mulching coupled with fumigation to manage verticillium wilt; root rot, phoma collar rot, pests, *Botrytis cinerea* infection, *Rhizoctonia solani* infection, *Fusarium oxysporum*, PHOMA Chrysanthemicola, antimicrobial activities, fungus disease, virus diseases; antifungal activity against *Candida albicans* and *Pityrosporum ovale*, use of meristem tip culture to create chrysanthemums free from Cucumber mosaic virus, serological examination and to detoxicate infected plants, Cucumber mosaic virus strain, extensive transcriptome studies of grafting on *Artemisia scoparia* W. to influence the aphid resistance of chrysanthemum, management of biosynthesis of lignin through escalating CmMYB15 to furnish chrysanthemum resistance to aphides, white rust reaction of chrysanthemum and its transcriptome Scrutiny, study of intake of macro and micro nutrient in tissue culture raised plantlets by Arbuscular Mycorrhizal fungi (AMF) strains, safeguarding and control of Chrysanthemum from the aphid *Macrosiphoniella Sanborni* CmWRKY53, role of <scp>CmMYB15-like-Cm4CL2 to balance lignin biosynthesis of chrysanthemum in reply to aphid feeing, improvement of aphid resistance through over-expression of CmMYB19, negative regulation of Chrysanthemum CmHRE2-like towards protection of *Chrysanthemum* to the aphid (*Macrosiphoniella sanborni*) [39–42].

2.6. Cytology/Genetics

The main intention of cytogenetical work on *Chrysanthemum*, as mentioned in 65 articles (RF 1-65) was both basic, creative, and applied to solve the evolutionary race history. Materials included in cytogenetical studies were Chinese *Chrysanthemum* cultivars, wild *Chrysanthemum*, *C. morifolium* cultivars, species Indigenous to Korea, artificial intergeneric hybrid, induced somatic mutations, colchicine induced polyploidy, sport varieties, allotetraploid hybrids, *C. carinatum*, *C. nerifolium,* edible chrysanthemum cultivars, *C. segetum*, *C. zawadskii*, homologs in *C. argyrophyllum* L, homolog in *C. nankingense*, *D. indicum* var. *aromaticum,* F1 Hybrid of *C. makinoi* × Ch. Vulgare, intergeneric hybrid between *Dendranthema in*dica and *Crossostephium chinense* by GISH., *C. fruticulosum* complex, Octaploid. C. ornatum. Hemsley, some species of Chrysanthemum sensu lato in Russia.

Cytological studies mainly confined to—determination of chromosome numbers, Karyomorphological analysis, B-chromosome, meiotic chromosome characteristics in diploid individuals, determination of ploidy levels by flow cytometry, accessary chromosomes, cytohistological studies, genetic linkage map, chromosomal characterization based on repetitive DNA distribution in tetraploid cytotype, genomic relationships in allotetraploid hybrids, GISH identification of ancestor or closely related genome, hybrid genome characterization, identification of parental chromosomes, species relationship analyzed by FISH and GISH, intra-chromosomal changes, chromosomal distribution of repetitive DNA sequences, genomic DNA extraction, cloning and structure analysis of LEAFY homologues of genomic sequence, localization of 45S and 5S rDNA sites of Chrysanthemum and its related genera by fluorescent in situ hybridization, rapid genome reshuffling induced by allopolyploidization in F1 hybrid in *Chrysanthemum*, nature of telomere and 45S rDNA sequences etc.

Complex polyaneuploid (2n = 36, 45, 47, 51–75) nature was confirmed through investigations.

Substantial reconstitution and structural modifications have taken place during the course of domestication. These modifications might have taken place due to outbreeding, spontaneous and intentional hybridization coupled with mutation, chromosomal differentiation repatterning, and polyploidy [11,43–50]. Chromosome-scale genome gathering investigation indicate that

C. morifolium is likely a segmental allopolyploid. Genomic resources generated will be very helpful in escalating chrysanthemum genetic improvement [51].

2.7. Biochemical/Pigments

The subjects of study under these headings (Biochemical 25 papers (RF 1-25) and Pigments 34 papers (RF 1-34) are very extensive and studies have remarkably enriched the knowledge. The investigated topics are—traditional medicine, antioxidant enzyme and lipid peroxidation during aging, oxidative stress and antioxidant activity as the basis of senescence, analysis of fragrance compounds, biosynthesis of volatile terpenoid secondary metabolites, absorption spectra of intact flowers, effects of sulfur fumigation on chemical constituents, Carotenoid and anthocyanin composition in petals, evaluation of volatile compounds in tea chrysanthemum cultivars and elite hybrids, identification of the phenolic components and neurotrophic/neuroprotective activity of cultivar extracts, Physiological response and comprehensive evaluation of heat tolerance of chrysanthemums under high temperature stress, chemical and biological comparisons on supercritical extracts, mitochondrial DNA B Resour, effect of water stress on phytochemical accumulation, essential oil, formulation of solid perfume, analysis of the complete chloroplast genomes, Surveillance and recognition of high temperature tension using chlorophyll a fluorescence and infrared thermography, UHPLC-QTOF-MS-based targetless metabolomics and mineral element examination insight into the geographical variation, exploration of bioactive composite and assertion of crucial genes involved in flavonoid biosynthesis, relative assessment of varied cultivars by an antiinfammatory-based NF-κB reporter gene assay coupled to UPLC-Q/TOF MS with PCA and ANN, antibacterial traits of nanofibers containing chrysanthemum essential oil and their application as beef packaging, impact of terpene alcohol on terpenes aroma expression, sucrose and Methyl Jasmonate Modulate the expression of anthocyanin biosynthesis genes and to accelerate the prevalance of flower-color mutants in Chrysanthemum. Studies covered a wide range of species and their cultivars (*C. morifolium*, *C. indicum*, *C. boreale*, Chinese Chrysanthemum, *D. zawadskii*) [52–59].

