

Characterisation of vegetative bermudagrasses

(Cynodon spp.) for turf use in Australia

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<u>Abstract</u>

In this study, up to 21 morphological traits including those describing growth habit, stolons, shoots and inflorescences were measured on all of the available recreational Cynodon spp. turf varieties in Australia including 19 hybrid interspecific bermudagrass (Cynodon dactylon x C. transvaalensis) varieties and 16 bermudagrass (C. dactylon) varieties at the former Queensland Department of Agriculture, Fisheries and Forestry (DAFF) Redlands Research Facility, Queensland, Australia (27°32'S lat, 153°15'E long, 25 masl) between 2002 and 2004. During this period 5 spaced plant experiments and a single sward experiment, were established to measure a range of morphologicalagronomic characteristics.

Up to 14,248 morphological-agronomic data points were collected from the 6 experiments monitoring stolon and shoot, inflorescence and growth habit characteristics. These data were analysed by first conducting a spatial analysis on each experiment adjusting for any environmental trends and to obtain estimates of residual variances and first order auto-regression coefficients for row and column to be input into a one-stage spatial analysis for each trait. Because varieties were treated as 'random' these analyses produced a set of variety averages or Best Linear Unbiased Predictors (BLUPs) for each trait. The BLUPs were used to construct a two way table of varieties x traits and used for a pattern analysis i.e. clustering and ordination.

Four 'variety groups' were identified, following a pattern analysis of the comprehensive data set. Each of the groups included members of both tetraploid C. dactylon (2n = 36) and triploid Cynodon hybrids (2n = 27) highlighting the large morphological variation that exists within the Cynodon taxa used in this study.

The morphological-agronomic traits studied were also grouped following pattern analysis to provide 6 'trait groups'. Four of the 6 trait groups contained individual traits, while 2 groups contained multiple characteristics. A set of informative traits were compiled consisting of a single characteristic from each of the 6 trait groups that would provide a distinct, uniform and stable morphological measure. Trait grouping could also provide beneficial information for the selection of varieties of common knowledge (VCK) needed for comparative testing as required for trials to obtain, for example, Plant Breeders Rights in Australia or Plant Variety Protection in the USA. Comparator varieties could be chosen from within a group containing parent/source material or a variety earlier identified following observations of morphological and developmental growth. Using this method to identify VCK would remove subjective and often bias judgement.

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Contributions by others to the thesis

Dr Vivi Arief of The University of Queensland assisted with undertaking pattern analysis of morphological-agronomic data and the construction of dendrograms and biplots within the present study.

Statement of parts of the thesis submitted to qualify for the award of another degree

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List of Abbreviations

ACT, Australian Capital Territory Adl., Adelaide AGCSA, Australian Golf Course Superintendents Association Aus./AUS, Australia AUSPBR, Australian Plant Breeder's Rights ARC, Australian Research Council chrom., chromosome DAFF, Department of Agriculture, Fisheries and Forestry DEEDI, Department of Employment, Economic Development and Innovation DNT, day/night temperatures DPI&F, Department of Primary Industries and Fisheries DPP, days post planting DUS, distinct, uniform and stable Exp., Experiment GDD, growing degree days GNTC, Greg Norman Turf Company GPS, Global Positioning System HAL, Horticulture Australia Limited IAC, Industry advisory Committee **IP**, Intellectual Property ISSR, Inter-simple-sequence-repeat LSD, Least Significant Difference L:W, length, width ratio Met., meteorological M-A, morphological-agronomic NZPVR, New Zealand Plant Variety Rights NSW, New South Wales PBR, Plant Breeder's Rights; A, represented as a PBR symbol PCR, polymerase chain reaction PPFD, Photosynthetic Photon Flux Density QP, Qualified Person OLD/Old. Oueensland DAFF, Queensland Department of Agriculture, Fisheries and Forestry DEEDI, Queensland Department of Employment, Economic Development and Innovation; DPI&F, Queensland Department of Primary Industries and Fisheries SA, South Australia US, United States USA, United States of America USDA, United States Department of Agriculture USGA, United States Golf Association RHS, Royal Horticultural Society **RRF**, Redlands Research Facility UWA, University of Western Australia USPP, United States Plant Patent UPOV, International Union for the Protection of New Varieties and Plants VarGr, VarGr VIC, Victoria WA, Western Australia WPP, weeks post planting

Chapter 1: Introduction

1.1 Cynodon Taxa Used as Functional Turfgrass

The two main *Cynodon* taxa used for turfgrass applications are *Cynodon dactylon* L. Pers. (bermudagrass) and the interspecific hybrid *C. dactylon* x *C. transvaalensis* Burtt-Davy (hybrid bermudagrass). The morphological and developmental diversity present between taxa and within each taxon is vast. An example of this is within the hybrid bermudagrass group where commercially available varieties (= cultivars) can be variously grouped as dwarf, ultradwarf or medium-to coarse textured hybrids.

"Ultradwarf" is a term coined by Dr Philip Busey of the University of Florida's turf research program, FloridaTurf, in the USA (Busey and Dudek, n.d.). The term refers to *Cynodon* varieties that are even shorter than the older standard dwarf hybrid bermudagrass varieties 'Tifgreen' (often referred to by its evaluation code number, Tifton 328) and 'Tifdwarf'. Ultradwarf varieties available in Australia include 'Champion Dwarf', FloraDwarfTM, 'MS-Supreme', MiniVerdeTM (P18), NovotekTM (TL2) and 'TifEagle'; however, only the last two varieties have been commercialised here. Novotek, TifEagle, and the older industry standards Tifgreen and Tifdwarf are predominantly used on bowling and putting greens because of their ability to be mown very short e.g. 2-5 mm.

The medium- to coarse-textured hybrid bermudagrasses are used in a similar matter to bermudagrass varieties because of their coarser texture, greater height and growth habit. Commercially available medium- to coarse-textured varieties (= cultivars) in Australia include 'AGRD', TifSportTM (Tift 94) and 'Santa Ana'; while 'Patriot', 'Tifway' and 'Premier' are only available experimentally.

1.2 Cynodon and Its Importance

The *Cynodons* are the major warm-season turfgrass taxa used for functional, recreational, and aesthetic purposes in Australia and internationally. Beard and Green (1994) list their functional benefits as: excellent soil erosion control and dust stabilization due to the dense sward that protects the soil; improved groundwater recharge and quality protection, plus flood control; enhanced entrapment and biodegradation of synthetic organic compounds; soil improvement (including CO₂ conversion); accelerated restoration of disturbed soils;

temperature moderation through urban heat dissipation; reduced noise, glare, and visual pollution problems; decreased noxious pests and allergy-related pollens; safety in vehicle operation on roadsides and engine longevity on airfields; reduced fire hazard via open, green turfed firebreaks; and improved security of sensitive installations provided by high visibility zones. Cynodon dactylon also has economic value for grazing and hay production (Harlan et al., 1970a). According to Beard and Green (1994), the recreational benefits of the Cynodons include low-cost surfaces for outdoor sport and leisure activities which enhance the physical health of participants, and provide a unique low-cost cushion against personal impact injuries. The aesthetic benefits include enhanced beauty and attractiveness; a complimentary relationship to the total landscape ecosystem of flowers, shrubs and trees; improved mental health through a positive therapeutic impact, social harmony and stability; improved work productivity; and an overall better quality-of-life, especially in densely populated urban areas. For whatever reason Cynodon is chosen or is found growing in a particular environment, these taxa will dominate areas receiving a medium to high level of management practices. However, if poorly managed, they will grow poorly and become invaded by other species (weeds) as frequently seen in general lawns, parks and gardens.

1.3 Morphological and Developmental Characterisation

To date, little information has been published on the origin, usage and morphologicalagronomic characteristics of each of the available *Cynodon* varieties in Australia, the majority of which have been introduced from the United States. However, there is an increasing number of varieties selected within Australia, which should be better adapted to its harsh growing environment.

Since 1987, the breeders of turfgrass varieties developed in or introduced into Australia have been able to seek protection of the intellectual property embodied in their variety through Plant Breeder's Rights (PBR). These rights were originally embodied under the *Plant Variety Rights Act 1987* which was later modified to conform with the 1991 Act of the UPOV Convention and become the *Plant Breeder's Rights Act 1994*. The grant of PBR for a new variety is based on a 'breeder testing' system which includes establishing, conducting and reporting on a comparative morphological-agronomic growing trial to describe the new variety and to demonstrate that is <u>d</u>istinct, <u>uniform and stable</u> (DUS) against the parent material and/or the closest varieties of common knowledge.

Ideally, such a process involves a number of morphological-agronomic traits of each variety being measured in a replicated trial and statistically analysed. However, in the absence of detailed guidelines having been prescribed by the examination authorities within the UPOV, the question remains: which traits are the most appropriate to measure? To this end, detailed morphological-agronomic data from previous growing trials is available for critical analysis to determine the traits that are the most effective in describing and distinguishing between turf varieties of *Cynodon* spp. and other warm-season grasses. From this, a standard set of descriptive guidelines based on 'informative traits' could then be developed, as well as providing a sharper focus to future breeding and management studies on these species.

1.4 Aims of the Study

This research study seeks (i) to characterise the available morphological-agronomic variation and relationships among the majority of current functional turf varieties of *C. dactylon* and hybrid bermudagrass within Australia; and (ii) to identify the most appropriate morphological-agronomic traits that can be used to describe and differentiate among varieties of *Cynodon* spp. and potentially other warm-season turfgrass varieties.

Chapter 2: Literature Review

2.1 Introduction

Bermudagrass (commonly referred to as green couch in Australia) is suitable for turf plantings under low to high levels of maintenance in home lawns, on roadsides, and in cemeteries, parks, sporting fields, and other venues (McCarty and Miller, 2002). The two main *Cynodon* taxa used for turfgrass applications are:

- *Cynodon dactylon* L. Pers., which is mainly tetraploid (2n=36 chromosomes) but with some hexaploid genotypes (2n=54 chromosomes); and
- *C. dactylon* x *C. transvaalensis* Burtt-Davy, an interspecific hybrid which is usually triploid (2n=27 chromosomes) but also tetraploid (2n=36 chromosomes) in one case ('Patriot') (Taliaferro 2003; Taliaferro et al., 2004b, 2006).

Within the latter taxon, the available varieties vary considerably and are often grouped into two broad categories based on leaf and stem texture: (i) dwarfs and ultradwarfs, which are predominantly used on bowling and putting greens; and (ii) medium- to coarse-textured hybrids, which are used for similar purposes to *C. dactylon* varieties.

Bermudagrass is adapted to a range of soil types, though best suited to a well-drained, acid or alkaline, fertile clay, clay loam or sandy soils (e.g. McCarty and Miller, 2002; Taliaferro 2003; Taliaferro et al., 2004b). If managed correctly, including providing adequate moisture and available nutrients to the plant, acceptable to optimum turfgrass quality will be achieved. As regards turfgrass quality, this is largely dependent on the variety. There is vast morphological and agronomic variation within the two Cynodon taxa suitable for turfgrass use. Around 50 years ago, the range of selected varieties was minimal. However, the last 10-20 years has seen the choice expand significantly (McMaugh, 2008). More varieties are currently being developed for release by breeders (e.g. private, university), both in Australia and overseas, in an attempt to supersede earlier varieties and target niche markets (e.g. global warming, increased urbanisation, lifestyle choices).

The following review looks at the history, taxonomy, including classification, species distribution and morphological aspects of the genus *Cynodon* and the two main taxa within the genus in particular. It also covers the numerous varieties which have developed and been introduced successfully or unsuccessfully into Australia during recent years. Since 1994, the

majority of the varieties have some form of proprietary protection (e.g. Plant Breeder's Rights, trademarks) which is also reported up on. Positive developments such as property rights and trademarks, along with numerous advancements in the turf supply chain has helped unite a previously fragmented industry and led to the recent emergence of Turf Australia, the national peak body for turf producers. For the consumer, greater choice has seen tremendous change in urban development (e.g. parks and gardens, multi-use sports facilities) and made alternative lifestyle choices possible with improvements in couch grasses that are better suited to our ever changing environment.

2.2 Origin of Bermudagrass

While many early writers believed that bermudagrass "evolved" in India, most of the improved strains have been developed from African genotypes (King, 1966; Mitich, 1989); According to the diary of Thomas Spalding, a prominent antebellum agriculturist and owner of Sapeloe Island, Georgia, one early path of bermudagrass entry into the United States was through Savannah, Georgia in 1751, by Governor Henry Ellis transporting cargoes of slaves in three voyages from Africa to Jamaica between 1750 and 1755 (Cashin, 2003). Kneebone (1966) indicated that bermudagrass may have been introduced to the new world, probably the West Indies, soon after its discovery by Columbus, and possibly on ships commanded by Columbus; he also noted that it may have been introduced to Savannah, Georgia, area prior to 1751 by Robert Miller, a botanist employed by the Lords Proprietor to collect plants from the Caribbean Islands and Central America from 1733 to 1738.

Such factual historic information and note taking by early pioneers is important when considering whether or not *Cynodon dactylon* was native to Australia pre-1770. Beehag (2006) stated that couch grass under the name *Panicum dactylon* was one of several grasses recorded at Port Jackson, Sydney by the Scottish botanist Robert Brown during his 1802-1805 voyage around Australia whilst aboard HMS "Investigator", under the command of Matthew Flinders. During this period and upon his visit to Australia, Brown made collections of *Cynodon dactylon* (Brown, 1810) and remarked in his notes that the species had possibly been introduced (Brown, 1814). There was great debate among early botanists upon whether or not bermudagrass in Australia was a result of an early introduction, or whether the species was native to the vast country. Beehag (1992) had initially reported that early records indicated common couch (*C. dactylon* var. *dactylon*) to be widely distributed in Australia at

the time of European settlement and was found in remote areas not apparently distributed by man (Taliaferro, 2003). However following colonisation, couch grass became widespread reported as the early 1850s along the Murray River, South Australia; and following mycological studies conducted in 1954 at the University of Queensland, Langdon (1954) concluded that *Cynodon dactylon* to be an introduced species. This conclusion was derived from historical fungal parasitic studies of smut and rust and the occurrence on the host plant *Cynodon dactylon*. While he noted that in light of these facts there was still some doubt if the species was indigenous to Australia, and it may have been possible that it became established in Australia at a time when a land connection with other parts of the Old World (Gondwana) was in existence.

Cynodon dactylon was ranked by Holm et al. (1977) high among the world's worst weeds and arguably the worst grass weed. In this context, it has been classified as a severe, principal or common weed (based on decreasing levels of damage) in most warm-climate countries in Africa, America, Asia, Australasia and southern Europe (Holm et al., 1979; Horowitz, 1996) because its rhizomes can penetrate the soil to a depth of one meter or more (Hanna, 1992) making it difficult to eradicate.

The species is widely distributed throughout all of the world's continents where it has been recorded from 53°N in Europe and Asia through to 45°S in South America, and at more than 3000 m elevation in the Himalayas (Harlan and de Wet, 1969; Harlan et al., 1970a; Taliaferro, 2003; Taliaferro et al., 2004b; Kenworthy et al., 2007); it is also found on the islands in the Pacific, Atlantic and Indian Oceans (Hanna, 1997). In Australia, C. dactylon is found in all six states and two territories, including Lord Howe Island (Sharp and Simon, 2002). The species is predominantly found growing between subtropical and warm temperate regions. In the transition zone or even cooler environments (at $>0^{\circ}$ C), the species remains in a nondormant state and is capable of maintaining a form of active growth throughout the year. Under such conditions, growth is significantly slower during the cool season and low temperatures may temporarily cause discolouration of foliage by inducing anthocynanin pigmentation or tissue damage from light frosts (Taliaferro, 2003). Studies conducted by Roche et al. (2005) identified minimum threshold temperatures for dwarf hybrid couch grass growth were approximately 9° to 10°C (air temperature) and 15° to 16°C at 10 cm soil depth at studies conducted at DAFF Redlands Research Facility, Queensland, Australia (27°32'S lat, 153°15'E long, 25 masl).

