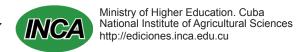
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REVIEW

Polianthes tuberosa L.: A REVIEW OF THEIR PHYLOGENETIC, MORPHOLOGIC AND OF CULTIVATION FEATURES

Revisión bibliográfica Polianthes tuberosa L.: revisión de sus aspectos filogenéticos, morfológicos y de cultivo

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ABSTRACT. The species *Polianthes tuberosa* L., of the family of the agavaceas, presents favorable qualities as cut flowers and ornamental plant, also is appreciated by its medicinal properties and peculiar aroma, from the national and international point of view. However, ignorance still exists on their characteristics and potentialities like cultivated plant. The current paper presents informations of interest about this species and discusses the importance of it multiplication and conservation.

Key words: agavaceae, cut flowers, nardo, ornamental plant, medicinal properties

RESUMEN. La especie *Polianthes tuberosa* L., de la familia de las agaváceas, presenta cualidades favorables como flor cortada y planta ornamental, esta especie también es apreciada por sus propiedades medicinales y su aroma peculiar, tanto desde el punto de vista nacional como internacional. Sin embargo, aún existe desconocimiento sobre sus características y potencialidades como planta cultivada. En el presente trabajo se exponen informaciones de interés sobre esta especie y se discute la importancia de su multiplicación y conservación.

Palabras clave: agavaceae, flor cortada, nardo, plantas ornamentales, propiedades medicinales

INTRODUCTION

The flora is an aesthetic and landscape element of significant environmental service. Particularly, floriculture is the horticultural branch aimed at exploiting flower growth and production. It is stated that since the beginning of humanity, the man felt the need to cultivate plants so as to beautify his environment rather than for his feeding, either for plant erection, attractive flowers, foliage, privacy, protection, its scenic-aesthetic

and socioeconomic value (1). At present, it is known that this sector constitutes a great opportunity for many farmers and has become a large-scale business.

Worldwide flower production started to be driven by the economic development of several countries, highlighting the boom achieved by developed societies, such as Western Europe, North America, Canada and Japan, which today comprise more than 50 % cut flower marketing. There is an annual flower demand of importing countries ranging from 10 to 15 % (2).

In Cuba, producers who have preserved flower crops over time have a small-scale

production. Marketing is very important for some people's sectors, as there are custom dates in the country demanding flowers, according to customers' preferences in terms of type, color and floral arrangements. However, national demands are not yet satisfied, despite our country is rich in botanical species diversity and has a tropical climate characterized by the absence of great temperature differences and high relative humidity, which are appropriate for producing a large amount of cut flower species and ornamental plant (3).

Nevertheless, it should be noted that flower production has increased in recent years (4),

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largely due to Urban Agriculture movement (5).

Polianthes L. genus, which is endemic to Mexico and belongs to Agavaceae family (6), is highly significant from the economic point of view. This genus consists of 15 species (7). Several genus representatives are important at a scientific, cultural and economic level. P. densiflora, P. durangensis, P. elongata, P. howardii, P. longiflora, P. palustris, P. platyphylla, P. graminifolia, P. bicolor, P. geminiflora, P. mexicana, P. gracilis and P. tuberosa may be mentioned among the species of Polianthes L. genus.

P. tuberosa species, commonly known as nard, has been cultivated as ornamental plant for more than 400 years. In addition, it is characterized by a high sapogenin concentration in its rhizomes and tuberous roots; besides, it has essential oils of interest to pharmaceutical and perfumery industry (8, 9). Therefore, it is suggested that, since prehistoric times, this species has been used for medicinal, ornamental and ceremonial purposes (10), as well as in various traditional rituals.

At present, bulb demand of this species has increased in various parts of the world. In Cuba, P. tuberosa is used in floral arrangements and appreciated for its unique aroma. However, it is still necessary to get more information for specialists and producers, in order to ensure a proper management, increase production culture and evaluate all considerations on the possible claims related to this species, as an attractive cut flower and ornamental plant. as well as to extract essential oils for industrial purposes, which would allow, in some way, to enhance our economic development and meet people's interests with high quality.

BOTANICAL FEATURES

Some aspects of P. tuberosa species related to Systematics are described below (11, 12): Kingdom: Vegetal Division: Esparmaphytes Subdivision: Angiosperms Class: Monocots Order: Asparagales Family: Agavaceae Genus: Polianthes Species: Polianthes tuberosa L.

A marilly daceae and Liliaceae families were joined together, as well as Agavaceae recognized as an independent family in 1988 (13). It is noted that when the latter family was studied, its delimiting criteria were very diverse, due to the convergence and parallelism processes of family phylogeny^A, which has led to various studies and reflections on its validity.

Agavaceae family is endemic to America and spread from the boundaries between Canada-USA and Bolivia, including Caribbean islands. This family has nine genera and 330 species. The center of its origin and diversity is found in Mexico, where there are 251 species, 76 % out of those described in the world and 177 of them are endemic, corresponding to 70 %. The members of this family reach higher growth at 1500-2000 m above sea level (14).

