State of the art of turfgrass breeding

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ABSTRACT

Population centers are constantly growing, even more in urban zones, for which the demand for plants for landscaping purposes and recreational areas also increases considerably. These zones represent unique and hostile conditions for plant growth, even that of turfgrass species. The environments where these species are grown are very influenced by man and are considered highly stressing, because they combine a large number of biotic and abiotic stress factors. In this sense, the development of new varieties adapted to abiotic stress is an aspect which has been unnoticed. That is why the development of specific breeding programs for local conditions constitutes an urgent need for the turfgrass industry. Conventional plant breeding methods have been successfully used to obtained improved varieties; however, the use of modern genetics and biotechnology tools has marked a new milestone in turfgrass breeding, which is aimed, mainly, at the attainment of abiotic stress tolerant varieties. This paper presents a bibliographic review about the state of the art in the topic of turfgrass species breeding.

Key words: breeding, plant biotechnology, turfgrass

INTRODUCTION

In the last 10 years, the turfgrass industry has grown more than any other in the agricultural sector and has become a multimillionaire industry, capable of generating thousands of jobs and/or services throughout the world.

The genesis of the Cuban turfgrass industry was based on a market test made in Varadero in 1994, which identified the interest of hoteliers in improving their gardens, which enhanced the strategic intention of the Ministry of Tourism (MINTUR) of building several golf courses (Hernández *et al*., 2007; Hernández, Suárez, Hernández and Martín, 2009). The determining factor in this process was the decision of the Cuban State to build, for the first time in Cuba, an 18-hole golf course. In this sense, in 1996 the Investment Group of the MINTUR requested the Pastures and Forages Research Station "Indio Hatuey" (EEPF-IH) to elaborate a project, in order to develop technologies to sow and establish the turf of the Varadero Golf Club (VGC).

The prestige achieved in the VGC, the beginning of the intensive use of turf in sports facilities –promoted by the National Institute of Sports, Physical Education and Recreation (INDER)–, as well as the constitution of the Program of Turfing Services of the EEPF-IH in 2000 have generated an area of research and turfing services highly accepted in the Cuban tourism and sports sector, and thus a technology-based turf industry has been consolidated (Hernández *et al*., 2007; Blanco, Martín, Suárez and Milera, 2010). However, in spite of this prominence and the priority given to turfgrass research, the Cuban turf industry faces a serious problem related to the predominance of turfgrass varieties introduced from foreign markets, which is manifested in the scarcity of suitable varieties for the local conditions, such as salinity, drought, soil infertility, etc. For such reason, obtaining new varieties adapted to local edaphoclimatic conditions, is an urgent need for the sector, which has been already included in one of the prioritized research lines of the EEPF-IH –"to introduce and evaluate under Cuban conditions, new turfgrass varieties, including the shade-resistant and salinity-tolerant ones" (Hernández, 2010)– and of the National Program of Plant Breeding and Genetic Resources- concerning the "introduction and generalization of new varieties with high genetic quality and adapted to the main edaphoclimatic conditions of the country".

Among the principal varieties used are *Cynodon dactylon* (L.) Pers (Bermuda grass) and its many hybrids; *Paspalum notatum* Flugge (Bahia grass), *Pennisetum clandestinum* Holst. Ex Chiov (Kikuyu grass), *Stenotaphrum secundatum* (Walt.) Kuntze (St. Augustine grass), *Axonopus affinis* A. Chase (Common Carpet grass), *Axonopus compressus* Sw. P. Beauv (Tropical Carpet grass), *Zoysia* spp. (Zoysia grass), etc. From them, many commercial cultivars have been obtained, especially at research stations of the United Sates, through the combination of different breeding techniques (Alteper and James, 2005; Trenholm and Unruh, 2006; Holton, Skabo, Lowe and Sinclair, 2007; Sandu and Alperter, 2008; Brosnan and Deputy, 2008).