2.3 Cynodon Morphology

Cynodon has a C4 photosynthetic pathway (Bogdan, 1977; Watson and Dallwitz, 1992), which responds to high light intensities and suffers from shading (Horowitz, 1996). *Cynodon* plants are perennial, naturally low growing, sod-forming species. Plants have flat, linear leaf blades, sometimes rolled when budding, with a membranous ligule often with hairs on the upper edge (Taliaferro, 2003). With the exception of three species, *C. barberi*, *C. arcuatus* and *C. transvaalensis*, there is a conspicuous range in leaf size making it difficult to consistently distinguish the various taxa within the genus (Rawal and Harlan, 1970). This also relates to leaf size on different plant parts and under different management conditions: e.g. studies by Roche and Loch (2005) showed that the leaves of hybrid bermudagrass varieties differ considerably in shape between stolons of unmown spaced plants and unmown tillers taken from a sward.

Culm nodes of *Cynodon* spp. could be interpreted as simple with the upper leaf subtending their terminal bud even though the terminal bud becomes far removed by subsequent growth of the culm (Rawal and Harlan, 1970) (Figure 2.1a). The stolon nodes of *Cynodon* taxa are compound and produce 3 leaves reported in general by Bogdan (1952) and Roche and Loch (2005). The multiple leaves at each compound node are typically closer to the apex of the stolon. The lower delimiting lateral buds are on the opposite side of the stolon (Figure 2.1b).



Figure 2.1 Diagram of *Cynodon* (a) culm leaves and (b) stolon leaves (Source: Rawal and Harlan, 1970).

Each stolon node typically produces 2 to 4(-7) branches (Roche and Loch, 2008a), one or two of which may become side stolons, thus creating a complicated network of branching stolons which develops into a sward. Such developmental morphology is an important consideration in the management of perennial grasses used for turf (Moore and Moser, 1995).

There is a high level of variability in the branching pattern of *Cynodon* species. Both *C. dactylon* and *C. dactylon* x *C. transvaalensis* are identified as having an opposite equal branching pattern as seen in Figure 2.2c (Rawal and Harlan, 1970). Other branching patterns of the genus include - alternate, having one bud suppressed (Figure 2.2a), or intermediate, which regularly has one bud delayed (Figure 2.2b). It is possible for the latter two branching patterns to be found also on *C. dactylon* and *C. dactylon* x *C. transvaalensis* plants, hence the variability among varieties.



Figure 2.2 *Cynodon* spp. branching patterns (a) alternate, (b) intermediate and (c) opposite equal (Source: Rawal and Harlan, 1970).

It has been well documented in several studies (e.g., Gray and White, 1999; Guertal and White, 1998; Guertal et al., 2001; Knoop, 2000; White et al., 2004; McCarty and Canegallo, 2005) that the ultradwarf varieties produce higher shoot densities which result in increased thatch production.

Temperature, photoperiod and light levels can all influence the growth and development of bermudagrass. Youngner (1959) found that new shoot production and stolon elongation occurred in 'U-3' bermudagrass when day temperatures were greater than or equal to 15.5°C and night temperatures were greater than or equal to 4.4°C – a mean daily temperature threshold of around 10.0°C. Under higher temperature conditions in a controlled environment study, Stanford et al. (2005) reported that internode and leaf length on the hybrid variety 'Tifdwarf' were greatest in their 27/19°C treatment and least under the 35/27°C day/night temperature regime, regardless of incident light levels. They predicted this change in growth

form would result in faster coverage from newly planted sprigs and faster recovery from disruptive cultural activities when daytime temperature is 27° C or less as compared with periods when the daytime temperature is 30° C or above and the PPFD is $1000 \,\mu\text{mol}\,\text{m}^{-2}\,\text{s}^{-1}$ as is typical in the Australian subtropical and tropical climatic zones.

At low light intensities as measured by photosynthetically active radiation (PAR) relative to full sunlight levels (<70% for most *C. dactylon* genotypes), the shoot growth progressively becomes more open and more etiolated ('prayer-like') with narrow elongated leaves on thin upright stems with elongated internodes; rhizome and stolon growth also become weaker (McCarty and Miller, 2002). As a result, a bermudagrass sward becomes sparse, weak and open when grown under shaded conditions. Dong and de Kroon (1994), for example, reported longer internodes and less stolon and rhizome branching in bermudagrass grown at 20% of full sunlight than when grown in full sun. In a study of the effects of photosynthetic photon flux density (PPFD), temperature and nitrogen fertilisation on growth and development of Tifdwarf, Stanford et al. (2005) showed that the internode and leaf length were greatest at 27/19°C day/night temperature (DNT) and lowest at 35/27°C DNT.

Marousky et al. (1992) applied a short 9-h daylength and a long 18-h daylength (achieved by interruption of the night period by 4-h of incandescent light) to 10 bermudagrass and hybrid bermudagrass varieties grown under low and high fertility regimes in a glasshouse pot experiment. They found that the long daylength increased leaf length in 9 of the 10 genotypes, but not stolon number and plant dry weight, while all three attributes were increased by high fertility. However, the 10 varieties did not all respond uniformly to daylength and fertility, with differences among them in the magnitude of treatment responses by each attribute measured.

The reproductive development of bermudagrass is influenced by daylength (photoperiod). Mes (1958) and Nada (1980) have reported a long-day response and a quantitative long-day response, respectively. Anecdotally, however, daylength control of flowering in bermudagrass and hybrid bermudagrass is likely to be more variable and/or complicated, given the numerous variations in flowering time observed in the various varieties and experiments reported in this thesis (D.S. Loch and M.B. Roche, unpublished observations, 2002-04).

There is a large range in leaf size among members of both *Cynodon* taxa, making it difficult to consistently distinguish between them (Rawal and Harlan, 1970). Variation within *C. dactylon* x C. *transvaalensis* is such that varieties are commonly grouped into two categories: the finer, denser, lower-growing grasses (e.g. ultradwarf and other greens quality grasses) and the medium-textured grasses (e.g. TifSportTM, 'Santa Ana'). Studies by Roche and Loch (2005) showed that greens quality ultradwarf leaves differed considerably in shape between stolons of unmown spaced plants compared with those from a mown sward. Marousky et al. (1992) also reported that leaf length is a morphological characteristic that varies with seasonal daylength changes and fertility level.

The genus *Cynodon* is essentially caespitose (growing in tufts) (Rawal and Harlan, 1970) with *C. transvaalensis* and *C. dactylon* both growing by stolons and rhizomes (Bogdan, 1977). Rawal and Harlan (1970) describe *Cynodon* as having two kinds of rhizomes, (i) a true rhizome that has a tendency to be relatively slender, straight, with long internodes in which the tip is always below the soil surface (Figure 2.3a); and (ii) a rhizome that produces large diameter, fleshy, usually crooked, short internodes and that emerges to the soil surface where it converts into a stolon (Figure 2.3b) (Rawal and Harlan, 1970). The true rhizome grows deeper and faster than compared to the porpoising growth habit seen in Figure 2.3b where the rhizome breaks the soil surface.



Figure 2.3 (a) a true type rhizome in which the tip always stays below the soil surface; (b) rhizome that emerges and is converted to a stolon (Source: Rawal and Harlan, 1970).

Little thought is often given to the inflorescence characteristics of turfgrass, as most people simply perceive that turf areas are routinely mown. To an extent, this is true. Facilities such as golf courses, lawn bowling greens and sports stadia mow their turf frequently, and at times the turf manager may decide to double-cut the turf surface. However, in recreational areas such as parks and gardens, roadsides and home lawns that are mown less frequently, inflorescence attributes are more readily seen.

The inflorescence characteristic of *Cynodon* spp. is a digitate or subdigitate (Figure 2.4) panicle of a few to several one-sided narrow racemes arranged in one to a few whorls; spikelets in two rows, ovate or narrow ovate, laterally compressed, one-flowered; glumes are shorter than that of the spikelet; caryopsis ellipsoid, laterally compressed (Bogdan, 1977).



Figure 2.4 The two commonly seen (a) digitate and (b) subdigitate panicle of the *Cynodon* spp. inflorescence (After Rawal and Harlan, 1970).

Varieties of *C. dactylon* are reproduced both sexually by seed and asexually by vegetative propagation. Hybrid bermudagrass varieties are predominantly infertile however, hybrid varieties (*C. dactylon* x C. *transvaalensis*) produce few, if any, viable seeds and are essentially infertile by virtue of chromosomal pairing irregularities involving the two different genomes during meiosis. In practice, reproduction of hybrid bermudagrass varieties is therefore restricted to vegetative (asexual) propagation. Nevertheless, viable seeds theoretically could still occur very infrequently and at an extremely low level of fertility in hybrid bermudagrass, giving it the potential to produce contaminant off-type seedlings, probably on a basis of one in several million spikelets.

Hybridisation of *C. dactylon* var. *dactylon* with a wider range of *Cynodon* species and intraspecific taxa by Harlan et al. (1969) showed an extremely high level of sterility, unlike some of the other genetic combinations in their study. Based on these data, Harlan and de Wet (1969) concluded:

"Cynodon dactylon *is completely isolated genetically from* C. arcuatus, C. barberi, and C. plectostachyus. C. dactylon *can be crossed with* C. aethiopicus, *but with great difficulty and no evidence of such hybrids have been found in nature. Hybrids between* C. dactylon *and the South African species* C. incompletus *and* C. transvaalensis *are easier to produce artificially and occur occasionally in nature. They do not seem to lead anywhere, and we can detect no evidence of introgression among them. The only other species in the genus is* C. nlemfuensis. *Genetic barriers between different races and varieties of* C. dactylon *and* C. nlemfuensis *range from very strong to weak.*"

Bermudagrass is an outbreeding species, reproducing sexually as seed through a high level of cross-pollination because of a self-incompatibility (SI) mechanism which enforces outcrossing (Burton, 1947; Burton and Hart, 1967; Richardson et al., 1978; Taliaferro, 2000). Kenna et al. (1983) and Taliaferro et al. (2004) reported that self-pollinated seed-set varies in bermudagrass, typically ranging from 0.4 to 3.6%. Within a uniform sward of a vegetative variety, the recruitment of a self-pollinated seedling can quickly cause significant issues in relation to non-uniformity of genotype. Modelling by Busey (2009) showed that even a small level of admixture, as little as 0.1%, by a contaminant from the same genus with a 50% faster growth rate could proliferate and increase its level 140 times in just one contaminated planting cycle.

Turf colour is a key component of aesthetic quality and a good indicator of water and nutrient status (Beard, 1973; Karcher and Richardson, 2003). Temperature, photosynthetic activity and full direct sunlight versus shaded environments can all influence turfgrass colour. For example, when temperature drops (particularly during late autumn and winter) Tifdwarf is characterised by a purplish cast (Burton, 1966a) that may be objectionable to some and is easily detected when comparing different varieties growing side-by-side. A change in stolon and/or leaf colour of this kind in Tifdwarf and other varieties, such as 'MS-Supreme' (Krans et al., 1999) or 'CT-2', is the result of the plant developing higher levels of anthocyanin pigmentation. Turfgrass colour for the overall sward is traditionally evaluated by visually rating turf plots on a scale of 1 to 9, with 1 representing yellow or brown turf and 9

representing optimal, dark green turf (Karcher and Richardson, 2003). However, for a more accurate determination of plant or leaf colour, either reference colours such as the 896 individual chips in the 2007 edition of the Royal Horticulture Society (RHS) colour chart or digital image analysis (Karcher and Richardson, 2003) should be used.

2.4 Taxonomic Classification and Species Distribution

The genus *Cynodon* is a member of the family Poaceae (previously Gramineae), subfamily Chloridoideae, tribe Cynodonteae (Bogdan, 1977; Clayton and Renvoize, 1986). A revised taxonomic classification was carried out by Harlan et al. (1970b) after collecting and crossing some 700 accessions said to cover all known taxa within the genus. Their revised classification included eight species and ten varieties of Cynodon, and is still largely followed in more recent literature (de Wet and Harlan, 1970; Harlan, 1970; Harlan et al., 1970c; Rawal and Harlan, 1970; Taliaferro, 2003; Taliaferro et al., 2004b). A second publication produced by Harlan et al. (1970c) recognised nine species and ten varieties of Cynodon (Ho, 1999; Kenworthy et al., 2007; Taliaferro, 1995; Taliaferro, 2003), due to the additional listing of the interspecific natural triploid hybrid C. x magennisii Hurcombe (2n = 27) between C. dactylon and C. transvaalensis (Harlan et al., 1970c). Kang et al. (2008) state that the genus Cynodon genus contains ten species, but did not provide a source for their statement. Since the revised taxonomic treatment published by Harlan et al. (1970b, 1970c), there has since been considerable discussion and argument among taxonomists as to which Cynodon taxa should be accepted and which species should be recognised as valid, particularly in relation to the great natural endemic variability shown among accessions collected worldwide.

One reason why so much confusion over this complex genus remains is that the *Cynodon* species are morphologically similar and, in many cases, are genetically introgressing at several ploidy levels (Sharp and Simon, 2002). With the genus represented in numerous countries worldwide between latitudes 53°N and 45°S, it is still possible to find accessions that are morphologically and genetically dissimilar to those described in previous studies conducted. To complicate the taxonomic situation further, *C. dactylon* resembles large crabgrass/summergrass [*Digitaria sanguinalis* (L.) Scop.], crowsfoot/goosegrass [*Eleusine indica* (L.) Gaertn.], and paspalum/dallisgrass (*Paspalum dilatatum* Poir.) (Hansen, 1918; Mitich, 1989). Essentially, as shown in the earlier pioneering studies by Harlan and coworkers (Harlan and de Wet, 1969; Harlan et al., 1969; Harlan et al., 1970b), taxonomic

treatment of the genus *Cynodon* is extremely difficult such that no entirely satisfactory classification has yet been possible.

More recently, several leading taxonomic authorities worldwide have listed or identified *Cynodon* species that are not clearly identified as being accepted or revised, or are now synonyms of another name and no longer recognised. Some of these groups include: The Royal Botanic Gardens, Kew (Clayton et al., 2002 onwards) listing nine species of *Cynodon*; The Integrated Taxonomic Information System (2001) identifying nine accepted species and two accepted varieties within *Cynodon*; AusGrass (Sharp and Simon, 2002) and AusGrass2 (Simon and Alfonso, 2011) showing seven species and four varieties of *Cynodon* native to or naturalised in Australia; the species nomenclature in Germplasm Resources Information Network (GRIN) taxonomy (USDA et al., 2006a) which identifies thirty-one species of *Cynodon*; Missouri Botanical Garden's Tropicos, Nomenclatural Data base (Tropicos, 2012) listing thirty-two subspecific taxa of accepted *Cynodon*, and The United States Department of Agriculture (USDA) Plants Database (USDA et al., 2006b) classification for Plantae down to the genus *Cynodon* L. C. Rich contains nine species, with twelve accepted taxa overall.

Bryan Simon, former Principal Botanist at Queensland Herbarium, identified four species that need verification as to their authenticity and possibly adding to the "accepted" list of Harlan et al. (1970b); They included *Cynodon barberi* from India, *Cynodon incompletus* from South Africa, *Cynodon mucronatus* from Argentina (synonymised with *C. dactylon* by Clayton) and *Cynodon parviglumis* from the Caroline Islands (B Simon 2006, pers. comm., 17 August).

2.5 Cynodon Turfgrass Varieties Introduced and/or Used in Australia

2.5.1 Vegetative Varieties

Today, the *Cynodon* taxa of major economic importance are the tetraploid *C. dactylon* and the triploid hybrid *C. dactylon* x *C. transvaalensis* turfgrasses used for high and low input turf management applications mainly in the subtropics and warm temperate regions. There is also one hexaploid (2n=56) *C. dactylon* variety, 'Tifton 10', but this and its hybrid progeny, 'Patriot', have not yet been commercialised in Australia.

Within the turfgrass industry, C. dactylon x C. transvaalensis varieties are generally referred to as a hybrid bermudagrass, and include both dwarf and medium- to coarse-textured varieties. With the recent selection of finer, denser varieties, the term "ultradwarf" is also used to identify these new improved greens varieties. 'Tifgreen' was the first fine-textured hybrid suitable for golf and bowling greens, and following its release in the US in 1956 (Hein, 1961) has revolutionised the golf and bowling industries. Tifgreen was soon followed by the dwarf variety 'Tifdwarf' in 1965 (Burton and Elsner, 1965). Together, these two varieties provided the basis from which a number of improved greens grade varieties were selected and released from the mid-1990s onwards (Table 2.1) as ultradwarf varieties; these include 'MS-Supreme', FloraDwarfTM, NovotekTM and MiniVerdeTM. Tifgreen and Tifdwarf are now considered 'old industry standards' in the golf and bowling industries within Australia. White (2006) recognised that hybrid bermudagrasses have had a great history of performance, but also considered their other attributes as well, including great playing quality, ease of weed control, low disease/insect susceptibility (which in some cases may be debatable), exceptional mowing quality, strong recuperative ability, reasonable installation cost, and excellent winter hardiness when properly managed.