So as to complement the taxonomic studies of Agavaceae family, different morphological and anatomical analyses have been developed, including

Agavaceae family is important from the economic viewpoint for different worldwide regions, as it comprises diverse plant species domesticated by pre-Columbian cultures of Mexico. This family includes Agave genus, whose species are recognized for providing leaves with high concentrations of sapogenins, terpenes, sterols and vitamins; consequently, several species of this genus are applied to soap production and medicinal purposes (16). Other species included in this genus have gained great significance when cultivated to obtain alcoholic distillation, such as tequila and mezcal. Moreover, they provide high strength fibers with durability employed in crafts, fishery networks and ropemaking, among others (17).

Other genera are found within this important family: Cordyline, Dracaena, Sansevieria and Yucca, which are species with attractive foliage used as ornamental plants.

Agavaceae family also includes *Polianthes* genus (11,18), supported by cytological and cellular studies in different genera and species, where five big chromosomes and 25 small ones have been registered, for n=30 (Yucca-Agave complex) (19-23). It is valid to note that this genus was initially

reproductive and vegetative structures, karyotype studies, molecular and phylogenetic analyses^B. It is one of the 10 monocot families with greater species richness in the state of Oaxaca, Mexico (15).

A Hernández, S.L. "Análisis filogenético de Agavaceae y Nolinaceae con base en su morfología". *Primer Simposio Internacional sobre Agaváceas*, edit. Instituto de Biología-Universidad Nacional Autónoma de México, México, 1994.

B Solano, C. E. Sistemática del género Polianthes L. (Agavaceae). Tesis de Doctorado, Universidad Nacional Autónoma de México, México. 2000.

included into Amarillydaceae family, but results of karyotype studies on *Polianthes* L. genus, specifically *Polianthes tuberosa* L. (nard), allowed to corroborate that Agavaceae is a different family (24).

POLIANTHES L. GENUS

Polianthes L. genus is endemic to Mexico and very important from the economic, scientific and cultural point of view^B. With respect to the term Polianthes, its Greek meaning is "many flowers". This genus gathers about 15 especies B (7). P. densiflora, P. durangensis, P. elongata, P. howardii, P. longiflora, P. palustris, P. platyphylla, P. graminifolia, P. bicolor, P. geminiflora, P. mexicana, P. gracilis and P. tuberosa may be mentioned among its species.

Similar to other genera of Agavaceae family, there are some difficulties when delimiting species of this genus, which is also poorly represented in herbarium^B. Certain studies have detected that this taxon, like many others of restricted distribution, is sensitive to disorders in its natural habitat; consequently, it is prone to be extinguished (25), a problem currently affecting various crops.

In this sense, these authors point out that farming and cattle activities are the main causes of natural habitat changes, which make essential a continuous checking of species preservation status (25) that seriously endangers *Polianthes* populations, because its corms and bulbs are vulnerable to

cattle trampling and cultural practices (26).

These reasons have led to develop important researches, in order to reevaluate the extinction risk of five species belonging to Polianthes L. genus (P. densiflora, P. howardii, P. longiflora, P. platyphylla and P. palustris) and following the results, some strategies proposed preserve these vulnerable species (25). Likewise, the authors considered the need to combine different alternatives, that is, in situ and ex situ preservation practices in these species, ensuring greater success to complete the tasks conceived through biodiversity preserving programs (27-29).

So as to succeed in preserving and using plant genetic resources, the role of very diverse persons is also required at each site of action, including those in charge of germplasm, breeders, scientists, farmers and their communities, as well as decision-makers (25,30).

In Cuba, like in other nations, Polianthes tuberosa L. is the most renowned species of Polianthes L. genus, which is used in floral arrangements and its aroma is highly appreciated with very special characteristics (31). Likewise, some authors have referred to the extraction of its useful compounds, such as steroidal sapogenins, largely applied to industry (9, 32-34).

ORIGIN AND GEOGRAPHICAL DISTRIBUTION

Polianthes tuberosanard, from the Latin "nardus", is originally from Sonora

desert in Mexico (35). Aztecs called it Omixochitl or bone flower. Endemisms are among the most important features of Mexican biological diversity (36); it has been estimated that from 20 to 30 % species are endemic and the highest floristic richness is found in wet areas, while the greatest percentages of endemic species are recorded in those floras where desert scrubs and grasslands are predominant of dry areas, as Tehuacan in Puebla, as well as Chihuahuense and Sonorense deserts (35).

Although some authors suggest African and South American origins for *P. tuberosa* (12), it is known that Mexico has a great biological diversity, due to its geographical location and different climates, so that it can provide more than 40 ornamental species worldwide (36).

Moreover, it is important to mention that nard, as a species, was firstly classified by Linnaeus in 1737 in his book entitled "Genera Plantarum" with a generic name of Polianthes. This crop was introduced in Europe by travelers from the Middle Ages in 1629, who generally moved planting material appropriately adapted under Mediterranean climate to royal gardens. Furthermore, P. tuberosa was widely cultivated in southern Italy (Sicily), French "Midi" and Iberian Peninsula, particularly in the southeast (Valencian community). According to literature, large producing centers of this esteemed cut flower are located in southern USA (12).

