APPROVAL SHEET

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Title : Chemical studies on oil constituents of *Polianthes tuberosa* and effect of some growth regulators on the oil constituents.

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بسم الله الرحمن الرحيم

INTRODUCTION

Tuberose (Polianthes tuberosa L.) a member of the family Amarylidaceae of Mexico, cultivated for both ornamental and extractive purposes.

The flowers of tuberose emit a most delightful fragrance, which in itself is a wonderful bouquet and has been compared with the perfume of a well stocked flower garden to evening close in the east. These white tubula flowers emit a most powerful odour which increases after sunset, and on this account is called by the Malays "Misteress of the night".

In despite of the importance of tuberose concrete and absolute to perfumary and its high price, hardly found any tuberose extraction products were manufactured in Egypt, as until, 1963. Now, Egypt export the tuberose oil, besides the flowers. During the next years, it is hoped that these exports will containue to raise.

Pure absolute of extraction of tuberose is perhaps the most expensive natural flower oil at the disposal of the modern perfume. Therefore, it can be used in perfumes of only highest grade.

The volatile oil of single flowered variety of tuberose has been analysed, but no quantitative figures have been puplished. While, there is no any information about the oil constituents of the volatile oil of tuberose double flowered variety.

So this investigation deals primarily with the physical and chemical properties of tuberose oil and chemical composition of such oil. While treatment with growth regulator substances has proved effectively on the regulating of flowering and quality of the oil in many ornamental plants. So, this study was carried, also to evaluate the effect of some growth regulators (GA_3 and CCC) on the quality of tuberose oil, as well as on the quality of tuberose plants.

REVIEW OF LITERATURE

Importance of tuberose :

The horticulture variety of tuberose (*Polianthes tuberosa* L.) exploited in Southern France for the extraction of its perfume was that with single flowers; the double flowered variety usually goes to the cut flower (Mazyer, 1908). The bulk of extraction products of tuberose was produced in Southern France and Lately also in Italy and Morocco (Meunier, 1950 and Gunther, 1952). The tuberose bulbs were planted in April and flowering season started from July to September. The flowers were collected every morning just when they start to open and were pick by hand and finish with the stalk. Each 1000 tuberose plants yield about 25 to 30 kg of flower per year (Gunther, 1952, and Poucher, 1974).

Ashour (1967) mentioned that there were not any tuberose extraction products were manufactured in Egypt as until 1963. The area cultivated with tuberose for extraction occupied just two thirds a faddan in Tanahoub from Qalioubia. In 1965 however, this area was increased to 3 faddans.

El-Gendy, et al (1974) found that the flowering season started about the middle of June and contained through the winter to early of January and the flower yield of the single tuberose variety was higher than that of the double one, also he stated that the yield in Egypt was somewhat higher than in Southern France and the flowering season was longer.

Extraction of volatile oil of tuberose :

Natural perfumes, one of the most marvellous phenomena of plant metabolism, probably reach their highest degree of the excellence in the fragrance exhaled by fresh flowers. This fragrance is due to the minute traces of essential oil which exist in the petals, sometimes in the free state, as in the rose and lavander and occasionally in the form of a glucoside which, under favourable conditions, decomposed in the presence of an enzyme or ferment as in tuberose. Consequentlly methods of extraction were modified according to the location and state of the volatile oils. The enfleurage process was used for the extraction of tuberose oil, but in the present years this process has been partly replaced by the extraction method with organic solvents (Poucher, 1974).

Hess (1903) found that the enfleurage method produced essential oil 13 times more than the organic solvents method. Naves and Mazyer (1939), found that the extraction of tuberose flowers with petroleum ether yielded about 0.08 - 0.11 % of concrete which was light to dark brown, waxy, quite a hard mass and only partly soluble in higher proof alcohol. Gunther (1952) mentioned that the ratio of absolute oil of tuberose to concrete was between 18-23 %, where 1150 kg. of tuberose flowers were required to yield 1 kg. of concrete. Ashour (1967), showed that the yield of concrete of single tuberose flowers in Egypt was between 0.14 and 0.20 % and the ratio of absolute oil to concrete was form 29 % to 34 %. The yield ratios obtained by enflurage and by organic solvents extraction method were 15.34 : 1 for the single flowered variety and 14:1 for the double flowered one. Stulk (1970), mentioned that the 1000 kg. of tuberose flower petals yielded 68 gm. oil. Poucher (197) indicated that 1200 kg. of tuberose flowers produced 1 kg. of concrete which in turn yielded only about 200 gm. of absolute oil.