The cutting height for couch grass varieties varies depending on their use and/or playability. Dwarf and ultradwarf hybrid bermudagrass varieties which have been selected for use on bowling and putting greens require extremely low cutting heights, e.g. 2-5 mm, with the use of a cylinder mower. The achievable cutting heights have been lowered by the release of ultradwarf varieties selected for their shorter vertical growth, increased shoot density, and finer leaf texture. However, persisting at such a low cutting height for an extended period of time places large amounts of biotic and abiotic stress on the turf and could ultimately result in decline and possibly lead to death.

For other turf uses such as home lawns, parks and sporting stadia, *C. dactylon* or medium- to coarse-textured hybrid bermudagrasses are utilised. There are currently twenty-one *C. dactylon* (Table 2.2) and six medium- to coarse-textured hybrid bermudagrasses (Table 2.1) introduced or currently available commercially in Australia. Mowing heights for these grasses can vary, but they are generally best suited to mowing between 15-30 mm in height. Examples where a lower mowing height is preferred include varieties with a very prostrate growth habit (e.g. Plateau, Mountain GreenTM) or for low-cut sports turf like that on tennis courts or cricket wickets. Routine management practices such as mowing to avoid scalping,

thatch control, top-dressing and a nutritional program are all necessary to maintain or improve turfgrass quality and playability.

A more detailed description of commercially available *Cynodon* spp. varieties (= cultivars) in Australia for turfgrass use at the time of publication can be found in Appendix I.

Variety	Release Date ^{\dagger}	Reported Origin	United States Plant Patent	Australian PBR Application #	Registered Name (PBR/PP)	Experimental Name	Trademark	References
Tifgreen	1956	Seedling cross	-	-	-	Tifton 328	-	(Hein, 1961; Roche and Loch, 2005)
Tifdwarf	1965	Spontaneous mutation from Tifgreen	-	-	-	-	-	(Burton and Elsner, 1965; Roche and Loch, 2005)
Tifway	1960	Chance F1 hybrid	-	-	-	Tifton 419	-	(Hanson, 1959; Burton, 1985)
Santa Ana	1966	Seedling selection of PI 213387	-	-	-	RC145	-	(Hanson, 1959; Augsdorfer, 1995)
FloraDwarf	1995	Spontaneous mutation from Tifgreen	PP9,030	-	FHB-135	FHB-135	FloraDwarf	(Dudeck, 1995a; Dudeck and Murdoch, 1998; Roche and Loch, 2005)
Champion Dwarf	(1996)	Spontaneous mutation from Tifdwarf	PP9,888	1996/203	Champion Dwarf	-	-	(Kaapro, 1999a; Roche and Loch, 2005)
MS-Express	1991	Seed, plant or mutation [■]	PP10,289	-	MS-Express	MSB-20	-	(Krans et al., 1995a; Krans and Philley, 1998a)
MS-Pride	1991	Seed, plant or mutation	PP10,290	-	MS-Pride	MSB-10	-	(Krans et al., 1995b; Krans and Philley, 1998b)
Tift 94	1994	Radiation mutation from Midiron	PP10,079	2001/063	Tift 94	MI40	TifSport	(Hanna, 1997, n.d.1; Hanna et al., 1997); Loch and Hanna, 2001b)
TifEagle	1997	Irradiation of Tifway II	PP11,163	2001/062	TifEagle	TW-72		(Hanna and Elsner, 1999; Hanna, n.d.2; Loch and Hanna, 2001a)

Table 2.1 Origin, release and proprietary protection of C. dactylon x C. transvaalensis varieties available in Australia

Variety	Release Date [†]	Reported Origin	United States Plant Patent	Australian PBR Application #	Registered Name (PBR/PP)	Experimental Name	Trademark	References
MS-Supreme	1999	Spontaneous mutation from Tifgreen	PP11,781	2002/305	MS-Supreme	MSB-40	-	(Krans et al., 1999; Loch and Roche, 2003a)
AGRD	2000	Spontaneous mutation probably from Tifway or Tifgreen	-	2004/299	AGRD	-	-	(Hunt, 1999; Roche and Loch, 2008a)
Patriot	(2002)	F1 hybrid	PP16,801	-	Patriot	OKC 18-4	-	(Anonymous, 2004; Taliaferro et al., 2006)
Novotek	(2003)	Spontaneous mutation from Tifgreen	-	2002/268	TL2	TL2	Novotek	(Loch and Roche, 2003b; Roche and Loch, 2005)
P18	2005	Spontaneous mutation from Tifdwarf	PP12,084	2007/179	P18	P18	MiniVerde	(Kaerwer, 2001; Roche and Loch, 2005; Roche and Loch, 2008b)
Premier	(2007)?	Seed, plant or mutation [■]	PP18,247	-	Premier	-	-	(Parsons and Lehman, 2007; M Engelke 2009, pers. comm., 20 November)

Notes: [†], for informal releases, dates of first commercial use are shown in parentheses.

?, lacking reliable information.

, collected from (i) a seed within the original seed lot, (ii) a seed or plant introduced unintentionally, or (iii) as a mutation.

* United States only

- FloraDwarf was identified by Dudeck and Murdoch (1998) as being a *Cynodon* sp. variety. 'MS-Express' (Krans and Philley, 1998a), 'MS-Pride' (Krans and Philley, 1998b) and MS-Supreme (Krans and Philley, 2001) were identified as being *C. dactylon* x *C. magenissii* varieties. However, for the purposes of this review, the latter varieties have been grouped with the other *C. dactylon* x *C. transvaalensis* varieties.

- 'Premier' is not a *C. dactylon* as first identified by Parsons and Lehman (2007). It is fact a hybrid bermudagrass (*C. dactylon* x *C. transvaalensis*) (M Engelke 2009, pers. comm., 20 November).

Variety	Release Date [†]	Reported Origin	United States Plant Patent	Australian PBR Application No.	Registered Name (PBR/PP)	Experimental Name	Trademark	References
Royal Cape	1930	Natural selection	-	-	-	PI 213387	-	(Busey, 1989)
Greenlees Park	(1969)	Natural selection	-	-	-	-	-	(Beehag, 2006; McMaugh, 1998)
Wintergreen	1983	Seed, plant or mutation	PP6,278	-	C84-135	C84-135	-	(McMaugh and Whiting, 1988; Ho, 1999; McMaugh, 2008)
CT-2	1991	Cross-pollination of selected <i>C. dactylon sp.</i>	PP6,841	610167	CT-2	C84-135	GN-1*	(Whiting, 1988, 1989; McMaugh, 1998)
MS-Choice	1991	Seed, plant or mutation	PP10,332	-	MS-Choice	MSB-30	-	(Krans et al., 1995c; Krans and Philley, 1998c)
FLoraTeX	1992	Introduced into the US from South Africa in 1954	-	-	-	FB-119	FLoraTeX	(Dudeck, 1995b)
C1	1993	Natural Selection	-	-	-	S-49	Legend	(P Semos 2006, pers. comm., 18 August; M Robinson 2008, pers. comm., 16 September)
Windsor Green	1993	Induced mutant of Wintergreen	-	1993/078	Windsor Green	-	-	(McMaugh, 1993, 1996)
Riley's Super Sport	(1995)?	Spontaneous mutation from Greenlees Park	PP11,181	1995/127	Riley's Super Sport	-	Celebration*	(Kaapro, 1996; Riley, 2000)
Riley's Evergreen	(1998)?	Spontaneous mutation from <i>C. dactylon sp.</i>	-	1998/053	Riley's Evergreen	-	Conquest	(Kaapro, 1999b)
Plateau	(2001)	Spontaneous mutation	PP13,059	1998/023	Plateau	-	-	(Brown, 2002; Kaapro, 1999c)

Table 2.2 Origin, release and proprietary protection of vegetative C. dactylon varieties available in Australia

Variety	Release Date [†]	Reported Origin	United States Plant Patent	Australian PBR Application #	Registered Name (PBR/PP)	Experimental Name	Trademark	References
JT1	2002	Seed, plant or mutation	-	2002/282	JT1	-	Hardy Turf	(L Davidson, pers. comm., 29 January 2010; Loch and Roche, 2003c)
TL1	2002	Chance seedling of <i>C.</i> dactylon sp.		2002/267	TL1	TL1	Mountain Green	(Loch and Roche, 2003d; T Anderlini 2009, pers. comm., 30 November)
Oz-E-Green	2006	Spontaneous mutation of chance seedling	PP19474	2004/035	Oz-E-Green	Sir William	OZ TUFF	(Loch and Roche, 2004; Morrow, 2008)
Grand Prix	2006	Cross pollination of Wintergreen and C5	f PP20017	2005/291	Grand Prix	DN12	-	(Roche and Loch, 2006a; Nickson, 2007, 2009)
Winter Gem	2007	Cross-pollination of Wintergreen and C5	f -	2005/290	Winter Gem	DN09	-	(Roche and Loch, 2006b; D Holden 2009, pers. comm., 17 June)
Hatfield	(2008)	Natural selection	-	2002/304	Hatfield	ES302	-	(Loch and Roche, 2003e; W Scattini 2008, pers. comm., 31 August)
WGP3	2011	Open pollination	-	2008/111	WGP3	-	CynoSport	(Paananen, 2008a)
LEG13A	2011	Open pollination	-	2008/110	LEG13A	-	CynoMax	(Paananen, 2008b)

Notes: [†], for informal releases, dates of first commercial use are shown in parentheses.

?, lacking reliable information.

, collected from (i) a seed within the original seed lot, (ii) a seed or plant introduced unintentionally, or (iii) as a mutation.

* United States only

2.5.2 Seeded Varieties

Bermudagrass seed from Australia and Arizona was widely planted for turf in the southern USA in the early 1900s and was a source of some superior plant selections (Taliaferro et al., 2004b). At one time, all the commercial seed used in the USA came from Australia (Tracy, 1917; Busey, 1989), but this market worldwide has long been supplied by large-scale seed production in Arizona and California.

Interest in seeded varieties for higher quality use has increased marginally following the recent development of improved seed-propagated varieties (Baltensperger and Klingenberg, 1994). 'Guymon' and 'NuMex Sahara' were the first varieties released in the US in 1982 (Taliaferro et al., 1983) and 1987 (Baltensperger, 1989) respectively. Guymon was selected for having greater cold tolerance (Baltensperger, 1989), whereas NuMex Sahara was chosen for possessing greater density, drought tolerance and uniformity (Philley et al., 1999). More recently developed seeded varieties also available in Australia include 'Sydney', 'Mohawk', 'Sultan' and 'Transcontinental', but these are not widely used. 'Princess 77' and 'Riviera' are both produced as first-generation seed from vegetative planting of the selected parent lines (cf. all other varieties undergo 3-4 generations of seed multiplication to get to commercial seed) (DS Loch, pers. comm. 2012); they are now the preferred seeded varieties used where higher quality turf is required. Interestingly, Princess 77 is from Arden Baltensperger's use of the Australian gene pool (McMaugh, 2008). However, generic "Speedy Couch" (much of it unlabelled NuMex Sahara because of its high yields – MJ Hills, pers. comm. 2001) still supplies the bulk of the seeded market on the basis of its lower cost compared with the named and labelled seed. It should also be noted that seeded varieties are less competitive and less rhizomatous than their selected vegetative counterparts (Loch 2008), often with poorer sward density (i.e. more open) and less tolerant of wear. However, for a cost effective establishment method of turfgrass, particularly over a large area, seed has a place when compared to the cost of laying full vegetative sod or even sprigging. Nevertheless, establishing turf areas by seed also has its disadvantages (e.g. soil erosion, germination rate, invasion by weeds) which should also be considered when choosing between seed or sod.

2.6 Morphological-Agronomic Testing

Morphological and agronomic studies (Sod Solutions, 2004; Busey and Myers, 1979; Ho, 1999; Hurcombe, 1947; Kang et al., 2008; Kenworthy et al., 2007; Sherman, 1994; Wofford and Baltensperger, 1985; Wu, 2004; Wu et al., 2007) including spaced plant or sward trials have been undertaken on only a limited number of *Cynodon* varieties. Of those studies listed above, only four varieties are available within Australia, TifSportTM, Tifway, Tifgreen and FLoraTeX[®]. The other grasses in these studies were either species or varieties that have not been introduced into Australia, or were clonal accessions of unknown origin (i.e. local collections of *Cynodon* spp. from China and Korea). Roche and Loch (2005) reported on morphological and developmental studies undertaken of seven hybrid bermudagrass varieties as the foundation for further descriptive information to be made available publicly in Australia. This process will assist the wider turf industry in appreciating the differences in growth and development among the wide range of *Cynodon* varieties used in Australia. Future decision making needs to be based on correctly identifying which turfgrass variety will meet their specific needs based on its morphological and developmental characteristics and cultural management information.

Comparative growing trials (similar to those described above) to provide descriptive morphological and developmental data must be undertaken by, or on behalf of, the breeder a new (candidate) variety prior to registration through Australia's Plant Breeder's Rights or New Zealand's Plant Variety Rights system (references for these varieties highlighting their studies can be found in Tables 2.1 and 2.2 and Appendix I). Testing of this nature also allows breeders and other researchers to characterise the available genetic variation and relationships among those varieties closest to their new variety. The comparable US Plant Patent system, however, also allows breeders to make claims based on more subjective and qualitative ratings data (e.g. turf quality and density), and may lack the rigour of data generated under the Australian plant patent system.

The wider objective of collecting and utilising morphological and agronomic data is to provide a structural framework enabling similar genotypes to be grouped and considered together, thereby providing a better defined focus to future breeding and management studies, rather than trying to consider genetic variation and management issues on a variety-by-variety basis. These data also provide a structural genotypic framework within which to assess the results of associated studies of drought, shade, herbicide and wear tolerance, as well as regional adaptation (climate, soil), of the various genotypes.

To obtain more accurate morphological and agronomic measurements of each variety, growing trials should be undertaken at different times of the year (e.g. winter and summer) over multiple years. Avis et al. (1980) and Wu et al. (2007) reported significant genotype x environment interactions for couch grass forage yield, necessitating the use of multiple environment testing through time (years) and space (locations) to more fully characterise the relative genotypic differences.

2.7 Proprietary Protection

There are seven forms of intellectual property rights with the first four used to protect aspects of plant varieties and their propagating material including vegetative and seed. The different forms include utility and plant patents, PBR, trademarks, trade secrets, copyrights, geographical indications and industrial designs (Loch, 1997). Of the sevens forms of intellectual property, PBR is the only one customised specifically for the purposes of protecting new varieties. In Australia, protection in the form of Plant Variety Rights (PVR) was introduced and passed into law in 1987. This legislation was updated in the Plant Breeder's Rights Act 1994, which conformed to the 1991 revision of the International Convention for the Protection of New Varieties of Plants (UPOV Convention) to ensure ongoing consistency and ensuring that the PBR system was in line with changing technologies (Australian Centre for Intellectual Property in Agriculture, n.d.). However, PBR-registered varieties increasingly are also being marketed under a trademark, which protects this marketing name but not the actual material (which is covered by the underlying PBR-registered name). The basic strategy is build up value in the marketing name (trademark) over the first 20 years during which time PBR registration will also protect the actual material. Once PBR protection runs out after 20 years, this enables the breeder/title holder to continue proprietary marketing of a good variety under the recognised marketing name (i.e. the trademark).

2.8 Conclusion

Numerous morphological studies in bermudagrass have been undertaken over the last five decades. However, there is no published research describing what morphological characteristics are important to distinguish between varieties and or species of *Cynodon* or other warm-season turfgrasses. Goals of the present study were to provide detailed methodology on how to collect morphological data, to identify which traits were able to differentiate varieties and determine if the Australian bermudagrasses could be placed into distinct groups.
Chapter 3: Materials and Methods

3.1 Location and Timing of Experiments

Five spaced plant experiments (experiments 1-5) and a single sward experiment (experiment 6) were conducted on a fertile red volcanic ferrosol (= krasnozem) soil at the former Queensland Department of Agriculture, Fisheries and Forestry (DAFF) Redlands Research Facility located in Cleveland (27°32'S lat, 153°15'E long, 25 masl), Redland City, Queensland, Australia between June 2002 and December 2004. Overall, these experiments included 25 different *Cynodon* varieties, not all of which were represented in each experiment as is described in more detail below in section 3.2. Varieties in each of the six experiments user arranged in a randomised complete block design with three replications, but in experiments 1-5 the numbers of spaced plants in each plot varied as noted below.