2.8. Improvement

Worldwide breeders and researchers made massive attempts to improve *Chrysanthemum* with special reference to developing new desirable varieties. Comprehensive and intricate breeding approach and analysis uncovered that all glamorous present-day varieties have evolved by way of intricate interspecific crosses among elemental species, natural pollination, haphazard hybridization, selective breeding, bud sport, artificial mutation, selection, etc. This knowledge motivated scientists to develop new varieties as per market demand through selective breeding.

2.8.1. Mutation

It is very enthralling to record that an enormous amount of classical and modern mutagenesis works have been conducted for the improvement of *Chrysanthemum*. The literature review exhibited a maximum of 259 articles (RF 1-259) covering a wide range of statistics on induced mutagenesis work. X-ray, Gamma rays, Fast neutrons, Thermal neutrons, Microwave, Radioactive phosphorous, Ethylene Imine, Ethyl Methane Sulphonate, Colchicine, etc. have been applied as mutagens. Monumental knowledge and literature have been evolved on mutation work covering almost all basic and applied aspects related to LD_{50} dose, radio-sensitivity, selection of material, methods of exposure to mutagens, combined treatment, recurrent irradiation, split dose, colchi-mutation, mutant genotype, detection of mutation, mutation frequency and spectrum of mutations, nature of chimerism, management of chimera, *in vitro* mutagenesis, isolation of mutants, etc. [23].

Efficient aim resulted in the development of trait-specific mutations. NaCl-tolerant *Chrysanthemum* plants have been produced through *in vitro* whole plant and callus selections [60–63]. All indispensable basic parameters have been precisely systematized for the success of the mutation technique and one can get a clear picture of all technological progress and its fruitful implementation to create new varieties. Many interesting and unique genotypes of commercial importance have been developed through the need-based application of technique [23]. Enormous knowledge generated so far has qualified *Chrysanthemum* as a model plant that enriches the entire technology package covering the relevant technological refinements of mutation technique [7,64].

2.8.2. Hybridization/Genetics/improvement

28 (RF 1-28) papers could be found covering all on the improvement of *Chrysanthemum* with special reference to hybridization. Literature expedition proclaims the involvement of the following materials in hybridization studies—*C. japonicum*, *C. boreale*, *C. vestitum*, *C. grandiflorum*, *C. indicum*, *Dendranthema morifolium*, *D. nankingense*, *Nipponanthemum nipponicum* early blooming varieties, hexaploid varieties, etc.

The focal points of such studies were—inheritance and breeding potential, morphological variations in newly developed hybrids, fluorescence in situ hybridization and genomic in situ hybridization to identify the parental genomes in the intergeneric hybrid, FISH physical mapping of 5S rDNA and telomere sequence repeats, isolation of chromosomes and mutation in the interspecific hybrid, interspecific hybridization and backcrossing using ovary rescue and their cold tolerance characteristics, genetic studies of self-incompatibility, overcoming pre fertilization barriers in the wide cross, identification self-incompatibility, and multigenic self-incompatibility, etc. Comprehensive and intricate breeding approach and analysis uncovered that all glamorous present-day varieties have evolved by way of intricate interspecific crosses among elemental species, natural pollination, haphazard hybridization, selective breeding, bud sport, artificial mutation, selection, etc. This knowledge motivated scientists to develop new varieties as per market demand through selective breeding. Hybridization work review established excessive heterozygosity and diversity among *Chrysanthemum* in growth habit, flower, blooming period, fertility, etc. Methodic exercise throughout the world resulted in the development of a vast amount of new varieties for multipurpose use [14,65–69]. Collective efforts of the author and his colleagues developed a few unique varieties like—pompon type cut flower varieties, No pinch no stake mini varieties, out-of-season blooming varieties, etc. (**Figure 2A–C**). Excessive breeding combinations and their results established chrysanthemum has endless genetic treasure to create new forms [14].