Population centers are constantly growing, even more in urban zones; thus, the demand for plants for landscaping purposes and recreational areas also increases considerably. These zones show unique and hostile conditions for plant growth: high temperatures (island effect), extremely variable soil conditions (depth, compaction, organic matter, soil type, traffic and intense use, etc.) scarcity of water resources, etc. For such reasons, turfgrass species face a unique challenge within the plant kingdom: adaptation to urban zones (Jonhson, 2008). In this sense, in recent years most breeding programs have aimed at obtaining tolerant varieties to abiotic stress factors, such as: salinity (Munns and Tester, 2008; Koc, Bas, Koc and Kusek, 2009; Abogadallah, Serag, El-Katouny and Quick, 2010), drought (Macar *et al*., 2009), shade (Patton, 2010; Trappe, Karcher, Richardson, and Patton, 2011), low temperatures (Li, Bruneau and Qu, 2010), among others.

That is why the objective of this work was to contribute to the knowledge of the state of the art of turfgrass breeding.

Turfgrass species

Within the plant kingdom, the grass family (*Poaceae*) is the one which provides all turf-forming species. With 700 genera and more than 7 500 species, this family is present in all ecotypes of the planet: from sea level up to 3 600 m of altitude, and from arid zones to tropical or temperate areas. Nevertheless, no more than 40 species belonging to the subfamilies *Festucoideae*, *Panicoideae* and *Eragrostoideae* are capable of showing the botanical-agronomic characteristics that allow their use as turf (Wiecko, 2008). Among the warm-season (megathermal) species, the following stand out:

a) *C. dactylon* (L.) Pers (Bermuda grass, Bermuda)

The *Cynodon* genus comprises nine species, with a chromosome number varying between 18 and 54. Common Bermuda grasses (*C. dactylon* (L.) Pers var. *Dactylon*) as well as their hybrids are widely used as turf. Most common Bermuda grasses have 36 chromosomes (Tifton 10 is an exception, with 54) and produce pollen and seeds. Triploid hybrids (*C. dactylon* x *Cynodon transvaalensis*) have 2n = 3x = 27 and are sterile. However, although the common species can produce seeds, this type of propagation is not advisable, due to the appearance of mosaics in time. For such reason, vegetative propagation is practically the only reproduction way to obtain a uniform turf (Blanchard and Miller, 2002).

Cynodon species are highly used due to their vigorous creeping growth, as well as their deep rooting and fast establishment through the production of lateral shoots, which provides a competitive advantage regarding the other undesirable grasses. These species produce a very dense turf –between light and dark green–, well adapted to most of the edaphoclimatic conditions in many regions. In addition, they have excellent tolerance to wear (trampling) and to adverse salinity and drought conditions, which makes them a good choice for their use in coastal zones. Among their disadvantages are susceptibility to low temperatures and low shade tolerance. Below 10 ºC, they enter a dormancy phase (rest) and their leaves become discolored (Blanchard and Miller, 2002). Thus, one of the breeding strategies for this genus consists in obtaining low-temperature resistant genotypes.

1. *C. dactylon* hybrids

Bermuda grass hybrids are the result of interspecific crossings of *C. dactylon* x *C. transvaalensis*. They show better quality, density and color, as well as higher tolerance to stress due to trampling, heat and drought. A very important aspect which has been improved in hybrids is the tolerance to very low mowing heights (2-3 cm). Among the most known ones –described by Blanchard and Miller (2002) and Brosnan and Deputy (2008) – are:

- Tifway (also known as 419): it was obtained in 1960. It shows fine texture, dark green color and is extremely tolerant to trampling. It is used in sports fields, golf courses and gardens.

- Tifway II: it was obtained from Tifway and kept its characteristics, but with improved tolerance to low temperatures and root nematodes.
- Tifgreen (known as 328): it was obtained in 1956. It has fine texture, low growth habit and fast development. It produces a dense and highly weed-resistant turf. It is susceptible to nematodes and highly used in sports fields and golf courses.
- Tifdwarf: it is very similar to Tifgreen, but with a much more intense color and shorter leaves. It is susceptible to worm attack and it is regularly used in golf courses, due to its intense green color.
- Tifsport (also known as Tif 94): it is similar to Tifgreen and Tifway II, moderately tolerant to cold and very low mowing height.
- Tiflawn (known as Tif 57): it was obtained in 1952; it is of very fast dispersion and trampling resistant.
- Txturf-10: It was obtained in 1957 and it is recommended for athletic fields. It is very dense and has fine texture.