Experiment 1 included 22 hybrid bermudagrass and *C. dactylon* varieties. There were 10 spaced plants per plot, 1.0 m apart within plot rows and 0.9 m between plots, giving a total of 30 plants per variety. Individual plants were established from rooted nodal cuttings in peat-vermiculite mix before planting in the field on 7 June 2002. Experiment 1 was conducted from mid-winter through spring to mid-summer before finishing on 6 January 2003 (213 DPP).

Experiment 2 included 23 hybrid bermudagrass and *C. dactylon* varieties. There were 5 spaced plants of the same variety per plot (15 per variety overall) on a 1.0 x 1.0 m grid (i.e. 1.0 m apart within plot rows and 1.0 m between plots). Individual 50 mm diameter cores were cut from established swards on 4 March 2003 and planted immediately into the ferrosol soil. The trial ran from late autumn through to late winter, finishing on 21 August 2003 (170 DPP).

Experiment 3 was primarily designed to characterise 15 *C. dactylon* varieties, but also included two medium- to coarse-textured hybrid bermudagrass varieties (Santa Ana, TifSport). Individual plots had 5 plants of the same variety, giving a total of 15 plants per variety. The individual plants were arranged on a 1.5 x 1.5 m grid. Individual plants were acquired in the same manner as in experiment 2 with planting taking place on 25 August 2003. The trial ran through spring and summer, finishing on 16 March 2004 (204 DPP).

Experiment 4 covered 15 *C. dactylon* varieties plus the same two medium- to coarse-textured hybrid bermudagrass varieties as in experiment 3. There were 5 spaced plants of the same variety per plot (15 per variety overall) on a 1.5 x 1.5 m grid. The individual plants were grown from 50 mm diameter cores (as in the previous two experiments) planted on 10 June 2004. Blocks were set up at 1.5 m spacings within plot rows and between plots. The trial ran through winter and spring, finishing in early summer on 15 December 2004 (244 DPP).

Experiment 5 included 9 hybrid bermudagrass varieties with 5 spaced plants per plot (15 per variety overall) grown at 1.0 m spacings within plot rows and between plots. Individual plants were established from 50 mm diameter cores planted on 15 April 2004. The study ran through late autumn, winter and spring, finishing on 15 November 2004 (214 DPP).

Experiment 6 was established as a sward comparison, including 20 *C. dactylon* and hybrid bermudagrass varieties, as a companion study to experiment 1. The 0.9×1.0 m variety plots (3 per variety) were established by planting rooted nodal cuttings at a spacing of 5 x 5 cm on the same date as experiment 1 (7 June 2002). The aim of experiment 6 was: (i) to measure leaf and inflorescence attributes on fertile tillers; and (ii) to enable a comparison of leaf characteristics from a mown sward (experiment 6) with those of an unmown spaced plants (experiment 1). The experiment ran through late winter, spring summer and autumn, finishing on 16 May 2003 (343 DPP).

3.2 Varieties Characterised by Experiment

The six experiments described in section 3.1 generated detailed comparative morphological data for nine *Cynodon dactylon* x *C. transvaalensis* (hybrid bermudagrass) varieties and 16 *Cynodon dactylon* (bermudagrass) varieties. The hybrid bermudagrass varieties were Champion Dwarf, FloraDwarfTM, MS-Supreme, NovotekTM (TL2), Tifdwarf, TifEagle, Tifgreen, Santa Ana and TifSportTM (Tift 94); while the *C. dactylon* varieties and accessions included Common, CT-2, FLoraTeXTM, Greenlees Park, Hatfield, Hardy TurfTM (JT1), Legend[®] (C1), Mountain GreenTM (TL1), OZ TUFF[®] (Oz-E-Green), Plateau, ConquestTM (Riley's Evergreen), Riley's Super Sport, SS2, Windsor Green, Winter Gem and Wintergreen. The varieties tested in each of the six experiments are shown in Table 3.1. Plant material used in these experiments was obtained from the former Queensland Department of Agriculture, Fisheries and Forestry (DAFF) Redlands Research Facility turf demonstration plots located in Cleveland, Redland City, Queensland, Australia (Roche, 2012).

Table 3.1 *Cynodon* spp. varieties that were characterised in spaced plant (experiments 1 to 5) and sward (experiment 6) experiments conducted at DAFF Redlands Research Facility between 2002 and 2004.

Exp.							<i>on s</i> arie		}					Cy	nod	lon		ylor	n va	riet	ies				
	Champion Dwarf	FloraDwarf	MS-Supreme	Novotek	Santa Ana	Tifdwarf	TifEagle	Tifgreen	TifSport	Common	Conquest	CT-2	FLoraTeX	Greenlees Park	Hardy Turf	Hatfield	Legend	Mountain Green	OZ TUFF	Plateau	Riley's Super Sport	SS2	Windsor Green	Winter Gem	Wintergreen
1	•	٠	•	•		•	•	•	•	•	•	•	•	•	•	•	•	•		•	•	•	•		•
2	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•		•	•	•	•		•
3					•				•	•	•	•	•	•	•	•	•	•	•	•	•	•	•		•
4					•				•		•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
5	•	•	•	•	•	•	•	•	•																
6	•	•	•	•		•	•	•		•	•	•	•		•	•	•	•		•	•	•	•		•

In the following chapters, variety names have been abbreviated. These abbreviations can be seen in Table 3.2 along with the respective ploidy level for each variety. Hybrid bermudagrasses are usually triploids (2n = 27) with the notable exception of the tetraploid variety 'Patriot' (which was not included in the present experiments), while the *C. dactylon* varieties studied are tetraploids (2n = 36). Santa Ana and TifSport are classified as medium- to coarse-textured varieties while the other hybrid varieties are either dwarf or ultradwarf varieties as described in Chapter 2.

Abbreviation	Variety	Ploidy
champ	Champion Dwarf	triploid $(2n = 27)$
com	Common	tetraploid $(2n = 36)$
rilgreen	Conquest	tetraploid $(2n = 36)$
ct	CT-2	triploid $(2n = 27)$
florad	FloraDwarf	tetraploid $(2n = 36)$
fltex	FLoraTeX	tetraploid $(2n = 36)$
gem	Winter Gem	tetraploid $(2n = 36)$
gpark	Greenlees Park	tetraploid $(2n = 36)$
hat	Hatifeld	tetraploid $(2n = 36)$
hturf	Hardy Turf	tetraploid $(2n = 36)$
leg	Legend	tetraploid $(2n = 36)$
mount	Mountain Green	triploid $(2n = 27)$
mssup	MS-Supreme	triploid $(2n = 27)$
novo	Novotek	triploid $(2n = 27)$
oz	OZ TUFF	tetraploid $(2n = 36)$
plat	Plateau	tetraploid $(2n = 36)$
rilsport	Riley's Super Sport	tetraploid $(2n = 36)$
santa	Santa Ana	triploid $(2n = 27)$
sstwo	SS2	tetraploid $(2n = 36)$
tifd	Tifdwarf	triploid $(2n = 27)$
tife	TifEagle	triploid $(2n = 27)$
tifg	Tifgreen	triploid $(2n = 27)$
tifs	TifSport	triploid $(2n = 27)$
windsor	Windsor Green	tetraploid $(2n = 36)$
witer	Wintergreen	tetraploid $(2n = 36)$

Table 3.2 Abbreviations of variety names trialled along with their respective ploidy levels.

3.3 Experimental Management

Slow release 18-10-9 NPK fertiliser (275 kg ha⁻¹) was applied at planting, then every 1 to 3-months to maintain a quality turfgrass plant/sward. For longer term experiments, maintenance dressings (210 kg ha⁻¹) of slow release 24-2-9 fertiliser plus 2% Fe was applied to the plots at approximately 3-monthly intervals. Basic weed control was achieved with pre-emergence oxadiazon (Ronstar®) broadcast at 150 kg ha⁻¹ of product. Supplementary control was achieved by regular manual weeding of each experiment, or by fluroxypyr (Starane[®], 750 ml/ha) + metsulfuron (Brush-Off[®]; 15 g ha⁻¹) for broadleaf weeds or spot-spraying with glyphosate (10 ml per L of 45% product) for grass weeds. In experiments 2, 4 and 5, lawn armyworms [Spodoptera mauritia (Boisduval)] and sod webworms [Herpetogramma licarsisalis (Walker)] were controlled with bifenthrin (Baythroid Turf®; 1.25 L ha⁻¹). For experiments 4 and 5 couch mite [Eriophyes cynodonsensis] inhibited plant development early, particularly lateral spread. In order to prevent further 'stunting' or damage to the plants the trial area was sprayed with abamectin (GremlinTM; 1000 ml ha⁻¹). During establishment of experiment 6, plot edges were sprayed with glyphosate at a rate of 10 ml per L of 45% product. to limit cross contamination from adjacent varieties. Plots of both experiments were allowed to develop into swards by growing unchecked until 8 January 2003 to allow for the development of flowering characteristics. From the latter date, the hybrid bermudagrass and C. dactylon varieties were mown to 5 mm and 30 mm respectively and top-dressed lightly with sand (sized to USGA greens specifications) to remove any (minor) surface undulations. Plots of both taxa within experiment 6 were routinely mown at their respective heights until 16 May 2003 when the trial ended. All experiments were irrigated regularly as necessary to maintain optimum growth.

3.4 Data Collection

Approximately 14,248 morphological and developmental data points (Table 3.3) were collected across 6 *Cynodon* experiments (Table 3.1). The characteristics that were grouped into three categories, including growth habit, stolon and shoot/inflorescence traits and measured in each experiment are listed in Tables 3.4 and 3.5.

Table3.3	Summary	of	morphological	and	agronomic	data	points	collected	within	the	six
experiments	s at DAFF l	Red	lands Research I	Facili	ty between 2	2002 a	nd 2004	4.			

Morphological Characteristic Measured	Exp. 1	Exp. 2	Exp. 3	Exp. 4	Exp. 5	Exp. 6	TOTAL
Growth Habit							
Maximum diameter of Spread (dsm)	660	345	285	285	135	-	1710*
Height of sward (hos)	-	-	-	-	-	60	60
Stolon Measurements							
Branches at node 4 (stbnfour)	1200	-	120	90	210	-	1620
Internode four length (stifl)	1200	-	120	90	-	-	1410
Internode four diameter (stifd)	1200	-	120	90	-	-	1410
Internode four sheath length (stifsl)	1200	-	120	90	-	-	1410
Leaf blade at node four length (stlnfl)	1200	-	120	90	-	-	1410
Leaf blade at node four width (stlnfw)	1200	-	120	90	-	-	1410
Leaf and stolon colour	44	46	30	30	18	40	208
Mown internode four diameter (msifd)	-	-	-	-	-	120	120
Mown internode four length (msifl)	-	-	-	-	-	120	120
Mown internode four sheath length (msifsl)	-	-	-	-	-	120	120
Inflorescence Measurements							
Peduncle length (ifpl)	-	-	180	30	-	120	330
Peduncle diameter (ifpd)	-	-	180	30	-	120	330
Average length of spikes (ifalos)	-	-	180	30	-	120	330
Flag leaf sheath length (iffls)	-	-	180	30	-	120	330
Flag leaf blade length (iffll)	-	-	180	30	-	120	330
Flag leaf blade width (ifflw)	-	-	180	30	-	120	330
Fourth leaf sheath length (iffourls)	-	-	180	30	-	120	330
Fourth leaf blade length (iffourll)	-	-	180	30	-	120	330
Fourth leaf blade width (iffourlw)	-	-	180	30	-	120	330
Inflorescence density (id)	-	-	180	30	-	60	270
TOTAL	7904	391	2835	1155	363	1600	14248

Note: Does not take into account missing values e.g. dead plants; * following analysis later in Chapter 4, it was identified that only 1,675 data sets were present when measuring maximum diameter of spread i.e. 35 data points were not accounted for.

Table 3.4 Morphological characteristics measured in spaced plant (experiments 1 to 5) and sward (experiment 6) experiments comparing hybrid bermudagrass varieties at DAFF Redlands Research Facility between 2002 and 2004. Note: the abbreviations shown in parentheses are used in Chapters 4 and 5; numbers within the table refer to experiment numbers.

Cynodon dactylon x C. transvaalensis varieties													
Morphological Characteristic Measured	Champion Dwarf	FloraDwarf	MS-Supreme	Novotek	Tifdwarf	TifEagle	Tifgreen	Santa Ana	TifSport				
Growth Habit													
Maximum diameter of Spread (dsm)	1,2, 5	1,2, 5	1,2, 5	1,2, 5	1,2, 5	1,2, 5	1,2, 5	2,3, 4	1,2, 3,4				
Height of sward (hos)	6	6	6	6	6	6	6	-	-				
Stolon Measurements													
Branches at node 4 (stbnfour)	1,5	1,5	1,5	1,5	1,5	1,5	1,5	-	-				
Internode four length (stifl)	1	1	1	1	1	1	1	-	-				
Internode four diameter (stifd)	1	1	1	1	1	1	1	-	-				
Internode four sheath length (stifsl)	1	1	1	1	1	1	1	-	-				
Leaf blade at node four length (stlnfl)	1	1	1	1	1	1	1	-	-				
Leaf blade at node four width (stlnfw)	1	1	1	1	1	1	1	-	-				
Leaf and stolon colour	1	1	1	1	1	1	1	1	1				
Mown internode four diameter (msifd)	6	6	6	6	6	6	6	-	-				
Mown internode four length (msifl)	6	6	6	6	6	6	6	-	-				
Mown internode four sheath length (msifsl)	6	6	6	6	6	6	6	-	-				
Inflorescence Measurements													
Peduncle length (ifpl)	6	6	6	6	6	6	6	-	-				
Peduncle diameter (ifpd)	6	6	6	6	6	6	6	-	-				
Average length of spikes (ifalos)	6	6	6	6	6	6	6	-	-				
Flag leaf sheath length (iffls)	6	6	6	6	6	6	6	-	-				
Flag leaf blade length (iffll)	6	6	6	6	6	6	6	-	-				
Flag leaf blade width (ifflw)	6	6	6	6	6	6	6	-	-				
Fourth leaf sheath length (iffourls)	6	6	6	6	6	6	6	-	-				
Fourth leaf blade length (iffourll)	6	6	6	6	6	6	6	-	-				
Fourth leaf blade width (iffourlw)	6	6	6	6	6	6	6	-	-				
Inflorescence density (id)	6	6	6	6	6	6	6	-	-				

Table 3.5 Morphological characteristics measured in spaced plant (experiments 1 to 5) and sward (experiment) experiments comparing *Cynodon dactylon* varieties at DAFF Redlands Research Facility between 2002 and 2004. Note: the abbreviations shown in parentheses are used in Chapters 4 and 5; numbers within the table refer to experiment numbers.