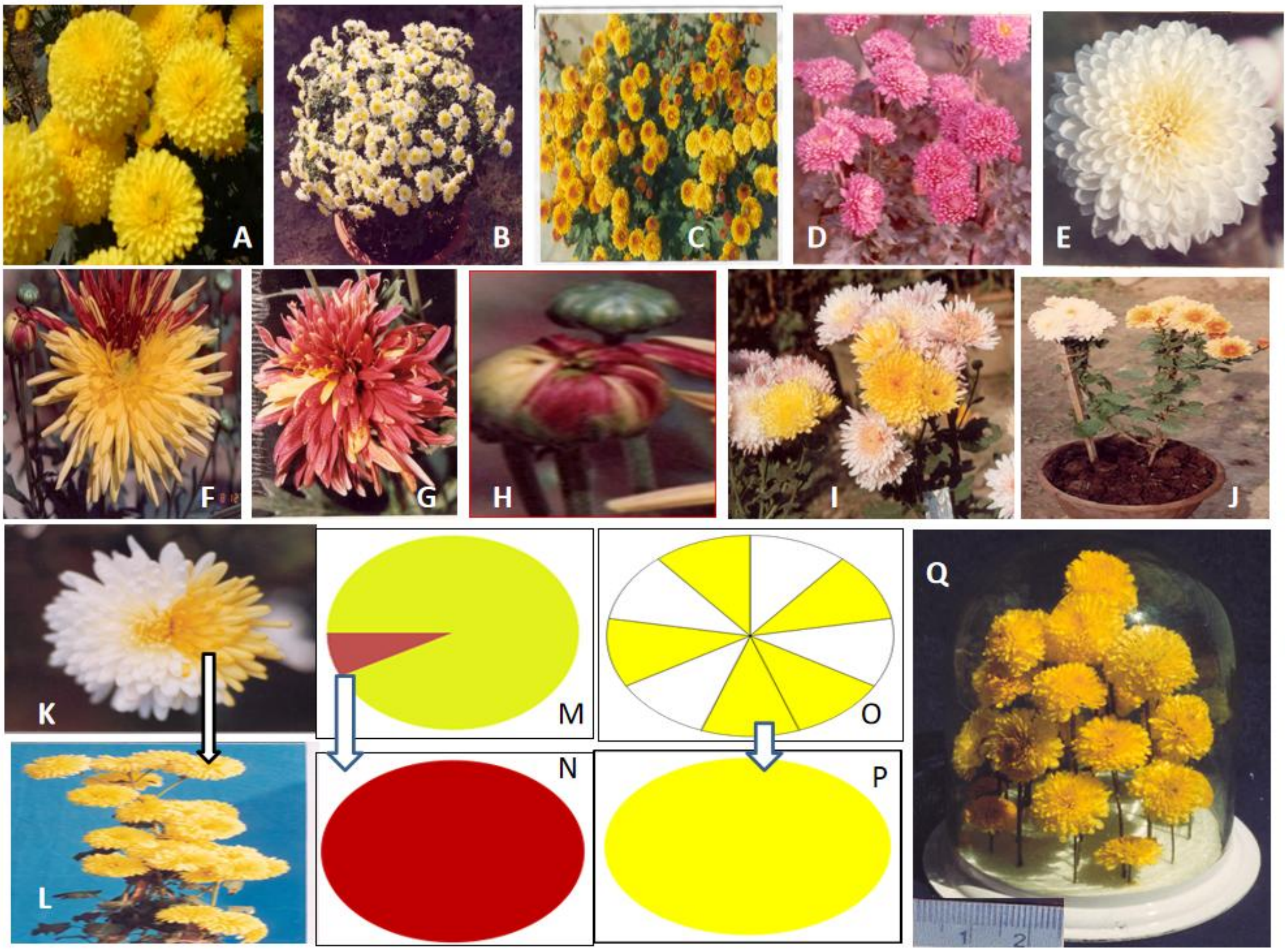


Figure 2. Showing new varieties, chimera, management of chimera and dry flower Chrysanthemum. New Hybrid Varieties: (**A**). Kundan, (**B**). Mother Teresa, (**C**). Little Darling; Gamma ray induced variety: (**D**). Shabnam, (**E**). Purnima; Chimera: (**F–H**): Flower chimera; (**I**,**J**): Branch chimera; (**K–P**): *in vitro* management of chimera; [K-original chrysanthemum cv. 'Lilith' (white) with sectorial chimera (yellow) and L-yellow sector established in pure form-cited from Dwivedi et al. 2000, IJAS 70(12): 853–855; Diagrammatic representation of chimera management–M-Chrysanthemum cv. Sharad Bahar with original (purple) and chimera (terracotta) and N-chimera established in pure form, and O-Yellow chimera in original white cv. Purnima and P- Yellow chimera established in pure form- cited from original Mandal et al. 2000, Euphytica, 114: 9–12]; Q: Dehydrated chrysanthemum.