2. Other less known hybrids:

- Celebration: dark green color, very aggressive, fine texture and highly resistant to trampling.
- GN-1: dark green color, moderate texture and nematode resistant.

3. Super-dwarf hybrids

Super-dwarf genotypes have been obtained through somatic mutations in Bermuda grass hybrids, especially from Tifdwarf and Tifgreen; they have begun to substitute the ones usually used in golf courses (Tifgreen and Tifward). In general, the turf quality has been improved, because they have a very small intermodal length, higher stem density and mowing tolerance –with a mowing height lower than 2,0-2,5 cm (Brosnan and Deputy, 2008)–. Among the most known are:

- TifEagle (also known as TW-72). It was obtained in 1997 and it is a mutant induced by mutations with cobalt 60, from Tifway II. It has leaves with a very fine lamina, extremely high stem density and high resistance to trampling. It is commonly used in golf courses.
- Miniverde: Mutant of Tifdwarf, with much finer leaves, high lateral growth rate and improved stem density. Like most super-dwarf hybrids, it allows a very low mowing height.
- FloraDwarf: it was obtained from Tifgreen. It is similar to Tifdwarf, but with finer leaves, increase in the stem density and extremely low growth habit.

4. Common Bermuda grasses

Common Bermuda grasses are frequently used to establish turf, athletic fields and golf courses, and green areas in general. They are propagated by seed, show vertical growth habit and coarse leaf texture, and they are little tolerant to mowing below 2-3 cm. In recent years, the breeding efforts have been aimed at obtaining genotypes with better cold tolerance, moderate green color, improved density and, especially, adapted to a mowing height lower than 2,0-2,5 cm. Some of the most widely-known are: Princess-77, Rivera, Savannah, Southern Star, Transcontinental and Yukon, among others.

5. Other *Cynodon* species

- Other *Cynodon* species, more or less used as turfgrasses, are:
- *Cynodon magennisii* (Magennis Bermuda grass)
- *Cynodon bradleyi* (Bradley Bermuda grass)
- *Cynodon transvaalensis* (African Bermuda grass)
- *Cynodon incompletus*

b) *P. notatum* Flugge (Bahia grass)

The *Paspalum* genus includes about 320 species (Duncan and Carrow, 2000); nevertheless, only two are used as turfgrasses: *Paspalum vaginatum*, which is used in golf courses of temperate climates, and *P. notatum*, used in warm climates.

P. notatum is a tropical species native from South America, autochthonous from northern Argentina and Paraguay. It forms a coarse texture turf with rustic aspect. It has short rhizomes and stolons and a deep root system, which makes it resistant to drought. Initially, it has a slow growth; but once implanted, it is very invasive. It adapts to different soil types, even to the poorest ones, and it is tolerant to drought, heat, and the attack by insects and nematodes. It is especially indicated for large land extensions, roadsides, etc., because it does not need much maintenance. The mowing height depends on the use; as a general rule, between 5 and 6 cm is recommended for parks, and on large surfaces, up to 10 cm. The planting density is 1 kg/100 m^2, although it depends on seed quality.

Two main cytotypes are known: the diploid cultivar Pensacola and the tetraploid Argentine. The latter is superior regarding intensity of the green color, density and seed production. Cultivar Tifton-7 has been recently obtained, from Argentine callus culture. The breeding studies in this species are focused on the development of protocols for the generation of commercially relevant cultivars from Argentine, through tissue culture and gene transfer techniques (Alteper and James, 2005; Sandu and Alperter, 2008).

c) *P. clandestinum* Holst. Ex Chiov (Kikuyu grass)

P. clandestinum is a tropical species that originated in Africa, which has been introduced in many regions of the world. It is known mainly for the tolerance of its varieties to salinity and drought (Radhakrishnam, Waisel and Sternberg, 2006). It shows fast growth, high cover and a well-developed root system, which provides it with an adaptive advantage in the competition with other weeds. In addition, it has an added value in the control of soil erosion in desert zones, and as pasture for its high nutritional value (Radhakrishnam *et al*., 2006).