	Cynodon dactylon varieties														T	
Morphological Characteristic Measured	Common	Conquest	CT-2	FLoraTeX	Greenlees Park	Hatfield	Hardy Turf	Legend	Mountain Green	OZ TUFF	Plateau	Riley's Super Sport	SS2	Windsor Green	Winter Gem	Wintergreen
Growth Habit																
Maximum diameter of Spread (dsm)	1 2 3	1 2 3 4	3 4	1 2 3 4	1 2 3 4	1 2 3 4	1 2 3 4	4	1 2 3 4							
Height of sward (hos)	6	6	6	6	-	6	6	6	6	-	6	6	6	6	-	6
Stolon Measurements																
Branches at node 4 (stbnfour)	1	1 3	1	1	-	1	1	1	1 3	3	1 3	1	1	1 4	4	1 4
Internode four length (stifl)	1	1 3	1	1	-	1	1	1	1 3	3	1 3	1	1	1 4	4	1 4
Internode four diameter (stifd)	1	1 3	1	1	-	1	1	1	13	3	1 3	1	1	1 4	4	1 4
Internode four sheath length (stifsl)	1	13	1	1	-	1	1	1	13	3	1 3	1	1	1 4	4	1 4
Leaf blade at node four length (stlnfl)	1	1 3	1	1	-	1	1	1	1 3	3	1 3	1	1	1 4	4	1 4
Leaf blade at node four width (stlnfw)	1	1 3	1	1	-	1	1	1	1 3	3	1 3	1	1	4 1 4	4	4 1 4
Leaf and stolon colour	1	1	1	1	1	1	1	1	1 3	3	1 3	1 3	1	1 4	4	1 4
Mown internode four diameter (msifd)	6	6	6	6	-	6	6	6	6	-	6	6	6	6	-	6
Mown internode four length (msifl)	6	6	6	6	-	6	6	6	6	-	6	6	6	6	-	6
Mown internode four sheath length (msifsl)	6	6	6	6	-	6	6	6	6	-	6	6	6	6	-	6
Inflorescence Measurements																
Peduncle length (ifpl)	6	3 6	6	6	-	6	6	6	3 6	3	3 6	6	6	3 6	4	3 6
Peduncle diameter (ifpd)	6	3 6	6	6	-	6	6	6	3 6	3	3 6	6	6	3 6	4	3 6
Average length of spikes (ifalos)	6	3 6	6	6	-	6	6	6	3 6	3	3	6	6	36	4	3 6
Flag leaf sheath length (iffls)	6	3	6	6	-	6	6	6	3	3	3	6	6	3	4	3
Flag leaf blade length (iffll)	6	0 3 6	6	6	-	6	6	6	3 6	3	6 3 6	6	6	0 3 6	4	6 3 6
Flag leaf blade width (ifflw)	6	36	6	6	-	6	6	6	36	3	3	6	6	3	4	3 6
Fourth leaf sheath length (iffourls)	6	3 6	6	6	-	6	6	6	3	3	3 6	6	6	3 6	4	3 6
Fourth leaf blade length (iffourll)	6	3 6	6	6	-	6	6	6	3 6	3	3	6	6	3 6	4	3 6
Fourth leaf blade width (iffourlw)	6	3	6	6	-	6	6	6	3 6	3	3 6	6	6	3 6	4	3 6
Inflorescence density (id)	6	6 6	6	6	-	6	6	6	6	-	6	6	6	6	-	6

3.4.1 Growth Habit Data Collection

Measuring Lateral Spread

For each plant, four measurements (i) the widest diameter of spread taken from stolon tip to stolon tip across the centre of the plant (0° to 180°), (ii) the diameter at right angles to this (90 to 270°), and (iii and iv) two intermediate measurements (45° to 225°, 135° to 315°) between the first two defining measurements (Plate 3.1) were collected following the guidelines by produced Roche and Loch, 2005. Four measurements per plant were initially taken with the premise that the mean value derived from these would give a more accurate representation of lateral spread for the overall plant. However, only the first measurement, the widest diameter of spread or the maximum diameter of spread (dsm) was analysed within the present study. Measurements were made approximately every two weeks once the majority of the plants had commenced active growth across the soil surface (refer to Appendix II for a complete list of dates). Only the final lateral spread measurement has been used within the present study, with the exception of a comparison between the two methods (e.g. cumulative verses final lateral spread measurements).



Plate 3.1 *Cynodon* plant picturing the angle and placement of the four lateral spread measurements which are taken per plant. However, only the first (i), maximum diameter of spread measurement (dsm) was statistically analysed within the present study.

In experiment 1, measurements of lateral spread of the *C. dactylon* and hybrid bermudagrass varieties were undertaken on 19 September 2002 (104 DPP). Lateral spread measurements within experiment 2 were carried out on 17 June 2003 (105 DPP) of the *C. dactylon* varieties and later on the 21 August 2003 (170 DPP) of the hybrid bermudagrasses. For the *C. dactylon* and medium- to coarse-textured hybrid bermudagrass varieties within experiment 3, lateral spread data were collected on 21 November 2003 (88 DPP). Taxa within experiments 4 and 5 which were planted eight weeks apart were measured on 2 November 2004 (229 DPP \pm 4 weeks).

Height of Sward

Sward height was measured as the distance between the soil surface and the apex of a randomly selected flowering tiller. Sward height was determined from one measurement per plant/sward within Experiment 6 only over two days, 16 and 19 December 2002.

3.4.2 Stolon and Shoot Data Collection

The assessment of stolon characteristics varied (Tables 3.4 and 3.5) within spaced plant experiments 1, 3, 4 and 5. The aim was to assess the number of branches present at node 4, length and diameter of the fourth visible internode, the length and width of the leaf blade from the fourth visible node at its longest and widest points, respectively and the sheath length at the fourth visible node (Plates 3.2 and 3.3). Measurements were made on two randomly selected stolons taken from each spaced plant. Within the sward study (experiment 6), measurements of leaf length and width at the fourth visible node of tillers/stolons were recorded after choosing ten randomly selected mature tillers/stolons from each plot. The timing of these assessments for each experiment can be seen in Table 3.6.

The decision to use the fourth visible node (spaced plant experiments and sward experiment) and internode (spaced plant experiments) was based on preliminary measurements showing that cellular expansion in the stolon tip region was virtually complete by this stage, and on the need to minimise the increasing risk of damage to stolon leaves particularly on older nodes (Roche and Loch, 2005).

Exp.	Taxa	Data collection period
1	Bermudagrass	6-15 November 2002
	Hybrid bermudagrass	18-29 November 2002
3	Bermudagrass	17-18 December 2003
4	Bermudagrass	1-2 December 2004
5	Hybrid bermudagrass	9-15 November 2004
6	Bermudagrass and Hybrid bermudagrass	8-16 May 2003
	Hybrid berniudagrass	

Table 3.6 Time period used to assess stolon characteristics of the bermudagrass and hybrid bermudagrass varieties within experiments 1 and 3-6.



Plate 3.2 Measuring position of (a) branches present at node 4 (stbnfour), (b) length and (c) diameter of the fourth visible internode (stifl and stifd respectively), (d) the length and width of the leaf blade from the fourth visible node at its longest and widest points (stlnfl and stlnfw), respectively and the (a) sheath length at the fourth visible node (stifsl) of a *Cynodon* stolon. The five asterisks present, indicate the positioning of each node (1-5).



Plate 3.3 Bermudagrass stolon segment of node four showing (a) sheath length of the outer sheath (stifsl), (b) turfgrass leaf blade length (stlnfl) and (c) width (stlnfw) at the widest point of the blade (measurements a, b and c are all taken of the outer sheath at the fourth visible node), (d) number of branches at node four (stbnfour), and (f) stolon internode colour which is taken of the surface area exposed to sunlight and (b,c) leaf colour of the blade which is also exposed to sunlight.

Leaf and stolon colour

Royal Horticulture Society (2001) colour charts were used to determine stolon (surface area of the fourth internode exposed to sunlight) and fourth primary leaf colours of the varieties trialled in the spaced plant experiments only. The RHS fourth edition (2001) colour charts were used in spaced plant experiments 1, 3 and 4. RHS colour observations were recorded at the following times: experiment 1, 29 October 2002 (spring); experiment 3, 16 March 2004 (autumn) and experiment 4, 1 December 2004 (summer). Refer to Appendix II for RHS colour information collected.

RHS colour was identified of each variety within each experiment. These data were not included within the analysis of data as it was a subjective measurement with potential variation among observers.

3.4.3 Inflorescence Data Collection

Inflorescence characteristics assessed included peduncle length and diameter, average spike length, flag leaf sheath length, flag leaf blade length and width, fourth leaf sheath length, and fourth leaf blade length and width (Plate 3.4). Within the spaced plant experiments 3 and 4 and the sward experiment 6, measurements of these characteristics (with the exception of inflorescence density) were taken from two randomly selected inflorescence-bearing (mature) tillers per spaced plant or sward. Inflorescence density was quantified within sward experiment 6 only, by taking 2 x 0. 1 m² quadrat measurements per plot. The timing of these assessments are given in Table 3.7.

hybrid bermuda	agrass varieties within experiments 3, 4 and 6.	C

Table 3.7 Time period used to assess inflorescence characteristics within of bermudagrass and

Exp.	Taxa	Data collection period
3	Bermudagrass	18 December 2003 and
		12 January 2004
4	Bermudagrass	13-15 December 2004
6	Bermudagrass	16 December 2002
	Hybrid bermudagrass	8 January 2003



Plate 3.4 Measuring position of (a) peduncle length (ifpl) and (b) diameter (ifpd), (c) average spike length (ifalos), (d) flag leaf sheath length (iffls), (e) flag leaf blade length (iffll) and width (ifflw), (f) fourth leaf sheath length (iffourls), and (g) fourth leaf blade length (iffourll) and width (iffourlw).

3.4.4 Excluded Data

Together with the morphological and developmental characteristics identified in Tables 3.4 and 3.5, a range of other traits were measured but were excluded from the study (first and second stage analysis) because of having insufficient data. This was largely for two reasons (i) varieties like OZ TUFF and Winter Gem were trialled only in later experiments because they were new at the time and (ii) because during the early stage of the study (while conducting PBR experiments), the methodology was still being fined tuned as to what characteristics could be measured and what appeared worthwhile, or provided distinctness, uniformity and stability (DUS) for PBR examinations. The list of other traits that were measured during experiments 1 to 6, but were excluded from the present study can be seen in Table 3.8.

Table 3.8 Other morphological-agronomic data that was collected during the present study but was excluded from analysis and write-up.

Other Morphological Characteristic Measured
Growth Habit
Tiller density [†]
Stolon Measurements
Branches at node 2
Branches at node 3
Branches at node 5-15*
Leaf blade at node four length:width ratio
Stolon and leaf blade colour using RHS‡
Inflorescence Measurements
Number of spikes present
Minimum length of spike
Maximum length of spike
Flag leaf blade length:width ratio
Fourth leaf blade length:width ratio

<u>Notes</u>: †, Tiller density was measured during experiment 6 only of the hybrid bermudagrass greens grass varieties. It was decided at the time to discontinue collecting this measure as it was difficult and time consuming to do so; * Braches at node 5-15 was measured in experiment 5 only. The reason behind this is detailed in Roche et al., 2005; ‡ stolon and leaf blade colour data are shown in Appendix III, however the collected data was not included within the analysis of data as it was a subjective measurement with potential variation among observers.

3.5 Data Analysis

The following analyses were done for each trait. Firstly, a spatial analysis was performed for each individual experiment to adjust for any environmental trends using the preferred models as outlined by (Quiao et al., 2004). Briefly, for the spatial analysis all the possible effects were accounted for in the model and dropped if they were found to be insignificant. These analyses provided estimates of the initial value of the residual variances and first order auto-regression coefficients for row and column to be included in a one stage spatial analysis. Varieties were treated as random, therefore, these analyses provided Best Linear Unbiased Predictors (BLUPs) (White & Hodge, 1989) using the method of Residual Maximum Likelihood (REML) (Petterson & Thompson, 1971; Harville, 1977; Searle, 1989). The BLUPs were used to construct a two way table of varieties x traits and

used for a pattern analysis i.e. clustering and ordination. Pattern analysis of the morphologicalagronomic data generated from the 6 experiments containing 25 varieties respectively was performed using R version 2.7.1 (R Development Team, 2008).

Clustering was performed using the Ward linkage method (Ward, 1963) as a strategy and the squared Euclidean distance as a dissimilarity measure. Ordination was performed using a principal component analysis based on a singular value decomposition (Gabriel, 1971). The clustering result was presented as a dendrogram and the ordination was presented as a bi-plot. The group number determined by examining the dendrogram and is often subjective (Freeman, 1985). In this study the square root rule was used as a starting point for determining the number of groups [i.e. the square root of the number of entries (varieties) evaluated] as described by DeLacy (1981)].

A series of boxplots containing information on informative traits and variety groupings were constructed using Microsoft Excel® 2010.

Chapter 4: Australian Cynodon Varieties: Grouping and Characteristics

4.1 Introduction

Several studies (Harlan et al., 1969, Harlan and de Wet, 1969a, Harlan et al., 1970, Jewell et al., 2012) have indicated that the genus *Cynodon* is difficult to treat taxonomically because of the huge morphological variability that exists within each taxa, therefore, no entirely satisfactory classification is possible. Harlan and de Wet (1969b) also described the variety *Cynodon dactylon* var. *dactylon* as containing the remainder of the variation within the species with enormous morphological variation, ranging from very small, fine turfgrasses to large, leafy robust types. It is hypothesised that the morphological variation among functional and commercially available varieties (= cultivars) of bermudagrass or hybrid bermudagrasses in Australia is also large although to date this variation has not been described.

Today, DNA profiling is a common technique used to assist in the identification and grouping (relatedness) of turfgrass species and or varieties. However, this technique is not always clear cut as specie or varietal groupings may not align as predicted due to the existence of large morphological variation, particularly in a complex genus like *Cynodon*. For example, Jewell et al. (2012) analysed genetic variation among 690 *Cynodon* accessions collected from across Australia using expressed sequence tag (EST)-simple sequence repeat (SSR) DNA markers. This study showed that multiple species and ploidy levels were represented within the same clusters and groups. Such variation present within the *Cynodon* taxa was also suggested by Anderson et al. (2009).

Prior to DNA testing, identification and characterisation of genotypes relied on morphological and passport data (e.g. Harlan et al., 1969, Harlan and de Wet, 1969, Harlan et al., 1970b, Hu et al., 2000 and Jewell et al. 2012). For turfgrasses, in particular *Cynodon* spp., numerous studies (e.g. Hurcombe, 1947, Harlan et al., 1969, de Wet and Harlan, 1970, Liu and Guo, 2003, Roche and Loch, 2005, Wu et al., 2007, Kan et al., 2008, Nasiri et al., 2012) have been undertaken to characterise collections using classical taxonomic descriptors.

For the grouping of *Cynodon* spp. using morphological-agronomic descriptors a diverse array of traits have been measured. Fifteen characteristics, including leaf and inflorescence traits, were measured by Liu et al. (2003) to characterize the morphological variation in *C. dactylon* accessions in China. More recently, a study by Nasiri et al. (2012) evaluated the morphological variation of 46 populations (460 individual plants) collected within Iran using a comprehensive list of qualitative and quantitative traits (Table 4.1). However, these traits were very specific and were assessed to provide a morphological description and taxonomic key for the species collected.

Table 4.1 List of evaluated qualitative and quantitative traits assessed by Nasiri et al. (2012) within their *Cynodon dactylon* collection made within Iran. Refer to Nasiri et al. (2012) for detailed information on the qualitative traits (characters) measured.

Qualitative Traits	Quantitative Traits
Stem node and internode hairs	Plant height
Basal hairs of inflorescence	Upper internode length
Hairs on inflorescence base	Inflorescence length
Position of spikes on the peduncle	Number of rachis (inflorescent branches)
Leaf upper surface hairs	Spike length
Density of hairs on leaf upper surface	Number of rachis veins
Shape of leaf upper surface hairs	Spikelet length
Leaf lower surface hairs	Spikelet width
Density of hairs on leaf lower surface	Number of spikelets per inflorescence
Shape of leaf on lower surface hairs	Upper glume length
Outer surface hairs of leaf sheath	Upper glume width
Hairs density on outer surface of leaf sheath	Lower glume length
Ligule marginal ornamentations	Lower glume width
Auricle	Ratio of glume to floret
Pedicle	Lemma length
Spikelet colour	Lemma width
Sterile floret	Number of veins on lemma
Spike colour	Palea length
Glume colour	Palea width
Lemma tip shape	Number of keel on palea
Lemma colour	Lodicule size
Lemma abaxial surface hairs	Number of lodicules
Hairs of central vein on lemma	Number of stamens
Lemma marginal ornamentations	Caryopsis length
Palea colour	Caryopsis width
Stigma colour	Ratio of density to caryopsis length

Of the 52 qualitative and quantitative traits collected by Nasiri et al. (2012) the authors concluded that for species identification presence of hair on leaf/sheath surfaces and spike colour had a major role on grouping at the species level. Other stable (similar to distinct, uniform and stable; part of the Australian PBR process) features included presence of pilose hairs on leaf surfaces and outer surface of leaf sheath (Nasiri et al., 2012). The latter examples are where the authors wanted to distinguish varieties within the species *C. dactylon*. There is no reported evidence of grouping attempts between varieties of functional (known) bermudagrass or hybrid bermudagrasses.