2.8.3. Biotechnology

Different protocols were standardized for the improvement of *Chrysanthemum* through biotechnology. The ultimate aim of biotechnology is to create new variety and to induce novel characters. For the achievements of biotechnology regularization of diverse methodology is a prerequisite. Multifaceted technical steps developed by different researchers are mentioned concisely—study of some factors affecting *Agrobacterium* transformation and plant regeneration of *Chrysanthemum*; production of a white-flowering variety; stable expression of the GUS reporter gene depending on binary plasmid TDNA; standardization of the improved protocol for Agrobacterium-mediated genetic transformation; regeneration and *Agrobacterium*-mediated transformation; quick assessment of host-bacterium synergy in

Agrobacterium-mediated gene substitution to *Chrysanthemum*; reproduction and operational evidence of the CmHSP17.9 gene from *Chrysanthemum*; decrease in plant height in response to depletion of gibberellin due to assertion of the Arabidopsis Gai Gene; insertion of *Agrobacterium rhizogenesis* intervened untransformed roots and callus; appearance of foreign genes in steady form; transfer of nucleocapsid gene of tomato spotted wilt virus to chrysanthemum through biolistic methods; studies on sensitive procedure of *Chrysanthemum* to low temperature and operative validation of *Chrysanthemum* DgWRKY4 gene; expression of rice chitinase gene in transgenic Chrysanthemum exhibiting increased resistance to gray mold; ectopic appearance of the tobacco phytochrome B1 gene and alteration of plant structure; flower color modification by prohibition of F3H and over assertion of the exogenous Senecio cruentus F3 5 H gene; operational investigation of *Chrysanthemum* heat scaring protein gene CmHSP90.5; development of white flower in florist's chrysanthemum; investigation of transcriptome and metabolome of the flowers and leaves; genetic engineering for pelargonidin production; enhancement of aphid tolerance due to increased expression of a *Chrysanthemum* WRKY transcription agent; maximization of particle bombardment and Agroinfection criterion after stable transgene expression; investigation on necessity of the transcriptional coactivator CmMBF1c in favor of waterlogging resistance; enhancement of salinity resistance due to overexpression of a *Chrysanthemum* transcription factor gene dgnac1; salinity and drought resisting in Arabidopsis through heterologous articulation of a *Chrysanthemum* Cys2/His2 zinc finger protein gene; evolution of rare floral character in Chrysanthemum due to *Arabidopsis* chimeric TCP3 repressor; *Agrobacterium*-mediated transformation and somatic embryogenesis; cultivar specific genetic alterations applying wild-type *Agrobacterium* strain; development of transgenic *Chrysanthemum* through hygromycin tolerance selection; application of promoter gene and effective transgene expression for Chrysanthemum chlorophyll-a/b-binding protein; application of gene of *Bac. thuringiensis* 6-endotoxin and development of transgenic strains; use of mutant ethylene receptor genes (mDG-ERS1) to transform reduced ethylene sensitivity in Chrysanthemum and analysis of transformants etc. [70–76]. The entire work was highlighted based on the perusal of 51 papers (RF 1-51).

2.8.4. Somatic embryogenesis (RF 1-9)

Somatic embryogenesis is incredibly an efficient methodology of micropropagation. A single somatic cell differentiates into a somatic embryo through this artificial process. This technique has been applied in *Chrysanthemum* to evaluate the genetic stability and color of somatic-embryo-derived plants of radio mutants. Author [77] standardized efficient somatic embryogenesis strategy for utilization in the management of single-cell mutation affairs. This technique will be very useful to isolate new flower colors/shape new mutant varieties and to avoid chimera formation through retrieval of single mutated cells [78–81]. The literature survey detected 9 papers (RF 1-9) related to somatic embryogenesis.

2.9. Characterization

The concept of characterization was realized for the exact documentation of varieties, to guess genetic diversity and phylogenetic relationship, taxonomic stature, cataloging, deviation design, documentation of beneficial traits, crossing, registration, etc. A wide range of classical and modern techniques was applied to study different parameters like—chromosomal status, morphology, micromorphology, palynology, Support Vector Machine (SVM) and Multilayer Perceptron (MLP) algorithms, DNA markers, chemical and biochemical, etc. [82]. Characterization work was reflected on two types of materials—the original species and varieties, and newly developed varieties. The objective was to explore the genetic variations in germplasm (original materials) and to detect the genetic background of new varieties, as mentioned in 57 (RF 1-57) articles.

Researches explored on original materials are—Collection, assessment and maintenance of germplasm; evaluation for cut flowers; analysis of the morphological, anatomical variability and karyomorphology of wild species; identification of self-incompatibility; path coefficient analysis for flower production; discrimination of different white varieties by electronic tongue; promising selections for early and off-season production; chemical composition, and anthocyanin content of different cultivars; high-density genetic map construction and identification of loci controlling flower-type traits; use of isozyme analysis in breeding; development of easy screening method to evaluate the non-branching characteristics trait in juvenile plantlet stage and *in vitro* stage; determination of flowering time and flower number at low and optimum temperature; use of isozyme analysis in breeding; classification study based on multivariate statistical analyses, flower blooming pattern and color composition; characterization of *in vitro* haploid and doubled haploid plants (phenotypical traits and DNA methylation pattern); rapid recognition of cultivars and authentication of chrysanthemum teas by computer technology and strong understanding; to resolve origin of cultivars applying testimony of chloroplast genome and nuclear LFY gene; calibration of etiquette for Distinctness, Uniformity and Stability Test for *C. morifolium* (Ramat.) etc. Suitability of newly evolved promising selections of chrysanthemum (*Dendranthema grandiflora*) for pot culture; palynological and micromorphological interpretation of gamma-ray and colchicine-induced mutant cultivars; Cytological characterization showed similar chromosome number, symmetrical karyotype in original and mutants, and no mutant special abnormality. Chromosomal evaluation has confirmed that gene mutation might have taken the main role in flower color changes and neither due to alterations in chromosome number, chromosomal abnormalities or variation in karyomorphology [83]. Noticeable modifications in micromorphological and pollen grain characters were recorded [84,85]. A good amount of variety-specific morpho-chemical characters and desirable genes have already been identified through characterization. Passport data highlighting brief descriptions and other details have already been prepared. It is now most needful to utilize these data for planned breeding.