Its chromosomal number is 2n = 36. Although it is only vegetatively reproduced, in most of its distribution zones there are indications of the existence of ecotypes with apomictic reproduction (Marais, 2001). There are several ecotypes and/or varieties described (Holton *et al*., 2007), among which the cultivar Whittet stands out.

This species provides a very nice turf due to the color and texture of its leaves. It has excellent resistance to intense traffic; nevertheless, like Bermuda grasses it does not tolerate shaded zones. The maintenance is very similar to that of Bermuda grass; but because of its high growth aggressiveness, it competes better with weeds and requires a more intense fertilization. It is important to mow it frequently and to keep it low, in order to prevent the formation of brush. Due to its high growth rate, it tends to "become padded". This effect could be attractive at first; but if it worsens, it damages the turf and could cause its death. The recommended mowing height is 3-4 cm. The sowing density is 400 to 700 g/100 m^2 at the beginning of spring.

d) *S. secundatum* (Walt.) Kuntze (St. Augustine grass)

S. secundatum is widely distributed in tropical and subtropical regions, in which it is mainly used as pasture. Its propagation is principally vegetative, although it also has sexual reproduction associated to high sterility levels (Busey, 1986). The studies in this species have focused on the response of cultivars to biotic and abiotic factors (Li, Bruneau and Qu, 2006). Among the most known cultivars are: Floratan (wide leaf laminae), Amerishade (shade tolerant), Classic (cold tolerant), Delmar (dense growth), Mercedes (cold tolerance), Palmetto (small and thin leaves), Raleigh (cold tolerance), Sapphire (for particular lawns) and Seville (shade tolerant).

e) *A. affinis* A. Chase (Common carpet grass)

In general, the species of this genus are extended throughout Australia, Central America, Malaysia, South America, North America, South Korea and Western Africa. Turfgrasses have a very fast lateral growth, through stolons, which provides an attractive and wear-resistant turf (Greene *et al*., 2008).

A. affinis is a perennial creeping species which is propagated by stolons and forms a tapestry of low vegetation, with stems up to 50 cm high, short stolons and narrow, smooth and blunt leaves. It grows on sandy, well-drained, humid-zone soils. It persists on poor soils, responds well to fertilization and is a popular pasture in the zones where it is adapted. As a turfgrass it is cultivated, mainly, for soil conservation. The seeds are easy to harvest and a good mass density can be established, with minimum land preparation.

f) *A. compressus* Sw. P. Beauv (Tropical carpet grass)

A. compressus forms a very dense perennial turf, of coarse texture and moderate green color. Likewise, it guarantees a very attractive and ornamental soft cover which, due to the tight closure of its leaves, hinders the establishment of weeds. It adapts to moderate shade, stands trampling and has good recovery capacity in case of deterioration. It requires the application of iron in the establishment and within the maintenance scheme.

g) Zoysia spp. (Zoysia grass)

The species of the *Zoysia* genus are vegetatively reproduced by rhizomes and stolons, and produce a very dense and trampling-resistant turf. They are adapted to a very wide range of environmental conditions: good tolerance to cold, shade, salinity, etc. Its establishment is difficult, due to its slow growth (Quian, Engelke and Foster, 2000; Wen and Chen, 2001; Trenholm and Unruh, 2006).