Regarding ornamental species in Cuba, it is stated that the first commercial gardens appeared in Havana about the middle of the XIX century, mainly dedicated to grow roses and ornamental plants (37). At the beginning of the XX century, when our population increased, the number of gardens surpassed 650 in this city. In this sense, literature indicates that the first germplasms of these species were introduced from USA in 1914. Bulbs and tubers started to be imported from countries like the Netherlands and Belgium in 1920 (3).

However, it is valid to note the results of some diagnoses made to P. tuberosa in different provinces of Cuba at present, which enable to corroborate that this species is not described as a cut flower produced for commercial purposes. Due to its potentialities, P. tuberosa can be an effective choice within the production field of cut flowers and ornamental plants in our nation, so that it would enable to give more offers to people and promote an increased species diversity of plants used with such aim^c.

BOTANICAL DESCRIPTION

Polianthes tuberosa L. is described as a perennial herbaceous tuberous plant, commonly known as nard, whet, tuberose, white tuberose, night friend, lilies and goldenrod. It is considered a cultivated species within Polianthes genus.

The following aspects are among the main features of this species:

Bulb: It is the reproductive organ with an oval-conic shape and a tunic. The bulb consists of a "basal disc" and a floral stem emerges from its apex. This structure stores reserves for reproduction (38), for instance carbohydrates (39), which are located inside bulbar flakes, which are usually very fleshy. This structure is called rhizome and it is the subsistence organ. Bulb weight varies according to its diameter; they are brown and a small disc or rhizome is located at its base that gives rise to short, abundant and rounded roots (12).

Leaf: They are sessile, fleshy, elongated, linear and narrow; in general, they are light green and reach up to 60 cm long and 3,5 cm wide. Basal leaves are bright green and lanceolate, 30-45 cm long with 8 to 12 reduced cauline leaves. They are gathered at the base of flower stems as flakes arched outwards (12).

Studies related to foliar anatomy of this species show that the open leaf profile, in cross section on immature leaves, has U-shape, coinciding with senescence state^B. However, that profile is straight on mature leaves. Furthermore, the epidermis is made up by a cell layer (mono-stratified); such cells may be square, rectangular or partially spherical^B.

With regard to cell configuration of stomatal complexes, it is parasitic, that is, with one or more attached cells lining stomata and arranged parallel to guard cells. Palisade parenchyma cells are poorly developed towards adaxial surface and virtually absent towards leaf underside, because its development is extremely poor^B.

Flower stem and inflorescence: Its stem is erect, reaching 50-100 cm high and has a flower spike at its end; it is bright green, round and its base is compact. It is characterized by small light green leaves. Aerial stems can reach up to 180 cm high (40). Inflorescence is a spike measuring 30-60 cm long and the number of flowers varies from 8 to 20. Flowers are larger at the base of inflorescence and smaller when approaching the apex (41).

Flowers: They are white with light pink tones (41, 42), very fragrant and waxy. They are small, 3-6 cm long, sessile with extended segments and curved tube, which usually occupy more than a third portion of stem and are arranged in spikes at its end. Flowers appear in pairs and emerge from a bract; they have a zig-zag-shape and open from the base to the apex (43).

Flowers are long-lasting in Polianthes tuberosa L., which is a profitable aspect (38). They are hermaphroditic and regular; their funnel-shaped corolline perigonium with a curved and long tube near the base divided into six lacinia. Stamens are included and joined at the central part of perigonic tube. A tricarpelar and trilocular ovary with three free stigmas at the apex; it is inferum with six tepals sprouting from its top.

González, M. E.; Ramírez, M. A.; Valcárcel, M.; Castilla, Y.; Rondón, L.; Suárez, A. y Lago, E. "El nardo (*Polianthes tuberosa*), atractiva flor de corte: control de la antracnosis con el empleo de la quitosana". En: *Primer Congreso Cubano de Horticultura*, edit. Instituto de Investigaciones Hortícolas, La Habana, Cuba, 2012.

Aroma: P. tuberosa is one of the most aromatic floral species. Its aroma is very peculiar (31), dense, pungent and sweet, due to the presence of indole as a major component of its essential oils, also accompanied by benzyl-acetate and methyl-autranilate (44).

Fruit: It is a loculicide capsule provided with three valves: it usually contains a high number of deltoid or semicircular and flattenshaped endospermic seeds. Micro-morphological and anatomical studies have shown that seeds lack starch and have exotesta with primary and secondary reticulation, cylindrical linear embryo and endotesta surrounded by the oily endosperm. Their black color is attributed to phytomelanes in the epidermis (45).

VARIETIES

P. tuberosa species is characterized by poor genetic variability; it is vegetatively propagated in absence of genetic recombination. It is generally multiplied by tubers (38), which, in turn, make its breeding tasks difficult. Varietal differences are mainly caused by spike and inflorescence length, flower opening shape and its degree of prematurity, among other things.

"Pearl" is the cut flower variety known of this species, which has a complete inflorescence with white flowers and double perigonium as well as a stem 80-160 cm long. It is widely grown in Mexico, USA, Argentina, Spain, India and Iran (12, 40, 45).

Some *P. tuberosa* genotypes are called dwarf or creeping, as for instance "dwarf Pearl" variety, which is characterized by a compact spike, very large, double and resistant flowers with stems 60-80 cm long.