2.9.1. Molecular analysis

Accessible 41 papers (RF 1-41) were explored to perceive the activities on molecular aspects. Molecular characterization was performed to detect genetic relationships, assessment of genetic diversity, molecular systematics, classification, intravarietal variation, exploration of genetic variability among induced mutants, valuation of somaclonal variation, genetic diversity of

Chrysanthemum plants derived via somatic embryogenesis, examination of sports, microsatellite profiling for DUS testing, documentation of chloroplast DNA variations, distinguishing genetic variation among plantlets regenerated from irradiated calli, etc. The techniques mostly used were RAPD, RFLP, SSR, STS, SNP, VNTR, STR, SFP, and AFLP, start codon targeted (SCoT) markers, isozyme analysis, etc. DNA-based technique efficiently differentiated hybrid populations and original groups, dissimilar cultivars, radiation-induced mutants, sports, commercial varieties, etc. Genetic diversity estimated through these techniques will be practicable for the breeding plan to expand the heterosis of hybrids and introgress the new genes in the gene pool [55, 86–90].

2.10. Dehydration

Seven papers (RF 1-7) were identified which cover the preservation of Chrysanthemum through dehydration. Shelf-life of cut chrysanthemum flowers is very limited even after applying the best chemicals for enhancement of keeping quality. Dehydration technology has been standardized by which chrysanthemum flowers retain their fresh look for several months or even years and can be converted into value-added products (**Figure 2Q**). The demand for dry flowers has increased and the dry flower market has grown aggressively as consumers become 'eco-conscious' and prefer dried flowers as the environmentally friendly and biodegradable alternative to fresh flowers. Techniques have been standardized by which entire chrysanthemum flowers and individual florets can be dehydrated by press drying, hot air oven drying, microwave oven drying, etc. The dehydrated materials can be utilized for the preparation of floral craft for the dry flower industry [91].

3. Conclusion and Discussion

The present *Chrysanthemum* review will spotlight a deeper understanding of floriculture industry dynamics, important basic aspects, technical advances, and practical implementation of theoretical hypotheses. Exploration of publications elucidates the entire research expedition on *Chrysanthemum* from the very beginning to the current circumstances. The assembled depth of know-how will help to prepare a future blueprint of research approaches. Although various research fulfillments of earlier ventures have been mentioned, efforts have been made to recognize the weak points of different approaches. A sincere attempt has been made to interchange vision regarding a few particular techniques that are very pertinent for future courses of action. Independent paper evaluation suggests that a significant amount of work is a duplication of similar work which does not add any new knowledge. We should very sincerely stop this normal gap-filling research. Repetition of many works necessitates a changed attitude, the right perspective, and accurate research subject matter that would help to achieve the targeted goal in the right direction. Because many present activities have already been investigated and are not in synchronization with its real demand. Many publications appear to be inadequate in advanced constituents. It is very much required to examine thoughtfully the past results and reshape and reschedule the present research design to secure the commanding research status of Chrysanthemum.

Keeping in mind the past duration of research there must be a ripple of know-how and helpful information from the scientific circle down to the 'grass-roots level'. Literature inquiry is distinctly intimate that even though there are eloquent technological advances we are still parroted many of our experiments as overused activities year after year. Over the years we have rehearsed germplasm collection, geographical distribution, agrotechniques, characterization, disease management, post-harvest, breeding, etc. Germplasm collection is a leading activity but there is no clear picture about the specific number. This normal exercise generated duplication of varieties and unnecessarily duplicated nomenclature of the same variety. This stereotypic collection procedure has to be changed to avoid duplication and a centralized variety registration system should be developed.

Knowledge of genus, species, origin, geographical spread, domestication, taxonomy, botany, genetic diversity, characterization, improvement, etc. has been enriched enough. Proper care should be taken before repeating the same experimental design.

Agronomical evaluation has been excellently optimized applying all cultural practices. Vast literature has already been generated on different agronomical trial at different geographical regions along with their techno-economics. Very cautious future experiments should be designed that can generate new knowledge.

All basic information on tissue culture has already been explored from different angles. But tissue culture has enormous importance in in different experiments like mutation breeding, biotechnology, large scale commercial cultivation etc.

Post-harvest management on *Chrysanthemum* has been broadly researched covering all influencing factors which cause deterioration of harvested flowers. Variety-specific protocols have been generated and optimized on different management methods like the use of holding-, pulsing- and bud opening- solutions, growth regulators, gamma irradiation, pre-cooling, cold storage, packaging, etc. Now, the mindset to increase the publication through such routine repeat experiments only by changing varieties has to be changed. Advanced work on postharvest management to gain deep insight into the physiological and biochemical changes for the causes of senescence is to be intensified [53,92].