At least five species which have been used as turf are known: *Zoysia japonica*, *Zoysia matrella*

(L.) Merr., *Zoysia tenuifolia* Trin., *Zoysia sinica* Hamce and *Zoysia macrostaycha* (Quian *et al*., 2000). The most widely used are:

- *Z. japonica*: is the most tolerant to cold and can be established from seed. The most widely known cultivars include: Meyer (improvement in texture, color, vigor and tolerance to different environmental conditions), Belair and El Toro (coarser texture and faster propagation).
- *Z. matrella*: it shows thinner, pointed and resistant leaves. It does not tolerate very low temperatures.
- *Z. teniufolia*: it has the finest texture and is less resistant to low temperatures. It forms a dense and soft turf. Its propagation is slow, for which it has been used mainly as soil cover.
- *Z. japonica* x *Z. tenuifolia*: it has produced the cultivar Emerald, which combines the fine texture of *Z. tenuifolia* with the cold tolerance and fast dispersion of *Z. japonica*.

Besides, there are other cultivars more or less used as turfgrasses: Crowne, Empire and Ultimate (Trenholm and Unruh, 2006).

Nevertheless, the most important species within this genus is, undoubtedly, *Z. japonica*. It is highly used in the East for its excellent characteristics: fine texture, very slow growth rate, resistance to diseases, tolerance to intense traffic, etc. At some moment it was considered the magic turfgrass, because of having many advantages and few disadvantages, but the fact that it does not produce adequate seed for planting caused it to be relegated. This is in addition to its extremely low growth, which is a favorable characteristic; but once established, it is a great disadvantage to be vegetatively reproduced. Through special techniques it can be harvested and treated to obtain viable seeds. In the United States, intense work is being done to obtain varieties with higher seed production than the current ecotypes. Until now it is not an important species in Cuba, but it could be in the future, when the new varieties are available in the market.

h) *Buchloe dactyloides* (Nutt.) J.T. Columbus (Buffalo grass)

Buchloe dactyloides produces a drought- and cold-tolerant turf. However, it does not adapted to shaded places and does not stand trampling.

i) *Eremochloa ophiuroides* (Munro) Hack (Centipede grass)

Eremochloa ophiuroides grows in sunny sites and has low tolerance to shade (it does not survive under trees) and to traffic. It is propagated by seed and vegetatively.

j) *Distichlis spicata* (L.) Greene (Salt grass, alkali grass, spike grass)

Distichlis spicata can be used for almost all types of turf, especially in the restoration of marshlands. It tolerates saline, alkaline and poorly-drained soils.

Objective of the breeding programs

The first step in any breeding program is the selection of the breeding object. The main selection criteria in turfgrasses depend, mainly, on their use: gardening, sports fields (athletics, baseball, golf, etc.) or aesthetic uses. In this sense, breeding programs are aimed at the following variables:

- Turf density
- Leaf fineness
- Resistance to trampling
- Resistance to diseases
- Summer aspect
- Winter aspect
- Establishment rate
- Leaf color
- Global aesthetic aspect
- Management
- Seed attainment

However, in general, the most important attributes that should be taken into account are: the appearance and resistance or tolerance to biotic (diseases) and abiotic stress factors (salinity, drought, temperature, shade) (Florkowski and He, 2007). This has caused the selection criteria to address the aspects that approach a more ecological management of turfs:

- Drought tolerance (decrease of irrigation)
- Heat tolerance
- Salinity tolerance (irrigation with recycled water)
- Shade tolerance (good growth under trees)

Abiotic stress

Plants are affected mainly by two stress types: biotic and abiotic. The former is caused by living organisms and comprises the susceptibility to pests and diseases; while the latter is environmental and includes: low/high temperatures, salinity, drought, shade, etc. The abiotic factors are considered one of the main limiting factors in agricultural and/or industrial crops, because they have incidence on plant growth and productivity and trigger a series of morphological, physiological, biochemical and molecular disorders. Drought, extreme temperatures, saline soils and, to a lower extent, shade are the most detrimental.

Water stress (caused by salinity and/or drought) is one of the most studied abiotic factors, because of two main reasons: firstly, because it severely damages the plants; and, secondly, due to the large quantity of soil extension that is affected because of these limiting factors –according to data of the FAO (http://www.fao.org/corp/statistics/es/), more than 20% of the world agricultural surface is affected–. Saline stress occurs due to an excessive concentration of salts in the soil, which interferes with plant metabolism and causes an imbalance in water balance. Salinity and drought damage plants in a very similar way, and the common factor to both stresses is the reduction of the water potential (Macar *et al*., 2009). Salinity can be intrinsically given by the soil characteristics (that it is saline) or be caused by external factors such as irrigation, agricultural activity, excessive use of fertilizers, among others (Zhu, 2001). Drought acts by decreasing the germination percentage and plant growth.