Some varieties of this species are "Double excelsior", "Tall double", "Florentiu", "Orange flower", among others. There is also a form called *P. tuberosa* f. plena^B and some heterozygous hybrids of different colors (red, pink and yellow) mainly in South America. However, several hybrids can be found on the plains of India at present (39, 40).

It is also valid to emphasize the usefulness of P. tuberosa as an ornamental pot plant; an example is the case of "Gracilis" variety. In general, different authors suggest the importance to keep on developing studies and breeding programs that lead to obtain new varieties of this crop, in order to get plants with flowers of various shapes and colors, since they are interesting from the commercial viewpoint (45, 46). It is also important to mention that P. tuberosa cultivars of single flowers have been generally relegated by those with double flowers, for their beauty and attractiveness.

BREEDING ASPECTS

Plant species asexually reproduced as *P. tuberosa* are characterized by low genetic variability; however, different crop results make

evident that it can be induced by physical and chemical mutagens (46-49). Thus, it is feasible to apply ionizing radiations like X-rays, gamma rays and neutrons, chemical mutagens such as ethyl methane sulfonate (EMS), methyl methane sulfonate (MMS) and hydrogen fluoride (HF), among others (50-52).

Breeding performed in P. tuberosa has been very poor compared with other crops and it is usually aimed at obtaining a strong, erect stem of considerable length, inflorescence with abundant flowers and wilt tolerance, as well as an adequate spike arrangement. Other aspects of interest for breeding consist of inducing new flower colors. shapes and sizes, from the commercial point of view (44). Moreover, investigations are focused on the search for growth, crop and flowering uniformity of cultivars, as it enables management and harvesting practices, which is a highly valuable selection criterion for producers.

Nowadays, there is another aspect of great relevance against climatic change effects that is related to the search for resistance or tolerance to adverse environmental conditions (53), as pest (insects, fungal pathogens, viruses, etc.) damages (54). It is interesting in P. tuberosa, because it is highly susceptible to pests, despite being a short-cycle species and it is strongly affected by several pathogens that influence negatively on its development and limit both its beauty and production.

Multiplication organs as the corm (9, 55, 56), flower bud (42), apex segments (57) and leaf segments (41) can be included within the planting material used for inducing mutations in vegetatively-propagated plants; however, it is known that they are multicellular organs, that make the development of new genotypes more complex, that is, they restrict to obtain mutated sectors to some extent (45).

Likewise, there may be some injure caused by toxic effects of chemical agents used for such purposes, which makes difficult its homogeneous application. Therefore, it is considered that ionizing radiations have strong penetration power as gamma and X-rays, which is an adequate choice for inducing mutations in vegetatively propagated plants with the aim of obtaining improved genotypes (58).

Plant biotechnology provides very useful tools that enable a better plant use; in this sense, tissue culture techniques have proved to be effective for spreading various plant species; they allow cell, tissue and organ growth isolated from the parent plant and are based on cell totipotentiality (59). These micropropagation techniques are generally superior to conventional vegetative propagation methods, since they give rise to high multiplication rates, guarantee pathogenfree material production and reduce the space required to multiply a large number of plants (60).

Some authors emphasize the significance of combining different techniques to improve biodiversity; an example is mutation induction to increase crop genetic variability (46, 61) and *in vitro* culture techniques to ensure the quick multiplication of selected genotypes (41, 45, 62). As it is known, the use of *in vitro* culture overcomes certain limitations to apply this technique and altogether can accelerate breeding programs (59).

Therefore, given all these advantages, there is an international interest in promoting the development of in vitro technologies that enable spreading and breeding of plant species with commercial values, among them those of nutritive and industrial purposes, medicinal and ornamental properties (40, 63). In addition, endangered species could be prioritized in terms of biodiversity preservation (64, 65), because they enable germplasm preservation and exchange, as well as studies related to molecular biology (66).

While developing different studies aimed at obtaining genetic variability in this crop, some authors subjected P. tuberosa planting material (tubers and in vitro plants) to varying doses of 60Co gamma radiation (0, 5, 10, 15, 20, 25 and 30 Gy) and obtained that as radiation dose increased, plant development decreased, either those from adapted outbreaks or the ones obtained from in vivo established tubers. Irradiated plants showed greater leaf length and width variation than non-irradiated plants. Moreover, LD₅₀ of plants from outbreaks (9,09 Gy) was lower than LD₅₀ of in vivo established tubers (25,91 Gy) (45).

Researches on this approach are quite valuable when considering the basic principle for physical and chemical mutagenic agents applied *in vitro*, which refers that more attention should be given to "efficiency", looking for doses in which only one or a few mutations occur per plant, to ensure the commercial value of desired mutants (62).

There is little information on P. tuberosa researches applying novel molecular biology techniques. However, some authors report studies related to the analysis of crop genetic diversity, who also highlight the importance of Random Amplified Polymorphic DNA (RAPD) and Simple Sequence Repeat (SSR) markers, when evaluating ten cultivars of this species, which revealed 53 and 73 % polymorphism, respectively. This characterization is interesting, because these materials can be used as reference patterns in tasks related to a quick crop identification and breeding (67, 68).