In the present study, morphological data collected from a series of spaced and sward experiments was used to characterise 25 varieties of *Cynodon* taxa that are available commercially in Australia. A series of dendrograms (unrooted trees) and bi-plots are presented based on growth habit, stolon, shoot and inflorescence characteristics as described in Chapter 3.

4.1.1 Biplot Interpretation

The term 'biplot' was introduced by Gabriel in 1971 (Gabriel, 1971). Biplots provide a 2-D or 3-D visual display of matrices containing detailed information of the main effects and interaction between two variables. The matrices which are represented by a vector for each row (e.g. variety) and another vector for each column (e.g. trait) allows for a simultaneous display of main effects and interactions. Biplots are especially revealing in principal component analysis, where the biplot can show inter-unit distances and indicate clustering of units as well as display variances and correlations of the variables collected (Gabriel, 1971).

In the present study, the use of biplots was the preferred method to display large amounts of collected data graphically showing interactions between varieties and traits. However, sometimes biplots can be difficult to interpret. The aim of this section was to provide background information on biplots and how to best interpret them.

A segment containing two traits 'dsm' (maximum diameter of spread) and 'stifl' has been selected from a 3D biplot, however only principal components (PCs) 1 and 2 are shown on a 2D plane (Figure 4.1). This Figure has been modified from Figure 4.3. The following information, including the references to the labels ('a' to 'l' in Figure 4.1), is aimed to provide a greater understanding of how to interpret a 2D biplot. Points to consider when interpreting a biplot are:

 the axes of a biplot are a pair of principal components, PC1 and PC2 (for this 2D example). Axes contain values proportional to the mean data collected for the variables. Each axis contains a zero value ('k'). The intersection of the latter two axes is the mean value for each variable measured, otherwise known as the graph origin;

- a biplot uses points (e.g. 'l') to represent the scores of the observations on the principal components and it uses vectors (e.g. 'b' and 'd') to represent the coefficients of the variables on the principal components (Young, 1999). In this example, the points represent turfgrass varieties and the vectors morphological traits. Varietal groupings (VarGr) based on traits have also been included which are derived following statistical analysis;
- 'b' and 'd' are vectors for dsm and stifl, respectively. Varietal or group values are obtained by dropping a perpendicular line to each vector (trait). Values that drop below the origin of the vector are less than the mean while values that fall above the origin are greater than the mean. For example, the relative differences between variety groups 2 ('g'), 3 ('h') and 4 ('j'), can clearly be seen in Figure 4.1;
 - 'dsm' Variety Group (VarGr) 1 ('f') had the highest average dsm and therefore the value cut the dsm vector high along the positive direction whereas perpendicular values for VarGr 3 ('h'), VarGr 2 ('g') and VarGr 4 ('i'), were all below the average (i.e. the origin). The ranking and scale of scores among the variety groups for 'dsm' are consistent with those observed in Table 4.2b;
 - 'stifl' The same variety group ranking can be seen as 'dsm' above;
- the closer the angle ('a') between vectors the greater the correlation between the traits; and
- the length of the vector is proportional to the variance explained by the corresponding variable. The shorter the vector, the smaller the contribution to variation between the varieties.



Figure 4.1 A section taken from Figure 4.3 showing principal components 1 and 2 of the two traits 'dsm' (maximum diameter of spread) and 'stifl' (stolon internode four length). Turf varieties OZ TUFF (oz), Plateau (plat) and Conquest (rilgreen) are also shown.

4.2 Results

Following pattern analysis of all the collected morphological-agronomic data, as described in Chapter 3 (Materials and Methods), mean values of each trait were obtained for the 25 varieties trialled (Table 4.2a). There was genetic variation for all traits (Table 3.3) measured except 'inflorescence – peduncle length' (ifpl) which showed no variation and was therefore not included in the present dendrograms or biplots. A combined group matrix was also generated for the 'variety groups' following truncation at the four group level (Table 4.2b).

(a)							Vai	riety Triall	ed					
	(a)	champ	com	ct	florad	fltex	gem	gpark	hat	hturf	leg	mount	mssup	novo
	dsm	-218.80	233.40	501.80	-279.00	91.26	-271.60	259.70	127.00	195.00	13.86	-457.80	46.87	-308.00
	stbnfour	0.38	-0.63	0.23	0.17	-0.62	NA	NA	-0.47	-0.48	0.81	0.08	0.17	0.54
-	stifl	-17.12	16.97	15.56	-19.99	13.64	NA	NA	6.02	15.29	17.27	-14.55	-14.60	-19.56
nrec	stifd	-0.53	0.27	-0.05	-0.42	0.23	NA	NA	0.27	0.37	0.27	0.10	-0.48	-0.46
easi	stifsl	-4.49	4.75	1.23	-4.88	3.60	NA	0.00	3.13	4.67	2.55	-2.60	-4.14	-4.41
ž	stinfi	-1.41	1.24	3.45	-2.75	1.55	NA	NA	0.58	1.97	0.90	-0.94	-2.09	-1.08
Characteristics Measured	stInfw	-0.02	-0.35	0.46	-0.29	-0.13	NA	NA	-0.14	0.16	0.60	0.56	-0.06	0.18
eris	hos	-21.42	-3.37	-11.44	-17.02	42.14	NA	NA	-7.51	20.23	-36.42	-4.29	-9.28	4.90
act.	id	-89.92	114.70	-37.96	-67.73	-29.89	NA	NA	-16.45	-4.73	156.40	105.90	-59.75	74.26
Chai	msifd	-0.20	0.06	-0.02	-0.19	0.38	NA	NA	-0.04	-0.02	-0.02	0.22	-0.20	0.02
iic (msifl	-2.73	1.15	1.71	-2.42	3.03	NA	NA	-1.33	1.44	-0.67	1.55	-2.21	-2.24
νοι	msifsl	-0.34	0.80	-1.25	-0.47	0.91	NA	NA	-0.30	0.86	0.84	-0.24	-0.86	-0.29
gror	ifalos	-2.37	0.27	-2.27	-1.34	1.40	NA	NA	1.26	0.57	0.58	0.04	-1.39	-0.55
βA-I	iffll	-0.77	1.72	-9.30	-4.30	18.63	NA	NA	0.44	2.93	-6.13	-2.26	-4.44	-5.24
gica	iffls	-7.71	3.81	-8.29	-4.42	1.76	NA	NA	0.30	3.02	1.34	1.07	-6.51	-0.87
golo	ifflw	-0.02	-0.01	-0.01	0.00	0.05	NA	NA	0.00	0.00	0.00	-0.01	-0.02	-0.02
phe	iffourll	-7.62	10.00	-8.83	-3.40	17.24	NA	NA	2.27	7.95	-2.64	-2.67	-4.97	-0.82
Morphological-Agronomic	iffourls	-4.08	1.91	-4.84	-3.02	3.60	NA	NA	3.18	1.80	-0.36	1.10	-3.14	-0.21
_	iffourlw	-0.15	0.05	-0.14	-0.02	0.16	NA	NA	0.07	0.17	0.07	0.07	-0.01	0.06
	ifpd	-0.05	0.10	-0.04	0.00	0.10	NA	NA	0.02	0.13	0.00	-0.02	-0.04	-0.04
	ifpl	0.00	0.00	0.00	0.00	0.00	NA	NA	0.00	0.00	0.00	0.00	0.00	0.00

Table 4.2 (a) combined matrix of the traits and turfgrass varieties trialled following pattern analysis and (b) the combined group matrix containing BLUPs (the mean values equal zero).

		Variety Trialled (Cont'd)											
		OZ	plat	rilgreen	rilsport	santa	sstwo	tifd	tife	tifg	tifs	windsor	witer
	dsm	-298.00	-124.30	373.20	75.64	-233.80	276.90	-221.00	-195.00	303.70	-221.20	-53.37	177.10
	stbnfour	NA	-0.16	NA	-0.34	-0.86	-0.61	0.60	0.73	0.52	NA	-0.21	0.14
-	stifl	NA	0.79	35.59	3.65	NA	9.70	-18.28	-17.81	-6.37	NA	-7.10	0.90
nree	stifd	0.18	0.33	0.21	0.39	NA	0.32	-0.39	-0.33	-0.37	NA	-0.01	0.09
easi	stifsl	-1.20	1.61	4.90	3.37	0.00	3.57	-4.41	-4.54	-2.57	0.00	-0.51	0.35
Š	stinfi	1.02	-0.75	2.99	0.84	NA	0.10	-1.15	-1.33	1.65	NA	-2.65	-2.14
tics	stlnfw	-0.17	0.16	0.53	-0.27	NA	-0.12	0.00	0.25	-0.16	NA	-0.57	-0.62
eris	hos	NA	-13.94	-2.31	43.39	NA	-22.25	-5.17	-10.72	14.72	NA	14.33	25.44
ract	id	NA	-47.47	-43.35	-7.90	NA	-38.04	180.20	-30.51	17.64	NA	-96.58	-78.76
Chai	msifd	NA	0.16	0.15	-0.01	NA	0.08	0.00	-0.15	-0.06	NA	-0.01	-0.16
nic C	msifl	NA	2.17	0.86	6.40	NA	0.04	-1.43	-2.11	0.72	NA	0.46	-4.38
L DOL	msifsl	NA	0.47	-0.68	1.46	NA	-0.30	-0.01	0.22	0.04	NA	0.54	-1.40
gror	ifalos	-0.16	1.39	0.51	4.86	NA	0.95	-0.78	-1.84	-0.53	NA	-1.01	0.40
Ι-Α _ε	iffll	4.30	NA	1.58	18.45	NA	-0.55	-6.59	-8.50	-5.56	NA	-3.01	8.62
gica	iffls	1.63	3.92	6.81	12.98	NA	-0.31	-2.08	-5.57	-0.30	NA	-0.72	0.14
olog	ifflw	0.04	0.01	0.02	0.05	NA	0.00	-0.02	-0.03	-0.01	NA	-0.01	-0.01
Morphological-Agronomic Characteristics Measured	iffourll	5.09	-2.03	3.97	11.46	NA	-3.78	-2.75	-7.47	0.61	NA	-13.67	2.07
Mo	iffourls	NA	-0.70	2.25	9.93	NA	0.35	-1.91	-3.57	0.06	NA	-4.22	1.89
<u> </u>	iffourlw	NA	0.02	0.06	0.14	NA	-0.09	-0.06	-0.17	-0.02	NA	-0.12	-0.08
	ifpd	NA	0.06	0.04	0.09	NA	-0.02	-0.04	-0.07	-0.05	NA	-0.12	-0.07
	ifpl	0.00	0.00	0.00	0.00	NA	0.00	0.00	0.00	0.00	NA	0.00	0.00

(b)		Morphological-Agronomic Characteristics Measured										
		dsm	stbnfour	stifl	stifd	stifsl	stinfi	stInfw	hos	id	msifd	msifl
0.0	1	365.00	-0.07	14.57	0.05	2.47	2.11	0.00	-3.32	0.00	0.00	0.67
iety pin	2	120.90	-0.11	7.32	0.13	2.16	0.40	0.00	12.33	0.00	0.00	0.65
Vari	3	-214.20	0.05	-6.91	0.09	-1.14	-1.53	0.00	-3.36	0.00	0.00	-0.11
- 0	4	-271.70	0.13	-14.98	-0.26	-3.48	-0.98	0.00	-5.66	0.00	0.00	-1.21

		Morphological-Agronomic Characteristics Measured (Cont'd)									
		msifsl	ifalos	iffll	iffls	ifflw	iffourll	iffourls	iffourlw	ifpd	ifpl
20	1	-0.23	0.52	-1.10	0.80	0.00	2.55	0.17	-0.03	0.01	0.21
Variety irouping	2	0.43	4.95	6.26	2.66	0.00	7.12	3.59	0.13	0.04	1.26
Var Grou	3	0.01	-1.21	-3.65	-0.49	0.00	-6.54	-3.53	-0.07	-0.04	-1.19
. 0	4	-0.21	-4.25	-1.51	-2.97	0.00	-3.13	-0.23	-0.03	-0.01	-0.28

<u>Variety abbreviations</u>: Hybrid bermudagrass - Champion Dwarf (champ), FloraDwarf (florad), MS-Supreme (mssup), Novotek (novo), Santa Ana (santa), Tifdwarf (tifd), TifEagle (tife), Tifgreen (tifg) and TifSport (tifs). Bermudagrass - Common (com), Conquest (rilgreen), CT-2 (ct), FLoraTeX (fltex), Winter Gem (gem), Greenlees Park (gpark), Hatfield (hat), Hardy Turf (hturf), Legend (leg), Mountain Green (mount), OZ TUFF (oz), Plateau (plat), Riley's Super Sport (rilsport), SS2 (sstwo), Windsor Green (windsor) and Wintergreen (witer).

<u>Trait abbreviations</u>: Growth habit – maximum diameter of spread (dsm) and height of sward (hos); Stolon measurements – branches at node 4 (stbnfour), internode four length (stifsl), internode four sheath length (stifsl), leaf blade at nods four length (stlnfl), leaf blade at node four width (stlnfw), mown internode four diameter (msifd), mown internode four length (msifl) and mown internode four sheath length (msifsl); Inflorescence measurements – peduncle diameter (ipd), average length of spikes (ifalos), flag leaf sheath length (iffls), flag leaf blade length (iffll), flag leaf blade width (ifflw), fourth leaf sheath length (iffourls), fourth leaf blade length (iffourll), four leaf blade width (ifflw), and inflorescence density (id).

4.2.1 Morphological Comparison

A dendrogram of 25 varieties was used to graphically represent a hierarchical cluster analysis of the relationship between varieties based on morphological-agronomic descriptors. Clustering of the 25 *Cynodon* spp. varieties was truncated at the 4 group level using data from the spaced plant and sward experiments (Figure 4.1). Four groups were identified at 22% dissimilarity (Figure 4.1) by using the square root rule (DeLacy, 1981) (where the number of groups = the square root of the number of varieties evaluated). Although this rule was not explicitly imposed; because the group containing the varieties SS2 (sstwo), Greenlees Park (gpark), and Common (com) had only a few members and it was then decided to take a more conservative approach with grouping and truncate to four groups rather than five because this resulted in more members (varieties) per group.



Figure 4.2 Dendrogram of *Cynodon* spp. varieties derived from pattern analysis of morphological-agronomic data collected from the 6 experiments. Four 'variety groups' are shown based on the truncation of the dendrogram.

<u>Variety abbreviations</u>: Hybrid bermudagrass - Champion Dwarf (champ), FloraDwarf (florad), MS-Supreme (mssup), Novotek (novo), Santa Ana (santa), Tifdwarf (tifd), TifEagle (tife), Tifgreen (tifg) and TifSport (tifs). Bermudagrass - Common (com), Conquest (rilgreen), CT-2 (ct), FLoraTeX (fltex), Winter Gem (gem), Greenlees Park (gpark), Hatfield (hat), Hardy Turf (hturf), Legend (leg), Mountain Green (mount), OZ TUFF (oz), Plateau (plat), Riley's Super Sport (rilsport), SS2 (sstwo), Windsor Green (windsor) and Wintergreen (witer).

4.2.2 Characterisation of the Groups

To present results from the principal component analysis and expand on the varietal grouping data shown in Figure 4.2, a 3D biplot is shown in 2D (Figure 4.3) containing varietal and trait relationships based on the combined group mean data (BLUPS) from the 6 experiments. The first three principal components (PC1, PC2 and PC3) accounted for 74.3% of the total variation in the data. The biplots show that there are clear differences between the 4 variety groups, although groups 3 and 4 are relatively close together and group 2 is more distant from all others. The variety groupings were as follows:

VarGr 1 consisted of 6 varieties, 5 of which were bermudagrass varieties (CT-2, Riley's Evergreen, SS-2, Greenlees Park and Common) and the sixth was the first-generation hybrid bermudagrass greens quality variety Tifgreen;

VarGr 2 – Similar to the former grouping, VarGr 2 also contained a greens quality variety, MS-Supreme, a second-generation ultra-dwarf variety. The other 6 *C. dactylon* varieties included Wintergreen, Hardy Turf, Hatfield, Riley's Super Sport, FLoraTeX and Legend;

VarGr 3 – This group contained 4 varieties, one of which was also an ultra-dwarf variety, TifEagle. The other 3 varieties were *C. dactylon* and included Windsor Green, Mountain Green and Plateau. The latter two varieties in particular possess wide leaf blades on the stolon (stlnfw) which was consistent with the observations for these varieties described by Loch et al. (2003d);

VarGr 4 – This group was the most diverse of all 4 groups. Group 4 contained 3 types, *C. dactylon* (OZ TUFF and Winter Gem), first- (Tifdwarf) and second-generation (Novotek and FloraDwarf) *Cynodon* hybrid greens quality grasses and medium-coarse textured hybrid bermudagrass varieties (Santa Ana and TifSport).