Regarding water, turf is often seen as synonym of wasting and, thus, it is not considered sustainable; if it is taken into account that water saving and good use is a priority objective worldwide. This hypothesis is vital for the functional management of a turf with relation to the integral management of the water resources in urban zones, with the subsequent reduction of inputs (for example, irrigation) (Florkowski and He, 2007). Hence the tolerance/resistance to drought by turfgrass has become an essential issue among the technicians and professionals of green areas, as well as among those who are public responsible regarding the environment.

Shade is a factor that limits turf growth. It acts on the reduction of the root mass, root number, carbohydrate reserves, rhizomes and stolons, for which it affects the general turf quality (Trape *et al*., 2011). In most cases, trees (essential components of the landscaping design in tourism and sports facilities) are responsible for this stress, by competing for the soil resource.

It is evident that the above-mentioned abiotic stress factors significantly affect turf quality and are very restrictive in the selection of a certain species for a turfing project. Thus, avoiding this type of stress has become an urgent need for researchers and/or entrepreneurs. There is an absolute consensus on the fact that the most effective method to reduce the effect of these limiting factors is to obtain resistant and/or tolerant varieties, through breeding programs, adapted to the particular conditions of each locality (Casler *et al*., 2003; Koc *et al*., 2009; Li *et al*., 2010; Patton, 2010; Trape *et al*., 2011).

Turfgrass breeding

Florkowski and He (2007) described the three selective forces that contribute to create and maintain the current germplasm pool in turfgrass varieties; they are:

1. Natural selection: process through which the species adapt to the changing edaphoclimatic conditions of a particular environment.
2. "Unconscious" selection or domestication: process through which man phenotypically selects the most valuable individuals and ignores or destroys those which are not of value. During this process no selection criterion is defined or predetermined.
3. Methodical selection: processes and/or methods through which man selects individuals following a predetermined selection criterion. It comprises all the breeding methods (conventional and biotechnological).

The improvement of turf quality in the last 50 years has been an important part of research in plant biology and in agronomic sciences. The first seeds commercialized for turf during the first half of the 20th century came from lots collected in natural populations. After World War II, the diffusion of Mendel's laws, the development of discoveries on genetics (chromosomes, DNA) and the theory of quantitative genetics allowed to establish efficacious selection methods. Since then, progress has been constant and fast.

Since centuries ago, conventional breeding has been carried out in turfgrass species, and only in the United States more than 246 cultivars have been obtained since 1946. Yet, the inaccessibility to the genetic material –due to the sexual reproduction barrier in these species–, together with the large amount of time that must be invested to develop these programs, has restricted the use of such methods. Plant biotechnology has allowed the discovery of new and more efficient breeding methods, through the introduction of desirable traits in a short time span. The combination of tissue culture techniques, along with the development of genetic transformation technologies, has facilitated the introduction of specific genes in a great quantity of turf-producing species (Liang and Skinner, 2006).

Among conventional methods, the following stand out:

a) Natural variation and domestication

The first efforts in turfgrass breeding were focused on using the genetic variation created by natural selection. The first stoloniferous cultivars (for example, Bermuda grass) were identified and propagated from their natural habitats, in the early 1907 (Roux, 1969).

b) Recurrent selection

Many of the new cultivars of turfgrass species have been obtained by phenotypic and/or genotypic recurrent selection, which consists in the evaluation of a large number of individual plants or replicated clones, followed by the selection and crossing of the best individuals. This method requires another stage: the plants or clones to be evaluated are arranged in a crossing block, and the seeds are obtained from each individual. To complete the cycle, the crossings occur between the parents of the best families (Caster *et al*., 2003).

c) Interspecific hybridization

Many turfgrass cultivars are the direct or indirect result of interspecific hybridization (Brilman, 2001; Belanger et al., 2003). Interspecific hybrids can emerge naturally or through a controlled crossing program. One of the most illustrative examples is the large quantity of hybrid triploids obtained from the crossing between the tetraploid *C. dactylon* and the diploid *C. transvaalensis* (Busey, 1989).