PROPAGATION

The most used propagating method is generally asexual in *Polianthes* L. genus and particularly in *P. tuberosa* species (12). Seed reproduction is mainly used to get new varieties or for very specific studies (44).

It takes a long time before the plant blooms when this species is propagated by seeds; in addition, the offspring is dispersed due to genetic recombination, often leading to lose desirable characteristics present in the donor material. These reasons help explain why crop

producers and fans prefer agamic reproduction (12).

Some authors have reported that, from a simple bulb collection of *P. tuberosa*, they succeeded in establishing germplasm multiplication for a period of two years, until reaching flowering and seed of good quality, with the aim of starting to develop a research line in this ornamental crop breeding (44). These authors report that, in Argentina, there are not any wild examples of this crop through agamic propagation.

In general, this species is multiplied by bulbs or bulblets surrounding the mother bulb, which grow, after one-year period or more, on a soil with a high percentage of clay and sand, abundant organic matter and pH between 5,6 and 7,0 and acquire appropriate characteristics, in terms of size, giving rise to a flower of good quality (12). P. tuberosa is a perennial but monocarpic species that blooms only once within its cycle; thus, bulb fattening period last from one to four years.

In the case of bulb storage, during its fattening period, it is necessary to intensify care to avoid pest damage, with emphasis on Thrips and red spider, and thereby to preserve its quality (69).

Lateral bulblets of *P. tuberosa* are derived from "mother" bulb, which are characterized by presenting diverse sizes and classified according to each size (70, 71). It is stated that the largest bulb in one season will be ready to bloom and belongs to the "first force in bloom" category, which in

turn is an indicator that not all of them are ready for it. Meanwhile, sizes of 8-10 and 10-12 do not bloom the same year they are obtained, but the next year. It is important to know that the smallest bulblet requires from three to four years to complete the "fattening" process or appropriate level of thickening. Therefore, it is convenient to obtain large and new bulbs to ensure a profitable plantation.

In this sense, some authors report that when evaluating flower production and quality indexes in "Pearl" bulbs of 9-10 and 10-12 cm diameter for four growing cycles and with subcultures every 30 days, the highest productivity was achieved by bulbs of greater diameter; thereby values of 83, 82, 75 and 65 % were respectively obtained. Moreover, their flowers had an adequate quality (70).

Other authors evaluated bulb size effect on some growth indicators, flowering and bulblet production in the "Single" cultivar of tuberosa under agroecological conditions (71). Results showed that larger bulbs gave rise to more vigorous growing plants, as well as increased bulblet number and production, compared with smaller bulbs. Its biggest bulbs had 3-4 cm diameter, so that it is smaller than the optimal for "Pearl" variety (70).

Several authors have used the benefits of these biotechnological methods on *P. tuberosa* and obtained satisfactory results for all indicators evaluated^D (45).

For example, tubers without any mechanical damage and irradiated with ⁶⁰Co in studies related to genetic variability induction were established under *in vitro* conditions in a Murashige and Skoog (MS) culture medium (72) with 1 mg L⁻¹ benzyladenine (BA) and 0,5 mg L⁻¹ naphthalene-acetic acid (NAA).

In this case, results showed that *in vitro* plantlets from nard outbreaks presented a lower LD $_{50}$ (9.09 Gy) than LD $_{50}$ of *in vivo* established tubers (25,91 Gy) (45). One advantage in this type of study is that when working with *in vitro* culture plants, only a small portion of plant tissue is irradiated, which involves using very low mutagen doses (67).

The massive regeneration of P. tuberosa plantlets from corm, leaf, flower and bud is also reported in a modified MS basal culture medium (72) containing 50 mL L-1 coconut water, 6,4 g L-1 agar and pH adjusted to 5,7, in order to prepare different variants of culture media supplemented with 6-benzyl-amino-purine (6-BAP), naphthaleneacetic acid (NAA), 2,4-dichlorophenoxyacetic acid (2,4-D), indole-acetic acid (IAA), thi-dia-zuron (TDZ) and kinetin as well as sucrose (10, 20 and 30 g L⁻¹). Results enable to confirm the feasibility of using these techniques in P. tuberosa for the massive regeneration of in vitro plants and the rescue of somatic mutants, with the purpose of increasing crop biodiversity^D (73).

D Hernández, M. F. Caracterización de las condiciones para la micropropagación de Polianthes tuberosa L. a partir de cormo, hoja, flor y yema. Tesis de Maestría, Colegio de Postgraduados, México, 2013.

Similarly, different studies reported the use of these novel techniques to obtain compounds of interest, for example, the synthesis of extracellular polysaccharides in *P. tuberosa* callus (74, 75).

CULTURAL ASPECTS

Planting *P. tuberosa* bulbs is advisable within spring season, although some authors argue that they can be practically planted throughout the whole year, although it does not guarantee to have flowers continuously (12). Anyway, rows should be separated 50-60 cm from each other and it is recommended to plant 20000-40000 bulblets per 1000 m² depending on their size.

Another choice is to plant 16 to 25 bulbs per net m² in 1-m-wide blank spaces; plant density should not be very compact, since this species is plenty of foliage and there are results showing its influence on bulblet thickening level (76). In addition, it is necessary to ensure bulb disinfection before planting, in order to reduce pollution load.