Physical and Chemical properties of tuberose oil :

Several authers determined the physical and chemical properties of the oil from the different varieties of tuberose. Table (1) refered to the physical and chemical properties of tuberose oil extracted from single flowered variety.

Ashour (1967) reported that properties of volatile oil obtained by steam distillation of absolute of enfleurage for tuberose double flowered variety as follow :

Specific graviety	0.9371 to 0.9506
Refractive index	1.4589 to 1.4700
Acid number	64.3598 to 81.5891
Ester number	109.5504 to 125.0134
Sponification number	171.3349 to 198.8293

Chemical composition of tuberose oil :

Essential oils in general are extremely complex substances incorporating a variety of chemically unrelated compounds belonging to many classes of organic substances. The quality of essential oils depends mainly on their chemical constituents and their levels. It is a well known fact that each essential oil is chracterized by a particular ingredient which plays an important part in determining its quality and subsequently its aromatic value.

The chemical composition of tuberose flower oil was investigated by various workers. Varely (1899), stated that it contained about 10 percent of ketone $C_{13}H_{20}O$ to which he assigned the name tuberone.

Hess (1903) reported that tuberose flower oil always contained benzyl alcohol, either free or in ester form of benzyl benzoate and methyl anthranilate, but methyl salicylate is only found in the oil obtained by enfleurage method. The flower oil of tuberose derived by enfleurage method contained 56 times more methyl anthranilate than the product of extraction method with organic solvents. He also stated that tuberose oil contained butyric acid and phenyl acetic acid.

Schimmel (1903), reported that tuberose oil contained methyl benzoate and methyl anthranilate. Elze (1928), studied the chemical composition of tuberose oil and found that it contained geraniol and nerol, both alcohols in free form or as acetates and probably as propionates also farnesol, benzyl alcohol, methyl benzoate, benzyl benzoate, methyl salicylate, methyl anthranilate, eugenol, butyric acid and phenyl acetic acid were found.

Ashour (1967), studied the chromatographic analysis of concrete oil of single tuberose flowers, and found that the oil contained the following compounds : farnesol, methyl anthranilate and eugenol in amounts below 0.1 %, while methyl benzoate and benzyl benzoate in amounts of 0.8 % and 3.3 %, respectively.

Poucher (1975) summarized the tuberose oil content and he mentioned that the oil has been analysed, but no quanitative figures have been bublished before.

The various chemical constituents of tuberose oil extracted from the flowers of single flower variety plants and their structural formulas are presented in Table (2), as stated by Elze (1928).

Effect of gibberellic acid (GA_3) and Cycocel (CCC) on growth and plant constituents :

Effect of GA_3 :

Winkler (1969) on gladiolus plants, found that GA_3 treatments increased the dry weight of the cormels. Jana and Biswas (1979) on Polianthes tuberosa, L., reported that bulbs soaked in GA_3 at the concentration of 100 ppm yielded more leaves. Whereas, GA_3 treatment at the concentration of 10 ppm produced the highest number of flowering spikes per plant. El-Abd et al. (1980) on potato plants, reported that GA_3 either used at 25 or 50 ppm significantly increased the fresh weight of aerial parts. El-Naggar (1980) on gladiolus cv."Oscar", showed that GA_3 - treated plants at 50 or 100 ppm yielded more fresh weight of both corms and cormels. Refaey (1982) on P. tuberosa, L. indicated that GA_3 treatment at 50 ppm increased the fresh weight of main bulbs and the bulb number increased with increasing GA_3 concentration up to 200 ppm.

On the other hand, El-Leithy (1983) on Ocimum basilicum, L., pointed out that GA_3 at 100 and 200 ppm significantly decreased the fresh weight of the whole plant. Whereas, GA_3 at 100 ppm decreased the plant dry weight. Bhattacharjee (1984) on gladiolus cv. "Friendship", found that GA_3 at 10 and 100 ppm stimulated the vegetative growth, increased the fresh weight of corms and improved corme size. El-Hanfy (1985) on dahlia plants, indicated that the plants treated with GA_3 at 200 ppm resulted in the heaviest fresh and dry weights of both tubers and flowers and the highest number of flowers per plant.