The new technologies have started a new era in plant breeding. Among the most widely used are:

a) Marker-assisted selection (MAS)

Marker-assisted selection for quantitative traits consists in: 1) the identification of hypothetical *Loci* for the trait (Quantitative Trait Loci, QTL), through the correlation of the phenotypic data with markers, within a linkage map; 2) the determination of which hypothetical QTL explains the higher quantity of phenotypic variance and carries the desired effect on the phenotypic trait; and 3) plant selection based on the molecular marker, together with the hypothetical QTL (Dudley, 1993).

b) Technology of molecular markers

The use of the technology of molecular markers has allowed important scientific advances for the knowledge of the genetics and genomics of turfgrass species. In this sense, genetic similarities and/or differences among cultivars have been identified, and the genetic variation patterns related to the history of the species crossing have been illustrated (Huff, 2001).

c) Transgenesis

Transgenesis allows the transference of functional genes between the species or between genotypes within them. In addition, it allows breeders to directly manage the traits of interest, such as the tolerance to edaphic and stress factors. Duncan and Carrow (2001) offered a review of the application of these technologies and mentioned a group of genes of interest for obtaining turfgrass varieties transformed to improve tolerance to stress situations.

d) Mutation breeding

Mutation induction has been applied during the last 70 years to obtain mutant cultivars of sexual- and asexual-propagation crops. The most common mutagens used are the chemical and physical ones. Among the physical agents, gamma radiations are the most used for mutation induction (Jain, 2005). More than 1 800 cultivars of the principal cultivated species have been obtained this way (Suprasanna, Sidha and Bapat, 2008). Radiation-induced mutations have been widely used in turfgrass species to generate cultivars with modifications in morphological traits (Lu *et al*., 2009).

e) Tissue culture techniques

Tissue culture induces a variation in the regenerated plants known as somaclonal variation (Larkin and Scowcroft, 1981). There are studies which prove that it is stable and as useful for plant breeding as the mutation-induced one (Jain, 2001). In this sense, the callus induction techniques, plant regeneration and later *in vitro* selection in selective environments and/or media are widely used for breeding turfgrass species (Lu *et al*., 2007).

f) *In vitro* mutagenesis

In vitro mutagenesis does not constitute a technique in itself; it is rather a combination of mutation breeding with the modern biotechnological *in vitro* culture techniques. The induction of mutations can be reinforced through the use of *in vitro* techniques. In this sense, the combination of the *in vitro* culture and mutagenesis becomes a powerful way to generate genetic variability. It is cheaper, simpler and more efficient than other technologies (Suprasanna *et al*., 2008; Lu *et al*., 2009). The main advantage of the use of *in vitro* mutagenesis is that a large quantity of cells can be treated and afterwards regenerated through different culture cycles and in a relatively short time period.

These techniques have been very successfully applied in Cuba, mainly in the national breeding programs of rice (Fernández, Peteira, González and Llanes, 2003) and, worldwide, for turfgrass breeding (Pongtongkam *et al*., 2005; Lu *et al*., 2007; Suprasanna *et al*., 2008; Lu *et al*., 2009; Li *et al*., 2010).

FINAL CONSIDERATIONS

Turfgrasses are exposed to biotic and abiotic stress, the latter needs to be contextualized according to the specific local conditions, such as: salinity, drought and shade.

The turfgrass varieties introduced in Cuba were bred in countries where the edaphoclimatic conditions differ from ours, which implies the need to implement a breeding program to obtain Cuban varieties, which are capable of tolerating and/or standing the abiotic stress.

Mutation induction is considered to be the adequate method to obtain new Cuban turfgrass varieties, taking into consideration that satisfactory results have been obtained in the breeding of other grasses.

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