Altitude is another factor to be considered in *P. tuberosa* and some authors emphasize its importance when selecting a planting area, as they relate its effect on anthocyanin content in "77A05" cultivar growing at different meters above sea level in Taiwan (77). It is stated that the highest crop growth was attained at 1500-2000 m above sea level (14).

The soil for growing this species should be sandy or sandy loam and rich in potassium. Planting depth may range from 5 to 10 cm; the

bulb is put with its rizogenous area towards the soil and the apex slightly buried. Mineral fertilization is recommended at crop beginning, employing a complete NPK formula at the rate of 2:1:3. It is favorable to increase potassium content in the so-called bulb thickening phase, as it enables its adequate development.

In some researches evaluating substrate effect on the vegetative growth and floral characteristics of P. tuberosa, different soil and poultry manure combinations were used. Results indicate the positive effect of this organic matter on most indicators evaluated, increasing flower stem length, fresh weight and diameter, flower diameter, bulblet and sprout fresh weight, dry weight and diameter, chlorophyll content and flower number per inflorescence (78).

This favorable behavior is attributed to animal manure, which usually contains essential microelements and macroelements for plant growth (79). However, in this case, flower length, leaf area, root fresh weight and dry weight were not influenced by hen dung. The authors emphasize the need of further studies to explain the mechanism of action of this organic matter on *P. tuberosa*.

Likewise, when studying the effect of Azotobacter and some organic manure on a double cultivar of *P. tuberosa*, the authors found that they had a positive influence on its flower characteristics (80).

Similarly, some researchers evaluated the effect of different levels of paclobutrazol (50, 100 and 200 ppm) applied three

times (60, 70 and 80 days) on growth, flowering and bulb production in a double cultivar of *P. tuberosa* after its plantation. The best results were obtained when applying 200 ppm paclobutrazol at 70 days of culture (81).

Moreover, when assessing bulb production, it has been informed about the profitable effects of applying Perlite (30 %) on some indicators as flower number of larger bulbs, total number of bulblets per plant, root development and significant bulb growth increase, when compared to traditional crop growth (82).

In a study aimed to learn about the effect of various substrates prepared with cow, goat, horse and hen dung applied at different doses (0, 100, 200 and 300 g manure per bag) on flower growth and production of P. tuberosa, there was not any interaction between substrates and doses for the variables evaluated. The type of substrate did not affect the variables under study; however, the authors emphasize that doses did have influence on the indicators analyzed, except stem length and flower number. In this sense, the best dose was 200 g manure applied twice a year (83).

With regard to light requirements, it is said that *P. tuberosa* bulbs should be planted in sunny areas, where they can reach a profuse growth in summer season. It is known that light is necessary for photosynthesis, which is one of the most important plant processes. According to light composition, intensity or lengthening will be its influence on species development (84).

With regard to temperature, P. tuberosa is a demanding crop of optimal values between 20 and 30 °C during the day and from 15 to 20 °C are more favorable at night. Similarly, it requires high soil temperature, which should range between 17 and 20 °C. This species demands light, so that floral abortions occur when it is not appropriate. Concerning relative humidity, values between 60 and 70 % are optimal; then, flowers are not efficiently developed when environmental humidity is very low.

Irrigation of *P. tuberosa* should be moderate throughout the year, except in summer season and during flowering, when more abundant water is recommended for the species, which is characterized by slow growth. In this sense, sprinkler irrigation enhances plant development, as it prevents soil compaction.

FLOWERING

Plants providing cut flowers have great economic value, due to their significant amount sold in the market (76). Thus, many specialists and producers are constantly investigating in order to obtain hybrids of diverse colors, more intense scents and highly resistant to the passage of time once they have been cut (44). In this regard, P. tuberosaB flowers are among the most commonly employed to make bouquets and decorate vases or centerpieces, due to its particular beauty and fragrance.

P. tuberosa blooms when there is high solar radiation, as its flowers are very susceptible

to low temperatures. The spike usually lasts more than 40 days ready to be cut; it is an advantage compared to other cut flower species. Although plants have long flower stems, they do not require stakes, because stems are rigid and usually very strong.

In searching for endogenous cytokinin content in *P. tuberosa*, some authors observe significant zeatine and dihydroxizeatine increases at flowering time. This behavior differs from that observed during its vegetative phase development, which indicates the role of such growth regulators on the above mentioned stage (85).

P. tuberosa flowers are usually collected when the first three flower pairs of inflorescence are opened, which is a two-to-three week period in a normal crop. It is advisable to collect flowers early in the morning. They should be cut with scissors at its base without leaves. since the latter must be kept to complete the multiplication cycle of new bulbs that are commonly extracted from the soil when temperature is lower. This species lasts in the vase from seven to ten days (38).

A notable aspect in P. tuberosa is that it is sensible to ethylene (86), since its high concentrations reduce flower opening, although its effects are not probably important from the commercial viewpoint.

In studies aimed at obtaining P. tuberosa plants of suitable size without reducing floral stem quality, the effect of daminozide (2,5 and

5 g L⁻¹) and chlormequat (5 and 10 g L⁻¹) compounds was evaluated during bulb development. Results showed that both products at their highest concentration reduced the period for obtaining plants in ten days, when compared to the control treatment and decreased its length to 15 cm (86).