To analyse the association and importance of each variety group to particular traits, the evaluated characteristics have been grouped into 3 morphological features. They include:

- Growth habit: maximum diameter of spread (dsm), height of sward (hos);
- Stolon and shoots: internode four sheath length (stifsl), leaf blade at node four length (stlnfl), internode four diameter (stifd), internode four length (stifl), mown internode four diameter (msifd), mwon internode four length (msifl), mown internode four sheath length (msifsl); and
- Inflorescence: flag leaf length (iffll), fourth leaf blade length (iffourll), fourth leaf sheath length (iffourls), flag leaf sheath length (iffls), average length of spike (ifalos), inflorescence density (id).

Generally speaking there was a strong correlation among stolon and among shoot characteristics and a stronger correlation among inflorescence characteristics, with the exception of inflorescence density (id) as seen in Figure 4.3. Inflorescence density is a shorter vector that lies close to the origin (0,0) and therefore does not explain much of the variation between varieties, contrary to Figure 4.4 where the trait is located on its own branch. Inflorescence density appears to be correlated to maximum diameter of spread (dsm) but PC2 v PC3 shows they are not highly correlated. Inflorescence density, sward height (hos), stolon internode four length (stifl) and maximum diameter of spread were unique traits in that, individually these traits did not group with any other trait (Figure 4.4). Stolon branching at node four (stbnfour) displayed an interesting position within Figure 4.3. Based on the PC1 v PC2 biplot (Figure 4.3), stolon branching at node four (stbnfour) is a trait which is clearly separated from all others and is negatively correlated to almost all the inflorescence traits. Also, stolon branching at node four does not contribute to the variation accounted for in PC2 unlike most of the traits trialled. Vectors that lie close to the origin e.g. leaf blade at node four width (stlnfw) indicate in this case that variation in stlnfw did not contribute to the variation explained by PC1, but most of the other traits did. Inflorescence traits (commencing with 'if') also appeared to be highly correlated in Figure 4.3.



Figure 4.3 3D biplot displayed on a 2D plane showing variety and attribute relationships based on combined group mean data from the 6 experiments. Four variety groups are shown which can also be seen in Figure 4.2. Traits measured are attached to the vectors, while the varieties are positioned independently (no lines).

<u>Trait abbreviations</u>: Growth habit – maximum diameter of spread (dsm) and height of sward (hos); Stolon measurements – branches at node 4 (stbnfour), internode four length (stifsl), internode four sheath length (stifsl), leaf blade at nods four length (stlnfl), leaf blade at node four width (stlnfw), mown internode four diameter (msifd), mown internode four length (msifl) and mown internode four sheath length (msifsl); Inflorescence measurements – peduncle diameter (ipd), average length of spikes (ifalos), flag leaf sheath length (iffls), flag leaf blade length (iffll), flag leaf blade width (ifflw), fourth leaf sheath length (iffourls), fourth leaf blade length (iffourll), four leaf blade width (ifflw), and inflorescence density (id).

The mean variety group values displayed in Table 4.3 clearly highlight the morphologicalagronomic variation observed within the *Cynodon* varieties trialled. The variety groups (within Table 4.3) have characteristics that can be easily described. For example; VarGr 1 – maximum diameter of spread (dsm) was higher than other groups as were many of the stolon (st-) and inflorescence (if-) traits; VarGr2 – sward height (hos), stolon internode four diameter (stifd), average length of spike inflorescence (ifalos) and inflorescence fourth leaf blade width (iffourlw) were larger than other groups; VarGr 3 – values tended to be intermediate between variety groups1/2 and group 4; and VarGr 4 - The latter group which comprised the most varieties of *Cynodon* spp. saw the largest production of seedheads/inflorescence (117.6/0.01m² ±124.5) and the smallest values for many traits.

Table 4.3 Variety grou	up means together with	their standard errors	of the 21 morphological traits
measured.			

	Variety						
	Group 1	Group 2	Group 3	Group 4			
dsm	1439.05 (± 544.05)	1206.71 (± 496.77)	799.88 (± 416.97)	736.80 (± 330.71)			
stbnfour	2.31 (± 0.70)	2.21 (± 0.74)	2.74 (± 0.51)	2.70 (± 0.68)			
stifl	44.49 (± 14.29)	39.75 (± 12.40)	24.71 (± 10.59)	17.05 (± 9.33)			
stifd	1.43 (± 0.30)	1.52 (± 0.29)	1.48 (± 0.28)	1.06 (± 0.36)			
stifsl	10.17 (± 2.96)	10.06 (± 2.76)	7.17 (± 2.54)	4.79 (± 2.26)			
stlnfl	8.43 (± 2.33)	7.03 (± 1.98)	5.35 (± 1.43)	5.16 (± 2.16)			
stlnfw	2.16 (± 0.38)	2.07 (± 0.30)	2.28 (± 0.59)	2.07 (± 0.34)			
hos	190.53 (± 66.43)	197.24 (± 84.89)	112.92 (± 116.01)	41.58 (± 18.07)			
id	116.00 (± 88.41)	90.10 (± 85.75)	100.13 (± 98.09)	117.58 (± 124.50)			
msifd	1.91 (± 0.28)	1.88 (± 0.37)	1.89 (± 0.36)	1.43 (± 0.11)			
msifl	22.29 (± 7.02)	20.08 (± 7.45)	15.17 (± 7.27)	7.28 (± 0.94)			
msifsl	9.67 (± 2.67)	9.70 (± 2.91)	7.73 (± 2.63)	4.33 (± 0.46)			
ifalos	37.93 (± 12.22)	38.06 (± 13.37)	23.95 (± 8.99)	17.13 (± 5.45)			
iffll	20.72 (± 11.03)	22.73 (± 12.72)	11.73 (± 7.98)	7.69 (± 6.53)			
iffls	56.15 (± 17.66)	53.42 (± 15.61)	40.10 (± 12.12)	32.79 (± 13.47)			
ifflw	1.33 (± 0.35)	1.39 (± 0.37)	1.23 (± 0.32)	1.06 (± 0.51)			
iffourll	43.85 (± 16.28)	41.95 (± 16.41)	24.35 (± 8.60)	21.12 (± 7.33)			
iffourls	19.28 (± 5.49)	18.73 (± 6.45)	12.61 (± 3.34)	9.73 (± 2.48)			
iffourlw	1.90 (± 0.36)	2.00 (± 0.39)	1.95 (± 0.37)	1.64 (± 0.46)			
ifpd	0.58 (± 0.12)	0.58 (± 0.10)	0.50 (± 0.06)	0.44 (± 0.05)			
ifpl	85.10 (± 33.19)	79.77 (± 30.52)	58.75 (± 30.67)	52.71 (± 29.60)			

<u>Notes</u>: With the exception of 'dsm' (cm), 'stbnfour '(count) and 'id' (inflorescence count/0.01 m²) all other traits are in millimetres (mm). Value shown in parenthesis is the standard error.

4.2.3 Trait Grouping and Informative Traits

In the dendrogram based on traits studied (Figure 4.4), characteristics were divided in 5 'trait groups' after applying the square root rule. However, like that of the variety grouping, the square root rule was not strictly enforced, and an additional group was chosen to provide 6 trait groups, not 5. If an additional trait group was not chosen the majority of the dendrogram, (now) trait groups 3 and 4 would have been grouped as one. This decision would have encompassed 16 traits out of the 21 traits trialled less 'ifpd' that showed no variation as discussed earlier.

The trait groupings in Figure 4.4 contain the following characteristics:

Trait groups 1 and 2 – contained single growth habit traits, maximum diameter of spread (dsm) and height of sward (hos) respectively;

Trait group 3 – contained four inflorescence morphological traits (iffll), (iffourll), (iffourls) and (iffls);

Trait group 4 – possessed the largest number of morphological traits, twelve, containing a mixture of stolon, inflorescence and mown sward traits; and

Trait groups 5 and 6 - like that of trait groups 1 and 3, trait groups 5 and 6 also contained only one morphological trait each; stolon internode four length (stifl) and inflorescence density (id).



Figure 4.4 Dendrogram of characteristics trialled derived from pattern analysis of morphological-agronomic data collected in the 6 experiments. The dashed horizontal line indicates the grouping based on informative traits. The latter traits 'trait groups' are numbered 1-6. Each trait group contains one informative trait which is highlighted with dotted lines.

<u>Trait abbreviations</u>: Growth habit – maximum diameter of spread (dsm) and height of sward (hos); Stolon measurements – branches at node 4 (stbnfour), internode four length (stifl), internode four sheath length (stifsl), leaf blade at nods four length (stlnfl), leaf blade at node four width (stlnfw), mown internode four diameter (msifd), mown internode four length (msifl) and mown internode four sheath length (msifsl); Inflorescence measurements – peduncle diameter (ipd), average length of spikes (ifalos), flag leaf sheath length (iffls), flag leaf blade length (iffll), flag leaf blade width (ifflw), fourth leaf sheath length (iffourls), fourth leaf blade length (iffourll), four leaf blade width (ifflw), fourth leaf sheath length (iffourls), fourth leaf blade length (iffourll), four leaf blade width (ifflw), fourth leaf sheath length (iffourls), fourth leaf blade length (iffourll), four leaf blade width (iffourlw) and inflorescence density (id).

4.3 Discussion

Variety Grouping

The 25 *Cynodon* spp. varieties and 21 traits measured (Figure 4.2) investigate in this study resulted in 4 varietal groups of Australian *Cynodons*. Within each of the groups there was significant overlapping of tetraploid *C. dactylon* (2n = 36) and triploid *Cynodon* hybrid (2n = 27) varieties.

VarGr 1 contained varieties that were relatively spreading (dsm) and also produced long stolon internodes (stifl) (Table 4.3). Loch et al. (2009) alluded to a correlation between internode length and lateral spread of a series of *Stenotaphrum secundatum* (St Augustine Grass (USA)/buffalograss (AUS)) varieties in morphological studies i.e. slow-spreading plants had shorter internodes and genotypes that spread rapidly had long internodes. However, within the present study conducted on *Cynodon* taxa, the collected data showed only a moderate ($R^2 = 0.576$) (Figure 4.5) correlation between lateral spread and internode length. The outliers within Figure 4.5 include the bermudagrass varieties Legend, Common, FLoraTeX and Hardy Turf which are all from the same trial, experiment 1. If these outliers are removed a stronger relationship ($R^2 = 0.83$) is observed. The latter varieties are not from the same morphological variety group identified within the present study (Figure 4.2).



Figure 4.5 Correlation of available mean diameter of spread data and stolon internode four length data collected from spaced plant experiments 1, 3 and 4 where both traits were measured. Mean data is shown of 21 varieties of *Cynodon* spp.

Cynodon species were spread across variety groups as identified above; this can also be seen within genetic fingerprinting of genotypes within collections. For example, Jewell et al. (2012) identified that multiple species, ploidy levels, and geographic regions were represented within clusters and

groups of the collected Australian *Cynodon* accessions (some 690) following DNA testing. It should also be noted that clustering will also be dependent on what varieties or genotypes are being analysed. For example, the analysis of a diverse collection of native (wild) genotypes with a widespread geographic location; compared to known turfgrass varieties (e.g. as listed in Appendix I), or selections made from native genotypes on the basis of desired traits (e.g. good lateral spread, produces a dense sward, dark green in colour) are likely to give different outcomes.

Trait Grouping and Informative Traits

Six trait groups were identified following pattern and data analysis (Figure 4.4). Upon investigation of Figure 4.4, 4 traits, including maximum diameter of spread (dsm), height of sward (hos), stolon internode four length (stifl) and inflorescence density (id), were located on their own branch of the dendrogram highlighting their uniqueness. Located on branches 3 and 4 (branch numbering left to right) were 4 and 12 traits respectively. One trait from each of these two branches was selected to form a list of informative traits that can be used for future PBR trials or experiments that aim to determine the morphological variation among groups of *Cynodon* genotypes. The decisions used to select traits form each branch is described below.

Branch 3 – Inflorescence fourth leaf blade length (iffourll) was chosen out of the other three inflorescence traits, flag leaf blade length (iffll), fourth leaf sheath length (iffourls) and flag leaf sheath length (iffls), because iffourll is a trait which is easier to measure, compared with flag leaf measurements, including iffls where it can be difficult to determine where the sheath starts if the adjoining node is not clearly defined. A minor problem with iffourll is being able to collect a tiller that still has its fourth leaf intact. However, if collections are made from newly established spaced plants and not swards that have been planted for some time, this should not pose a problem.

Branch 4 – Stolon leaf blade at node four length (stlnfl) was chosen out of stolon internode four sheath length (stifsl), inflorescence average length of spikes (ifalos), mown stolon internode four length (msifl), mown stolon internode four sheath length (msifsl), stolon internode four diameter (stifd), mown stolon internode four diameter (msifd), inflorescence fourth leaf blade width (iffourlw), inflorescence peduncle diameter (ifpd), inflorescence flag leaf blade width (ifflw), stolon leaf blade at node four width (stlnfw) and stolon branches at stolon node four (sbnfour), because the trait is consistently easy to measure and that leaf blade length is a standard measurement collected as part of the Australian PBR process for the registration of *Cynodon* spp. (Appendix III).

The relevance and value of the other four traits maximum diameter of spread (dsm), height of sward (hos), stolon internode four length (stifl) and inflorescence density (id) in distinguishing varieties of *C. dactylon* and *C. dactylon* x *C. transvaalensis* is described below:

Maximum diameter of spread (dsm) – Throughout the study, lateral spread has proven to be a significant trait in providing morphological-agronomic variation between species and varieties of bermudagrass. This is why it is important to undertake varietal testing trials in the field and not within pots. This will allow the turfgrasses to express their phenotype more fully via unrestricted growth;

Height of sward (hos) – Sward height is a clear visible trait that is easy to measure. Significant differences can be observed between species and varieties when measuring sward height, making this characteristic less ambiguous;

Stolon internode four length (stifl) – Internode length like that of leaf blade length is a standard attribute to be evaluated within the Australian PBR process (Appendix III for a list of traits to be measured), as requested by IP Australia. However, the PBRO does not stipulate what node to measure and is therefore left open to interpretation. Following the guidelines by produced by Roche et al. (2005), the fourth visible node was chosen because their research showed that cellular expansion in the stolon tip region was virtually complete, and on the need to avoid the increasing risk of damage to stolon and stolon leaves;

Inflorescence density (id) – Inflorescence density like sward height is a clear distinguishable characteristic. This trait not only provides a reliable characteristic to measure, flowering and quantity of flowering can easily differentiate turf species and varieties;

The trait groupings within the present study also showed that stolon branches at node four (stbnfour) was negatively correlated to inflorescence traits. It is possible that varieties with higher stolon branching (Mountain Green, TifEagle, Plateau and Windsor Green) are investing carbohydrates into branching compared with inflorescence attributes e.g. flag leaf blade length (iffll), fourth leaf blade length (iffourll), fourth leaf sheath length (iffourls) and peduncle diameter (ifpd).

Observation of the raw data collected showed there may also be a relationship between sward height (hos) and peduncle length (ifpl). The shorter the peduncle length of a variety the shorter the sward height of the turfgrass. This was particularly evident in variety groups 3 and 4. Such an observation can potentially be extremely useful as the sward height of a grass could be estimated from peduncle length, an easy trait to locate on the inflorescence preventing the need to wait for the turfgrass to develop physically to measure sward height (hos).

A series of boxplots (Figure 4.6a-f) have been constructed relating to the six beneficial traits (including maximum diameter of spread (dsm), height of sward (hos), inflorescence fourth leaf blade length (iffourll), stolon leaf blade at node four length (stlnfl) and internode four length (stifl) and inflorescence density (id)) chosen based on merit. The purpose of the boxplots is to complement the data contained in the variety grouping biplot (Figure 4.4).

