INTRODUCTION
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Oil and fat industry:

Oils and fats rank among the most important agricultural commodities in both production and trade. Vegetable oils and fats represent an important source of food for humans. They are high calorie foodstuffs supplying required calorie in the diet and important vitamins. Vegetable oils derived from the processing of oil-seeds, account for 70% of the total availability of all oils and fats, the remainder being animal fats and fish oils. Vegetable or plant oils are usually classified into two major groups depending on their behaviour of heating: Volatile or essential oils and non-volatile or fatty oils. Volatile oils are produced from different aromatic plant species, belonging to different plant families mainly Lauraceae, Myrtaceae, Apiaceae, Lamiaceae and Asteraceae. Most of the volatile oils are used for manufacturing perfume and other scented items. Non-volatile or fatty oils are consumed as food, as raw materials for foodstuffs such as margarine, salad oils or shortenings, vegetable oils and fats are highly interchangeable with each other; their demand is mainly determined by availability and consumer habits. About one quarter of the production is used for technical purposes such as to obtain raw materials for decorative and protective coatings, for detergents, lubricants and synthetic resins, for plasticizers and precursors for various other chemical syntheses. Fatty oils are stored up often in large amount in seeds (40-60%) and to less extent in other parts of the plant. Fatty oils are produced from many plant species belonging to different families mainly Brassicaceae, Asteraceae, Fabaceae, Euphorbiaceae, Pedaliaceae and Arecaceae.

Oil seeds, their oil and high protein meal products are the most valuable commodity on the world trade. Over the last 30 years the oil and fat industry has increased production threefold (>80 million tons) and this increase has come primarily from plant sources (Rattray 1991). The particular
requirement of the oil and fat industry involve both edible and non-edible products. Oil-meals, the joint product of processing oil seeds, are almost exclusively used for livestock feed, although small but increasing quantities are consumed as processed protein foods for human consumption. Adoption of a variety of technologies will be required if the oil and fat industry is to keep pace with the ever-increasing consumer demand for a higher standard of living. To this end, applications of biotechnology in its several forms will have a major role to play. In particular, combinations of the newer genetic engineering techniques, the use of double haploids developed through microspore and anther culture and breeding procedure with the conventional agricultural practices associated with crop growth are expected to give the required increased productivity and provision of products of uniform desirable properties.

The major objectives that has to be considered in oil crop improvement are: better adaption to different environments, higher yields, higher oil content, changed and improved oil quality, resistance to pests and resistance to pesticides. The increasing importance of tropical and semi-tropical agriculture reflects the greater production of oils and fats that can be obtained from indigenous crops. These indigenous plants of tropical and semi-tropical can capitalize on their potentially high oil productivity as well as being of considerable economic value to the countries located in their climatic zones. Niger, belonging to Asteraceae, is one such plant receiving active consideration (Weiss 1983).

The family Asteraceae:

The Compositae or Asteraceae is one of the largest and most diverse families of flowering plants, comprising one-tenth of all known angiosperm species. The size and adaptive success of the compositae have stimulated
considerable research into its systematics and evolution. It is characterized by the compound inflorescence that has the appearance of a single “composite” flower from which it derives its name. They are predominantly herbaceous, although woody species also exist. The Asteraceae is cosmopolitan, found in diverse habitats; anaerophytic, xerophytic and halophytic specialists thrive in some of the more inhospitable habitats (vertisols, deserts and salt marshes). *Lactuca* and *Helianthus* are both drought tolerant taxa. The oilseed, niger (*Guizotia abyssinica*), is an anaerophyte that grows on waterlogged vertisols in Ethiopia and India, where few other species and no other oilseeds can be grown.

**Economic importance of Asteraceae:**

The Asteraceae (Compositae) contains over 40 economically important species including food (*Helianthus tuberosus* and *Lactuca sativa*), oil (sunflower, safflower and niger), medicinal (*Anthemis nobilis*, *Artemisia* spp.) and many ornamental (daisies, zinnia, chrysanthemum, dahlia, marigold) crops. The high quality edible oils of sunflower, safflower and niger are low in saturated and high in mono and di-unsaturated fatty acids. Additional novel industrial and edible safflower and sunflower oils have been developed (e.g. high oleic sunflower oil). Several novel industrial fatty acids are found in seed oils of composites, e.g. conjugated dienolic fatty acid in *Dimorphotheca*, acetylenic fatty acids in *Crepis*, and epoxy fatty acids in *Vernonia* and *Stokesia* (Smith 1985). The compositae are renowned for the production of a variety of secondary chemicals. The family is a rich source of powerful insecticides and industrial chemicals, e.g. pyrethrum (chrysanthemum) and rubber (*Parthenium argentatum*). Several species are grown as medicinal and culinary herbs. *Echinacea* is a source of biologically active compounds with medical or nutritional benefits. The compositae also includes several detrimental weeds
Let us consider species of the composite family (Taraxacum officinale, Sonchus spp., Ambrosia spp.). Lettuce and sunflower are the best genetically characterized members of this family.