El-Quesny (1977) on gladiolus cv. "Lovely melody", when sprayed the plants with GA_3 at 12.5, 25, 100 and 150 ppm found that the concentrations 25 and 50 ppm were the most effective concentrations of GA_3, which resulted further development of flower primordria, moreover, they gave more vegetative growth and dry weight, due to the increase in the mobelization of polysaccharides. Abou-Zied (1977), mentioned that soluble and total sugars in leaves of <u>Ocimum basilicum</u> L. plants were considerably reduced by GA_3 at 20, 40 and 80 ppm. El-Naggar (1980) on gladiolus cv. "Oscar", showed that reducing sugars of the spikes were significantly decreased as a result of GA_3 treatments, but the polysaccharides content was increased, compared with the control. In most cases, reducing sugars and polysaccharides contents increased by increasing the concentration of GA_3. Towfic (1980) on lemongrass found that carbohydrate content generally decreased as the level of the applied GA_3 increased (75, 100, 125 and 150 ppm). El-Shamy (1982) on carnation, indicated that GA_3 at 50, 100 and 200 ppm decreased the total carbohydrates content of roots, herbs and flowers, at mature stage, whereas, at senescence stage, the GA_3 at 200 ppm greatly increased the total carbohydrates content in both herbs and flowers. Zeid and El-Ghamarawy (1984) on carrot and table beet plants, mentioned that spraying the plants with GA_3 decreased the dry matter accumulation, crude protein, carbohydrates and carotene contents in roots. Whereas, in case of tubers of table beet, GA_3 treatments increased the dry matter accumulation carbohydrates and proteins. El-Hafny (1985), on dahlia, found that the higher concentration of GA_3 (1000 ppm) greatly increased the non-soluble sugars in leaves, stems, flowers and roots than the lower concentration and the control.

Towfic (1980) on lemongrass, indicated that plant phosphorus content, in general, slightly was responsed to GA_3. Increasing the level of GA_3 resulted in a noticable decrease in total nitrogen in the first cut of the first season, while, all treatments had no effect in the second cut. In the second season nitrogen content, in the first cut, increased followed by general decrease in the second cut. Harridy (1982) on <u>Calmintha officinalis</u> cv. "Hepetoides", mentioned that application of GA_3 decreased both nitrogen and phosphorus contents in the plant. While, El-Leithy (1983), found that spraying sweet basil plants with 50 ppm, increased phosphorus content in the leaves, while it decreased potassium in the leaves and stems. But, GA_3 at 100 or 200 ppm decreased nitrogen, phosphorus and potassium contents in the plants, except nitrogen and phosphorus contents in the stems. El-Hanfy (1985) on dahlia pointed that GA_3 at 1000 ppm caused a great increase in the nitrogen content in leaves, stems and flowers, when compared with the lower concentration of GA_3. Application of GA_3 at 100 ppm, showed the lowest value of phosphorus. The untreated plants contained more phosphorus in tubers than those plants treated with GA_3 at 100 ppm. Increase GA_3 concentrations to 1000 ppm, increased the potassium content in leaves, stems, flowers and herbs than 100 ppm treated plants.

Effect of CCC :

On begonia plants, Patakora (1969), revealed that CCC at 0.5 % caused a considerable reduction in tubers yield. Similarly, Tawagen and Hassan (1974) on *Chrysanthmum morifolium* reported that fresh and dry weights of CCC-treated plants were less than untreated ones.

Hassan and Egina (1979) sprayed *Polianthes tuberosa*, L. plants with CCC at 0, 1000, 2000 and 3000 ppm and found that CCC treatment of 2000 ppm significantly affected the fresh weight of leaves whereas, the number of flowering stalks per plant was markedly affected by the highest CCC concentration (3000 ppm). Dry weight of florets was not significantly affected by CCC application. Jana and Biswas (1979) on *P. tuberosa*, L. plants, reported that plants treated with CCC at 2000 ppm produced the maximum number of flower spikes and florets. El-Abd *et al.* (1980) on