P. tuberosa flowers are classified into three categories: extra, I and II (12).

Extra category: top quality flowers, pathogen free, with neither malformation nor bruise and very rigid stem, with more than 14 pairs of flowers that must be over 100 cm long.

Category I: good quality whole flowers, pathogen free, with neither malformation nor bruise that are grouped together and may have slight features of foreign materials without affecting its appearance, very rigid stem with a minimum of ten pairs of flowers that must be over 80 cm long.

Category II: it includes flowers that do not enter into the above categories; they may have slight malformation or bruise and some pathogenic damage with a minimum of eight pairs of flowers that must be over 60 cm long.

In general, it is recommended that once *P. tuberosa* flowers are cut, they should be placed in water and it is convenient to add some floral preservative to lengthen their vase life. In this regard, a significant spike life lengthening in the vase and better flower bud opening has been observed in flowers of

this species preserved for 24 hours at room temperature and in a sugar solution (20 % sucrose). Other compounds used for the purpose of preservation are silver thiosulfate, silver nitrate and citric acid, the latter being very effective at concentrations of 350 mg L^{-1} (12).

SOME PEST DAMAGES AND ITS CONTROL

Ornamental and cut flower plants are exposed to pest attacks, which have a negative influence on their development, limiting both beauty and production. P. tuberosa is vegetatively propagated (38) and is sensible to some pest attacks, which is in correspondence with its growing season, in this case summer; although it is a short-term species (three months and a half), it is strongly affected by typical pathogens at this season of the year.

Among harmful insects to this species are aphids (Aphis sp.), both green and black, which cause leaf curl and deformation, since they parasitize its underside and may also affect inflorescence. It can be controlled by applying Aldicarb to the soil or spraying acephate, pirimicarb or pyrethrins. Meanwhile, Thrips (Frankliniella occidentalis, Pergande), which causes severe damage, either as larval or adult stage, is able to fly easily, affects floral stem, inflorescence and flowers at any time of the year. It can be controlled by combining cultural practices and chemical control with acephate, acephate plus malathion, endosulfan, endosulfan plus pyrethrin, among others (87).

Red spider (Tetranychus spp.) mainly attacks leaf fold. so that affected leaves lose vigor and get a yellowishwhite to reddish tone. This pest also attacks flower spike and it is favored by extended drought periods or insufficient water supply, associated with high temperatures (88). It can be chemically controlled by Vertimec and Mitac. In the case of whitefly (Trialeurodes vaporarorium, West.), it attacks seedlings outdoors and affects leaf underside, provoking plant weakening. It can be chemically controlled by Lannate and Applaud.

Aqave weevil Scyphophorus acupunctatus Gyll., also known as "henequen max", in southeastern Mexico and "black weevil" or "nard acapiche" is associated with several economically important plant species from Agavaceae family. This insect species is of American origin and cosmopolitan distribution at present, which has increased due to the international agave trade for ornamental purposes and to obtain fibers (69).

S. acupunctatus species causes severe damage to P. tuberosa plantations (89) by drilling plant tissues. This insect is controlled by chemicals mainly directed to the adult, since it is the most susceptible stage. However, some biological control agents, such as entomopathogenic fungi based on different formulations of Beauveria bassiana (Balsam) Vuellemin and Metarhizium anisopliae (Metschnikoff) Sorokin, are currently applied as an alternative and obtained encouraging results that are consistent with other methods

among agroecological pest management programs (90).

Referred t o S acupunctatus control, it has also been reported on studies related to the use of pheromonal bait to capture this insect. Investigations are based on the fact that males of this species release an aggregation pheromone that attracts both sexes, which is composed of two alcohols and two ketones. Results show that the greatest captures were obtained by combining 2-methyl-4heptanone and 2-methyl-4octanone. Moreover, different pheromonal bait rates were evaluated in field trials without affecting insect-capturing process (91).

Rootand neck rotare among the main diseases affecting *P. tuberosa*, which are mostly caused by *Phytophthora parasitica* Dastur, causing plant wilting and yellowing, whereas neck area and root tip become necrotic (92), which leads to vascular system discoloration. This plant pathogen makes outer leaves dry and fall off. It is controlled through soil disinfection and applying Ridomil solutions during irrigation.

Some P. tuberosa leaf stains and mottling are caused by different microorganisms, such as Ramularia primulae (Thum); spots are light yellow on top and white on the undersides that later turn into reddish-brown, while mottling caused by Phillosticta primulícula (Thum) is light ocher with black dots. Anthracnose, blackish spots caused by Gloeosporium and Colletotrichum (93), a filamentous fungus, also affect leaves of this species.

In the case of Colletotrichum, specialists agree that it is one of the most common and important fungal pathogens, causing a significant group of diseases (94); it ranks eight within the 10 most severe plant pathogens (95) that can be controlled by applying Captan, Zineb and Copper oxychloride.