Niger:

Niger [Guizotia abyssinica (L.f.) Cass.] plant belonging to family Asteraceae, native to tropical Africa, has been regarded as a minor oilseed crop in India. It occupies the fifth position in the total production of oilseeds and India accounts for 79% of total niger cultivation and 75% of total niger production in the world (Joshi 1990). Although it has a fatty acid composition typical for seed oils of Asteraceae members, it has been underutilized and highly neglected crop. Although niger has been regarded as a minor oilseed crop as it accounted for very less percentage of export economy in early years, recently it is gaining importance as it is exported to U.S.A., U.K. as birdseed where bird feeding is second only to gardening in popularity. The surprising current level of the niger seed market coupled with high cost of niger seed ($1.10/kg for niger seed compared to $0.26/kg for sunflower) indicates that the market potential is great.

Niger is mainly grown in India and Ethiopia. Weiss (2000) has reported that the production of India and Ethiopia together is estimated to be 318,000-340,000 tonnes. The major niger growing states in India are Andhra Pradesh, Bihar, Karnataka, Madhya Pradesh, Maharashtra, Orissa and West Bengal. In India, about 75% of the niger crop is used for oil extraction. The remainder is exported, as bird feed to U.S.A., Canada, and U.K. Niger oil is bluish-white in colour, with a faint odour and nutty, slightly sweet taste. The fatty acid composition of niger oil is similar to that of sunflower oil except that sunflower oil contains 2% lignoceric acid (Seegler 1983). Linoleic acid content in niger oil is approximately 55% in seed grown in India (Nasirullah et al. 1982). The Niger oil is used for cooking, lighting, anointing, painting, soap making and
cleaning of machinery. Niger oil absorbs flower fragrance making it useful as base oil in perfume (Sharma 1990). Niger meal from India contains high protein (30%) and lower crude fiber (17%) levels than meal from Ethiopia. The oil cake is used as a cattle feed, especially for dairy cows. Niger is grown as intercrop with finger millets, maize and pulses in India. Seed is sown in July or August after the first heavy rains. Niger farmers both in Ethiopia and India, report that the crop helps to reduce weed infestation in subsequent crops. Preliminary work in Ethiopia indicated that a water-soluble extract from niger plants inhibited the germination of monocots. This raises the possibility of selecting for enhanced allelopathic ability in niger to suppress weeds.

Niger is a completely out crossing species with sporophytic incompatibility mechanism (Prasad 1990). Insects, particularly bees are the major agents of pollination. Both improvement of existing cultivars and development of new high yielding cultivars are common goals for breeders of all crops. The self-incompatibility nature of niger complicates the production of self seed and results in low seed yield. When conditions for vegetative growth are favourable, niger is prone to lodging, hence shorter plants with stronger stems and better harvest index are needed (Riley and Belayneh 1989). Testing and selecting plants or accessions from local collections have been the usual method of developing improved varieties. Genetic improvement of niger has been limited mainly due to the crop’s self incompatibility. A sharp increase in oil content could probably be achieved through selection for thin hull types, as was successfully done in sunflower and safflower.

Homozygous lines have the potential for crop improvement. Self-incompatibility of niger hinders the production of homozygous lines, ultimately crop improvement also. Conventional methods of obtaining haploids are not only laborious but also difficulties are encountered during detection because of lower frequency of haploid production. One of the main applications of anther
culture has been to produce diploid homozygous pure lines in a single generation, thus saving many generations of back crossing to reach homozygosity by traditional means or in crops where self-pollination is not possible.

*In vitro* culture studies using cotyledons and seedling explants have been reported in the past (Nikam and Shitole 1993; Sarvesh et al. 1993a) Induction of embryogenesis from cultured anthers and plantlet regeneration has been reported in niger (Sarvesh et al. 1993b; Murthy et al. 2000). Sujatha (1997) has reported *in vitro* adventitious shoot regeneration for effective maintenance of male sterile lines. Ashok Kumar et al., (2000) have reported direct somatic embryogenesis and plantlet regeneration from leaf explants.

Considering all the above factors, the major objective of the present investigation was to establish successful *in vitro* techniques for the production of haploids by anther and microspore culture in niger. The other specific objectives were:

1. To elucidate the effect of auxins and cytokinins individually and in combinations on induction of embryogenesis.
2. To evaluate the effect of temperature pretreatment of capitula on embryogenesis.
3. To study the effect of different carbohydrate sources on embryogenesis.
4. To determine the effect of different amino acids on embryogenesis from cultured anthers.
5. To test the effect of polyamines on embryogenesis from cultured anthers.
6. To study the effect of different concentrations of silver nitrate on embryogenesis.
7. To determine the effect of jasmonic acid on embryogenesis in cultured anthers.
8. To test the effect of different media on embryogenesis from cultured anthers.

9. To study the effect of different genotypes of niger in induction of embryogenesis.

10. To evaluate the effect of cytokinins (BA, KN, TDZ, ADE and 2-ip), amino acids (glutamine, glycine and proline) and polyamines (putrescine and spermidine) on differentiation of embryos.

11. To elucidate the effect of different concentrations of Abscisic acid on maturation of embryos.

12. To successfully acclimatize in vitro raised plantlets and assessment of haploid nature.