All symptomatic expressions of *Chrysanthemum* diseases have been scrutinized at different cultivation levels. But for the management of different diseases, accurate technical protocol knowledge has to be developed.

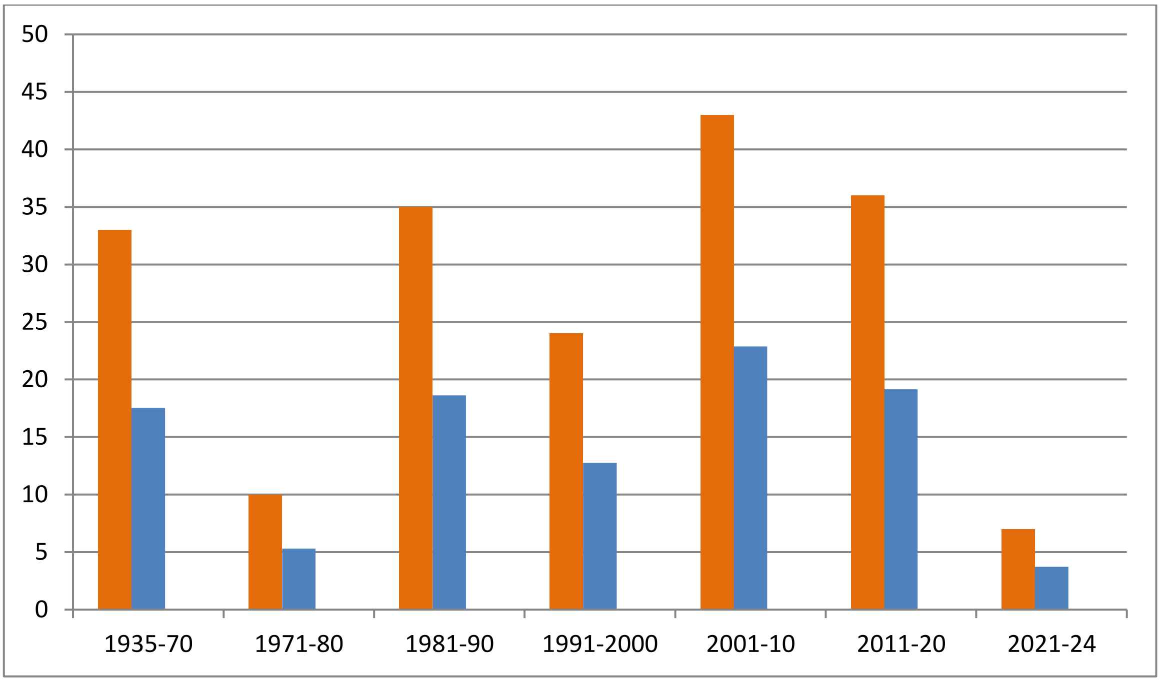
Chromosomal studies illuminated genetic-evolutionary race history and the genus *Chrysanthemum* was categorized as a poly aneuploid complex (2n = 36, 45, 47, 51–75). Assessment of chromosomal elements brought to light that there was substantial reconstruction and modifications during domestication. The main process of cytogenetical mechanism in the modernization of chrysanthemum has been itemized as outbreeding, spontaneous and intentional hybridization coupled with mutation, chromosomal differentiation repatterning, and polyploidy [11,43].

Cytogenetical exploration of *Chrysanthemum* are very meaningful but presently work on this area is negligible and the new report is almost at the ground level.

Biochemical research on *Chrysanthemum* is very important for multidisciplinary research. Early research has already achieved sensitive success in this regard. This is very significant topic of research in *Chrysanthemum* that needs special attention to achieve desired results.

Mutation has created not only flower color but also very attractive flower shape, chlorophyll variegated traits, leaf form, etc. (**Figure 2D**,**E**) Mutations in *Chrysanthemum* mostly develop as chimera, and isolation of such chimeric tissues is the main bottleneck in mutation breeding (**Figure 2F–J**). Novel *in vitro* technique has been standardized for restoring the chimeric mutant tissues. Further refinement of the *in vitro* technique resulted in to development of solid mutants through *in vitro* mutagenesis [23,93]. A massive amount of repeat experiments with color combinations have established that desired flower color mutations (directive mutation) can be induced in *Chrysanthemum* [23,94]. *In vitro*, techniques for the management of spontaneous and induced chimeric mutations and the development of solid mutants in *Chrysanthemum* have opened new vistas to produce new mutant varieties through chimera management (**Figure 2K–P**).