Nevertheless, the high occurrence of a disease caused by Colletotrichum gloeosporioides in P. tuberosa suggests that the chemical treatment with fungicides alone is not entirely effective to control anthracnose; thus, it is necessary to conduct new studies in the search for other more environmentally friendly alternatives. Some authors reported the antifungal activity of certain essential oils of this plant species against such pathogen, who observed a mycelial growth decrease and total inhibition of C. gloeosporioides in some treatments evaluated with dextrose potato agar medium (96).

Meanwhile, researchers from the National Institute of Agricultural Sciences (INCA) of Cuba remark the effect of chitosan, a bioactive product that works as an activator of innate plant resistance and physiological conditions, to in vitro and in vivo control C. gloeosporioides in P. tuberosa^c. Results show a pathogen development decrease that reach the highest values with 50 mg L-1, attributable to the inhibitory properties of such chitin deacetylate derivative. In vivo results confirm the antifungal potential of this bioproduct, a nontoxic and biodegradable

polymer, to control such disease. This active ingredient is a viable choice to obtain cut flower, from the economic and environmental viewpoint. On the other hand, in 2003, scholars from New Delhi, India, observed a new disease called rust in P. tuberosa and analyzed its symptoms and pathogenicity; then, they proved the causal agent is Colletotrichum capsici Penz (97), which confirms the fact that Colletotrichum genus is the causative agent of a significant number of plant diseases (94).

Botrytis cinerea Pers: Fr affects P. tuberosa leaves and inflorescences when there is high humidity, causing gray mold, which is controlled by Bennlate and Ronilan, whereas Fusarium oxysporum affects roots severely (98). Similarly, Curvularia sp. and Alternaria sp. fungi that cause leaf blights have been isolated (99). Aster yellow virus may also damage this crop, as well as other kinds of mosaic viruses. Recently, molecular and serological methods allowed detecting a pot virus in this species affecting leaves and peduncles, which can be mechanically transmitted or by Myzus persicae (100).

In general, it is suggested that this damage can be only controlled by destroying affected plants (91, 95).

With regard to nematode injury, it is important to note that *Meloidogyne incognita* is another plant pathogen causing severe effects. In this sense, an experiment was carried out under field conditions, so as to get an integrated nematode management through *Paecilomyces lilacinus* as a

fungal antagonist combined with Castor plant and Nim tree extracts; the latter combined with *P. lilacinus* was more effective on fresh weight and flower production (101).

Also, some authors report that the successful growth of *P. tuberosa* has been affected in Ranaghat, Haringhata and Panskura towns, some West Bengal areas of India (102), due to *Aphelenchoides besseyi* nematode, which has become a serious problem to this species and others of ornamental interest in that nation (103).

In general, it is valid to emphasize the need to deepen on the study of pests affecting P. tuberosa and particularly important to consider the new damaging ways emerged in recent years. In this sense, it is convenient to apply integrated pest management practices in this species by combining diverse methods and adapting them to local crop conditions, with the aim of ensuring an adequate phytosanitary status by the friendliest possible manner with environment.

PROFITABILITY AND UTILITY

P. tuberosa species is considered a perennial plant with valuable extracts, which is characterized by the presence of numerous ergastic substances found in mesophyll cells containing tannins, raphids and styloids. In addition, its roots are characterized by a high sapogenin concentration and also have essential oils of interest to perfumery industry (7), to make incenses and medicines (86, 104).

Flowers are structures that have been deeply studied and produce a wide range of volatile compounds, for example, essential oils that are important for perfumery in countries like India, Iran and Mexico. Other authors emphasize the value of different metabolites of this species with bioactivity, which is associated to its fungicidal effect at very low concentrations and gives great significance for their possible synthesis or analogues to control fungal diseases of interest (102).

P. tuberosa plants have a remarkable place in Indian culture and mythology. Its flowers are used in wedding ceremonies, garlands, decoration and various traditional rituals. In Mexico, it represents an important source of employment and income for farmers in several states and localities. It is also traditionally used in Hawaii to create necklaces and it was considered a funeral flower at Victorian times (2, 12, 45).

In Cuba, just as in other countries, *P. tuberosa* is used as an ornamental species and mainly in floral arrangements, which is highly valued for its particular scent. Furthermore, different authors highlight the possibility of extracting important volatile compounds useful for the industry (32-34).

FINAL CONSIDERATIONS

Considering the information analyzed and that *P. tuberosa* are among the flowers used to make bouquets, decorate vases and centerpieces, as a result of its particular beauty and fragrance, its characteristics as an ornamental plant

and potentialities, from the industrial viewpoint, either for medicinal purposes or to prepare perfumes, confirm the validity of trying to develop actions that lead to improve this important species in those places where there is interest in such crop and to create favorable conditions for its growth. Besides, to complete this goal, it is necessary to provide available information to specialists and producers, in order to ensure a proper management, increase production culture and evaluate potential demands related to this species as a cut flower and ornamental plant. Undoubtedly, these actions may contribute to grow plant species diversity and its optimal exploitation must represent benefits from the aesthetical, economic and social viewpoint. Consequently, it is important to keep on making researches about this attractive crop, which in turn is a new culture choice; therefore, it is essential to join forces and have creativity to perform strategic actions that enable to generate knowledge, preserve and use this ornamental resource sustainably.

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