Sensitive scrutiny of individual mutation papers under each broad category identifies that one-fourth of papers focus on the same experimental design and results with only changing varieties. Mutation work was chosen as a representative case as mutation work on *Chrysanthemum* requires notable allusion to assess its impact on inducing mutations and its benefaction in enriching the mutation technology. 259 papers were accessible to the author of which 188 papers cover classical mutation work and 71 papers spotlight the management of chimera and *in vitro* mutagenesis. All papers were screened fervently and represented chronologically in **Figure 3**. Sixty percent of classical work chronologically enriched basic knowledge and the rest was on the whole repetition of earlier experiments/knowledge. It is very fascinating to mention that the new concept and its practical applications chronologically enriched the knowledge and finally a full package of mutation technology developed in *Chrysanthemum* (**Figure 4**). It will be realistic not to repeat now only routine classical mutation experiments but to follow the full mutation technology package (**Figure 4**) (classical + management of chimera and *in vitro* mutagenesis for creation of new and novel varieties) [94]. *Chrysanthemum* is, perhaps, the only ornamental species that may be pronounced as a model plant as one can obtain every enlightenment and idea for designing extensive mutation work on any ornamental crop. The mutation technique is now well-methodized, systematic, and cost-effective. Classical mutagenesis combined with the management of chimera and *in vitro* mutagenesis is most propitious for creating new and novel varieties [64,94].



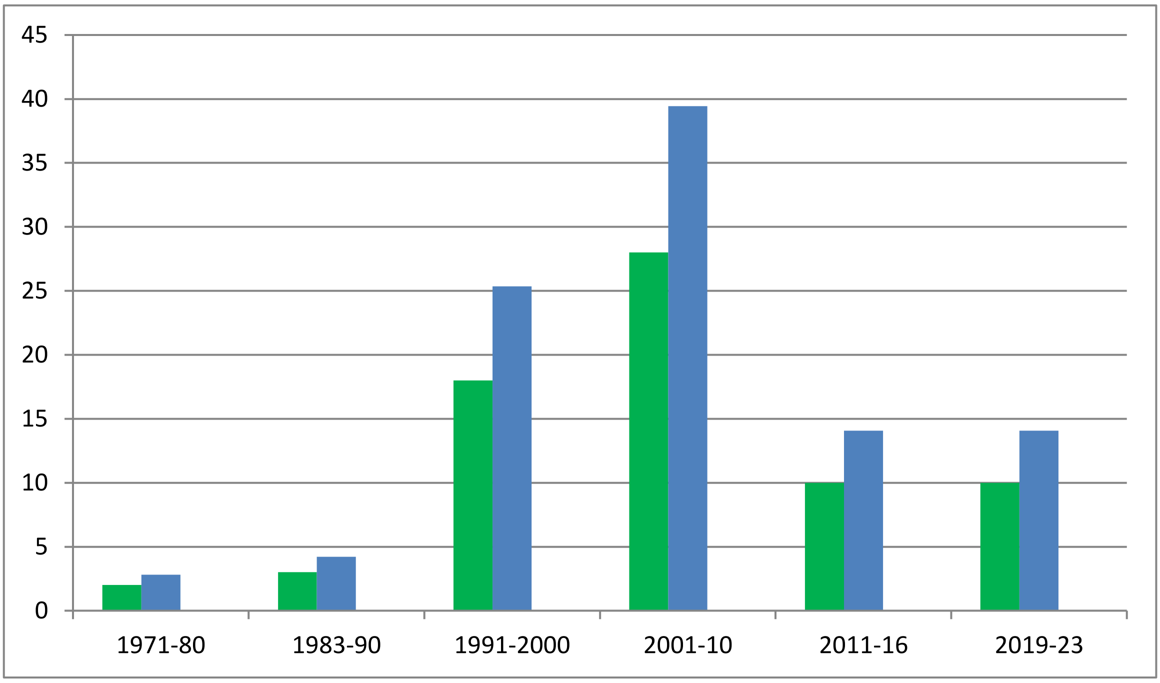


Figure 3. Chronological number of papers on classical [red box] and *in vitro* mutation [green box] and their percentage values [blue box].

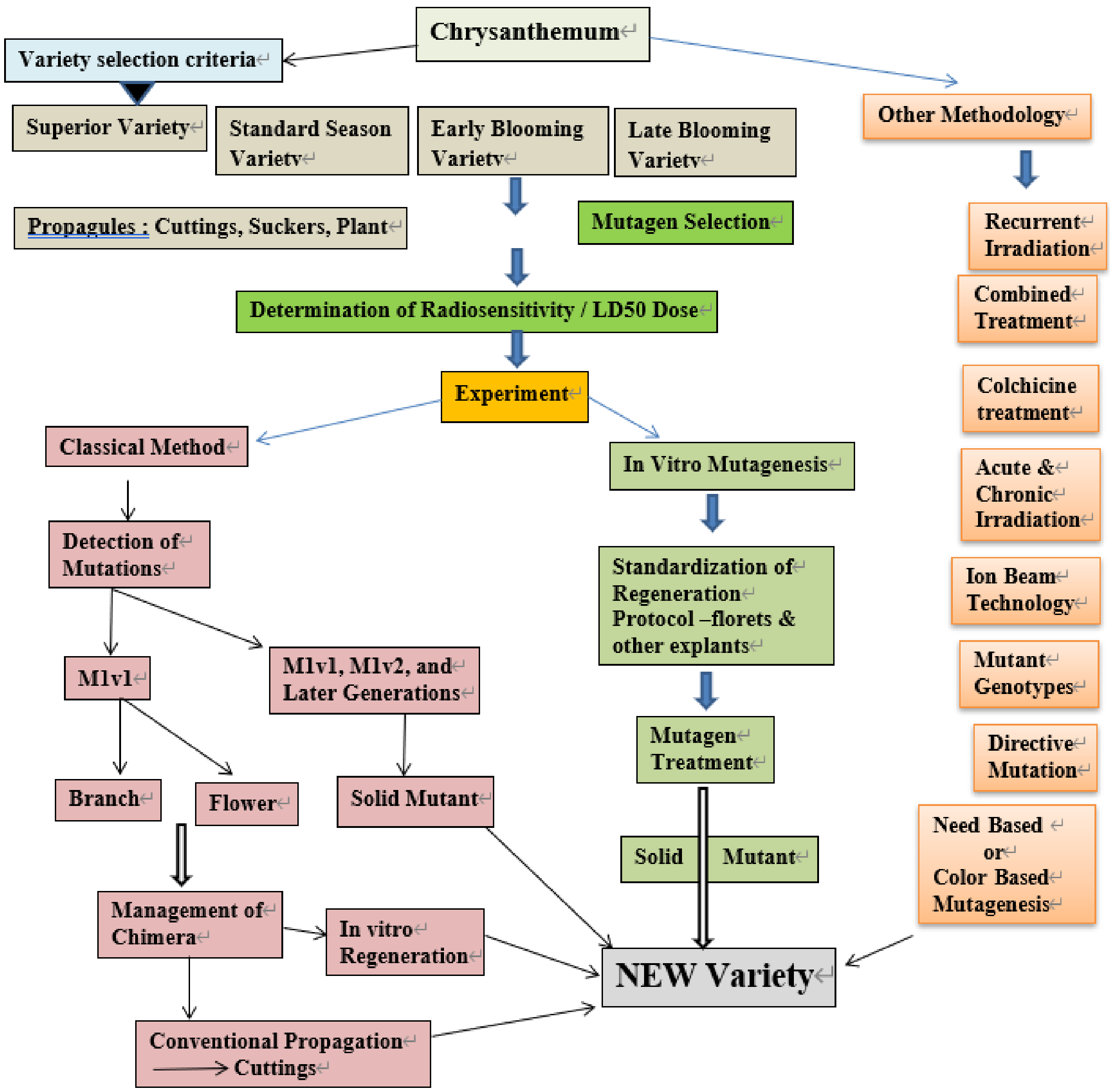


Figure 4. Mutation technology package for Chrysanthemum.

Literatures on the genetics and breeding of Chrysanthemum has appreciably enriched. But experiments on selective breeding are very valuable to explore the complex genetic system.

A variety of classical and modern breeding procedures have been applied to develop desirable traits. Heterozygosity and polyploid nature make it complicated to improve *Chrysanthemum* even after using molecular and omics techniques [95]. Selection of desirable genotypes and application of classical breeding, mutation, *in vitro* mutation, and management of chimera appear to be best in developing novel traits. The development of day-neutral varieties for year-round blooming through selective cross-breeding has a significant role in floriculture [3,14,23].

The biotechnological approach for creating new variety in Chrysanthemum has generated incredible basic knowledge, which may be exploited for future predicted molecular breeding. It hasn't yet significantly contributed consumer-friendly new variety in the floriculture trade. It is the beginning of an exciting new era.

DNA-based molecular techniques have been immaculately systematized cultivars, hybrids, mutants, etc. as a routine strategy for better discrimination. However, these molecular profiles have not been judicially utilized to establish the phylogenetic relationship. It is now very meaningful to use this technique not only for routine characterization but also for selective breeding [96]. Routine experiments on any DNA-based parameters may be avoided to increase the bulk of the literature.

All the leading *Chrysanthemum* varieties have been classified into their Response Groups and their susceptibility to photoperiods for year-round programmed blooming. Planting date and light intensity inside the greenhouse can be influenced and varieties can be marketed as per definite flowering dates and market demand. Now, this concept of year-round blooming and technology may be commercially exploited [21].

Efforts should be made for commercial exploitation of all earlier standardized techniques.

There is a necessity to evaluate the utilization percentage of laboratory techniques for commercial use not only for *Chrysanthemum* and other ornamentals but also for all plant-based experiments.

Different themes of early developed technology should be scrutinized at many stages which will help to reschedule the future mission plan. Researchers should be conscious of the prospective and the restrictions of many early viewpoints and can pick out the right blueprint that will be very relevant to reach the target. After a considerable period of research on a particular topic, new experiments should be designed to initiate new findings based on results that have come out from the last experiments/reports. Repetition of studies is easy to do and is a fairly common process but it does not add new knowledge in the technology domain. One should design, mastermind, and propagate new important ideas that will speed up innovation, creativity, and excellence.

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Data availability statement: The review article has been prepared through a critical survey of the literature on Chrysanthemum from the beginning up to the present status. The review has been prepared from approx. 992 articles. Only the relevant references have been cited and others are stored as Reference File (RF) 63 pages. It has been mentioned in the manuscript that a complete ‘Reference File’ (RF) is preserved with the author whom one can get on request (subodhskdatta@rediffmail.com).

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