(a)



(b)







(d)





(e)






Figure 4.6 Boxplots showing the variety group association of the informative traits (a) maximum diameter of spread, (b) height of sward, (c) inflorescence fourth leaf blade length, (d) stolon leaf blade at node four length and (e) internode four length and (f) inflorescence density.

Further Testing and DUS Studies

To validate the usefulness of the 6 important traits chosen, it is recommended that additional studies be undertaken long term using a larger range of genotypes in different seasons and environments where the likelihood of greater genotype x environment interaction may be evident. These studies should be conducted to assess the success of the 6 traits to see if they are capable of providing distinctness, uniformity and stability (DUS) across a range of species and varieties within C4 functional turfgrass species.

As there are currently limited guidelines in place by the PBRO for conducting a PBR trial (Appendix III). It is recommended that as a minimum, Qualified Persons undertaking warm-season PBR trials undertake the following:

- (i) Conduct a field trial, not a pot trial. This will allow for the plants to better express their phenotype and spread laterally across the soil surface; and
- (ii) Collect the minimum set of turfgrass descriptors (traits) for warm-season turfgrasses, which would include: maximum diameter of spread (dsm), height of sward (hos), stolon leaf blade at node four length (stlnfl), stolon internode four length (stifl), inflorescence fourth leaf blade length (iffourll) and inflorescence density (id).

Chapter 5: General Discussion

The aim of this research was to (i) characterise the morphological-agronomic variation among Australian *Cynodon* turfgrasses (ii) identify the most informative morphological-agronomic traits to describe varietal differences and (iii) determine the most appropriate methodology for quantifying these traits. These objectives have been largely achieved (Chapters 3 and 4), though more work could be done to make further improvements in the methodology used to document varietal differences. The discussion presented here will assess the outcomes of the present research in the context of future research directions and opportunities.

5.1 Morphological-Agronomic Variation

The morphological variability that exists within *Cynodon* spp. is well documented (e.g. Harlan et al., 1969, Harlan and de Wet, 1969a, Harlan et al., 1970). Likewise, the study presented here has suggested that there is large morphological variation present within and between *Cynodon* taxa grown commercially in Australia. The large overlap between the taxa observed is not entirely clear but may relate to the presence of a range of dwarfing genes that exist within each taxa.

In recent studies of Australian bermudagrasses (e.g. Jewell et al., 2012) using expressed sequence tag (EST)-simple sequence repeat (SSR) markers, two *Cynodon* accessions thought to be *Cynodon* hybrids were in fact *Cynodon dactylon* (C Lambrides 2013, pers. comm., 2 March). The varieties had a similar appearance to a dwarf or ultradwarf greens grass variety, however, genetically they did not group with other (known) *C. dactylon* x *C. transvaalensis* varieties.

With the exception of ultradwarf hybrid bermudagrass varieties and planned manipulation of *Cynodon* spp. genes through intraspecific hybridization to reduce internode length and leaf length/width (Duncan, 2004), no studies, with the exception of Harlan and de Wet (1969b) who eluded to very small and fine turfgrasses being present, have specifically reported on natural dwarfing genes in *Cynodon dactylon*. It's possible that natural dwarfing genes are present within *Cynodon* spp., but particularly varieties of *C. dactylon* which adds to the confusion when breeders have previously attempted to identify the species taxonomically. However, given the ongoing improvements in DNA technology and having numerous international bermudagrass collections available, it may prove beneficial to once again look deeper into identifying the species taxonomically and account for the presence of dwarfing genes.

5.1.1 Stolon and Inflorescence Interaction

A strong correlation among stolon and among shoot characteristics and a stronger correlation among inflorescence characteristics, with the exception of inflorescence density (id) was observed (Figure 4.3) in the present study. The vectors pointing in the same direction (i.e. with a narrow angle between them) correspond to variables that have similar profiles and therefore could be interpreted as having similar meaning in the context set by the data. An observation like this highlights the requirement to select and measure informative traits that will provide varietal differentiation. To achieve this, traits should be chosen from different morphological features (e.g. growth habit, stolon and shoot and inflorescence). Consequently, 6 informative traits were chosen within this study.

Based on the principal component relationship of Figure 4.3, stolon branching at node four (stbnfour) was a trait which clearly separated from all others and was negatively correlated to almost all the inflorescence traits. It is possible that bermudagrass and hybrid bermudagrass varieties with higher stolon branching are investing carbohydrates into branching compared with inflorescence attributes. If this is true, further maintenance practices like mowing to remove or reduce flowering may also increase branching and sward development.

5.1.2 Varietal Grouping

C. dactylon varieties and *C. dactylon* x *C. transvaalensis* hybrids were both present within each of the 4 cluster groups identified through pattern analysis. Variety group 4, the largest of the groups, contained both first- and second-generation greens-quality grasses and medium- to coarse-textured hybrids, along with *C. dactylon* varieties.

The overlap of these different taxa in each of the 4 clusters highlights the enormous diversity within each of the classes of *Cynodon* turfgrasses; including varieties selected as functional turfgrasses. Although, when grown as spaced plants in the field, clear differences could be observed that differentiated between species and most of the 25 varieties studied. This emphasises the point that clustering uses information from all the collected data, not just individual traits. For example, within variety group 4, Novotek and OZ TUFF are two very different plants. Novotek is a triploid (2n = 27) and OZ TUFF is a tetraploid (2n = 36).

Four cluster groups were identified of the 25 varieties studied. It is unknown if the number of cluster groups would have remained the same if a greater number of varieties were trialled? Clearly, further studies and analysis of already collected morphological data are needed.

When applying for PBR in Australia, prior to commencing varietal testing it is essential that comparator or known varieties be chosen to trail against the new or candidate variety undergoing evaluation. The decision on which varieties to choose if not directly related to the candidate variety (e.g. parent/source material) is difficult and often based on subjective judgement and experience of the person conducting the varietal testing. However, in future it may be possible to better identify varieties of common knowledge through cluster groups if a parent or similar variety can be identified.

5.1.3 Informative Traits

Following pattern and data analysis, 6 trait groups were identified out of a possible 20 branches (Figure 4.4). This analysis was used to identify a subset of traits that potentially could be used to preliminarily differentiate other groups (species and or varieties) of bermudagrasses. Within 6 trait groups identified, a single trait was selected based on merit.

Being able to ascertain informative morphological-agronomic traits will not only save resources, but it will provide the breeder or agronomist with distinct, uniform and stable data to differentiate between species and or varieties of bermudagrass. It is possible that the set of informative traits could also be used to preliminarily differentiate between other varieties of *Cynodon* and or other species of warm-season turfgrass. However, DNA fingerprinting should also be undertaken to provide additional information to morphological observations.

5.2 Use of Biplots and Dendrograms

The biplot was a simple method to graphically represent thousands of data points and display relationships between both the variables and cases (in this instance, turf varieties) being analysed, while a dendrogram was used to represent graphically the hierarchical cluster analysis of the data collected.

Replicated spaced plant and sward trials containing multiple varieties provide an opportunity to generate morphological-agronomic data across a diverse range of genotypes. The data collected for a range of traits can then be analysed for each experiment individually (as, for example, for PBR and plant patent applications) or analysed across a group of experiments to distinguish clusters as was done in the present study. The main issue with grouping is then how best display the data for interpretation.

In the present study, pooling the data across all experiments and years and displaying the results in the form of dendrograms and biplots contributed to clearer, more informative presentation of the data than by using separate analyses for each experiment conducted between 2002 and 2004. The latter option would have been potentially confusing and difficult to understand because of the number of varieties trialled, the number of experiments undertaken, the large number of traits measured and the large volume of data generated.

5.3 Genotype X Environment Variation

Active turfgrass growth and development, particularly lateral spread, are highly dependent on soil and air temperature such that varietal responses to seasonal temperatures influence how and when particular genotypes will perform. The present study conducted 6 experiments over varying seasons between 2002 and 2004. It may be suggested that seasonal variation could have influenced the findings. However, a total of 14,248 morphological-agronomic data points were collected (excluding missing values) encompassing 25 *Cynodon* varieties and 21 traits, thereby generating a comprehensive data set available for statistical analysis.

5.4 Varietal Testing

Necessary Measurements

Lateral spread has traditionally been a useful trait for determining active plant growth and performance and therefore distinguishing varietal differences. Experimentally, four measurements per plant were taken at regular intervals with the premise that the mean value derived from these would give a more accurate representation over time of lateral spread for the overall plant. However, the present study has shown for the purpose of differentiating between varieties of *Cynodon*, that there is no advantage in collecting four measurements per plant, or collecting data over multiple dates.

Two linear regressions were analysed using (i) cumulative mean data and cumulative maximum data encompassing 8,946 data sets (missing data excluded i.e. dead or contaminated plants) (Figure 5.1) and (ii) average and maximum lateral spread measurements collected on the last day of data collection (i.e. excluding earlier measurements) encompassing 1,675 data sets out of a possible 1,710 data sets (excluding missing data) (Figure 5.2). Interestingly, both linear regressions, cumulative data collected over the duration of the study provided very strong positive correlations

 $(r^2 = 0.96)$, meaning that only 0.04 % of the variance was not shared between either the maximum and average diameter of spread means (Figures 5.1 and 5.2).

Being able to take a single measurement (not cumulative data) of the maximum diameter of spread (dsm) across the centre of the plant before encroachment occurs, will save the breeder/agronomist/Qualified Person (for the purposes of conducting a PBR trial) significant resources previously devoted to making lateral spread measurements.



Figure 5.1 Linear regression of cumulative (all) average and maximum lateral spread measurements of *Cynodon* taxa varieties acquired throughout spaced plant experiments 1 to 5.



Figure 5.2 Linear regression of average and maximum lateral spread measurements of *Cynodon* taxa varieties at the *last day of data collection* (i.e. excluding earlier measurements) in spaced plant experiments 1 to 5.

It is recommended that height of sward (hos), inflorescence fourth leaf blade length (iffourll), stolon leaf blade at node four length (stlnfl) and internode four length (stifl) and inflorescence density (id) also be collected when assessing varieties of *Cynodon dactylon* or *C. dactylon* x *C. transvaalensis*. It is also probable that these traits may also be beneficial in distinguishing varietal differences in other warm-season turfgrasses, but further research is warranted.

Unnecessary Measurements

With the exception of published plant patent applications in the US, PBR applications in Australia, and a limited number of scientific studies (e.g. Hurcombe, 1947, Harlan et al., 1969, de Wet and Harlan, 1970, Liu and Guo, 2003, Roche and Loch, 2005, Wu et al., 2007, Kan et al., 2008, Nasiri et al., 2012), there is limited quantitative data describing morphological-agronomic variation in *Cynodon* turfgrasses. However, the aim of conducing a PBR experiment or morphological comparative study is not to collect data for the sake of collecting data but rather to focus on traits that repeatedly allow the separation of varieties.

Within the present study, several traits showed minimal variation in terms of being able to differentiate between varieties of *Cynodon* and in the case of inflorescence peduncle length (ifpl), the trait showed no variation at all. Other traits of limited value (from Figure 4.4) included: stolon characteristics – mown internode four length (msifl), diameter (msifd) and sheath length (msifsl), branches at node four (stbnfour); internode four diameter (stifd) and leaf blade at node four width (stlnfw); and inflorescence characteristics – peduncle length (ifpd) average length of spikes (ifalos), flag leaf blade width (ifflw) and fourth leaf blade width (iffourlw).

Location and Conduct of Trial

Varietal testing studies are on the most part either conducted in (squat) pots or in the field. Both environments will provide breeders or agronomists with growth data. However it's possible that the pot trials will not allow candidate (new) or comparator varieties to express their phenotypes sufficiently to draw differences. Also, within the present study, maximum diameter of spread (dsm) has shown to be an informative characteristic in providing distinct, uniform and stable data. If turfgrass plants are to be planted in pots, the plants may not provide the examiner with accurate or uniform information e.g. stolon nodes are unable to produce roots and supplementary branching.

5.5 Practical Implications of This Study

The present study developed a structural framework in which genotypes with similar morphological/agronomic characteristics were grouped together. Using these techniques could potentially simplify issues for turf breeders and agronomists by enabling multiple varieties or even multiple experiments containing similar varieties to be considered at a group level rather than on a variety-by-variety basis. Breeders, for example, can initially select promising genotypes based on grouping characteristics that balance producer preferences for faster grow-in and shorter production cycles (through more aggressive lateral growth) with high turf quality to meet consumer needs for reduced thatch development and mowing inputs through stolons with shorter internodes and increased branching. For agronomists, varieties with similar morphological-agronomic characteristics may also have similar management requirements.

For descriptive purposes under Plant Breeder's Rights or similar schemes, the groupings developed for current varieties in this study will assist in the identification of the closest varieties of common knowledge to include in a comparative growing trial with a new candidate variety. Informative traits identified through the present study, including maximum diameter of spread (dsm), height of sward (hos), inflorescence fourth leaf blade length (iffourll), stolon leaf blade at node four length (stlnfl) and internode four length (stifl) and inflorescence density (id), will enable more time- and resource-efficient comparisons of varieties to be made through the growing trial rather than duplicating efforts through measurements of closely-related or less informative traits. A list of recommended morphological traits to be measured of *Cynodon* spp. varieties under the Australian PBR scheme can be seen in Appendix III.

5.6 Future Work

Varietal descriptions of warm-season turfgrasses for the PBR Office within IP Australia cover a range of genera and species including *Cynodon* spp., *Paspalum vaginatum* (seashore paspalum), *Pennisetum clandestinum* (kikuyugrass), *Sporobolus virginicus* (marine couch), *Stenotaphrum secundatum* (buffalograss/St Augustinegrass), *Zoysia matrella* (Manilagrass) and *Z. japonica* (Japanese lawngrass). Many previous studies have also involved similar methodology to that followed with *Cynodon* spp. as described in Chapter 3. It is of future interest to determine if the same informative traits identified through the present study are also of value for grouping varieties within other C4 turfgrass species.

Because each of the six experiments was conducted at only one geographical location, environmental and edaphic parameters such as soil type and seasonal climatic conditions potentially limited the phenotypic expression of some or all of the *Cynodon* varieties evaluated at that single site. Future studies are warranted to determine how well the findings from the present study hold up across a wider range of environmental conditions, seasons and cultural management factors. Genotype X nutrient availability within a growing site may also affect phenotypic expression and should also be considered in future studies. Nutrient availability is another limitation on reliance exclusively on morphologic separation. This and additional factors are why a combination of measures of morphological traits and DNA fingerprinting are important to characterising the differences amongst commonly utilised varieties.

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Appendix I: Background to the Approved Beneficial Vegetative Cynodon taxa varieties within Australia

Introduction

Appendix I contains a description of the current (1950s to 2012) *Cynodon* taxa found in Australia. Detailed information has been sourced in relation to the origin and development of the grasses which are suitable for turfgrass use. At no stage within the Australian turfgrass history have such particulars been collated and made available in a single publication. Such detail provides an interesting picture of the source of proliferation of newer varieties and how the Australian industry has evolved with the introduction of overseas and Australian selected varieties. The information adds to the bigger picture of that contained in the preceding thesis, the morphological and agronomic attributes and how closely each selection or variety is related.

The varieties discussed in this paper (listed alphabetically) are derived from one of the four classifications, being (i) *Cynodon* sp. (although the variety contained within the taxa fits best being classified as a *Cynodon* hybrid), (ii) *Cynodon dactylon* x *C. transvaalensis* (hybrid bermudagrass), (iii) *Cynodon dactylon* (bermudagrass) and (iv) *Cynodon dactylon* x *C. magenissii*. The latter species is debatable and may be best suited to fit within *Cynodon dactylon* or *Cynodon dactylon* x *C. transvaalensis*. However, given the information at hand, the varieties identified as *Cynodon* sp. or *Cynodon dactylon* x *C. magenissii* by the breeder/author they will remain as is.

At the time of publication, the turf research activities and entire Lifestyle Horticulture program (turf, flowers and nursery) within the Queensland Government operating under the Department of Agriculture Fisheries and Forestry (DAFF) had been terminated. Within the current Annex, numerous varieties are listed as being housed at Redlands Research Facility, Cleveland, QLD, Australia. The author can't guarantee that the warm-season reference collection or foundation plots which were located at Redlands for 13 years will remain. As such the future of the entire C4 collection containing over 170 varieties of commercial and experimental lines from Australia and overseas is under threat.

AGRD

'AGRD' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was a result of a spontaneous mutation that was selected by Dr Warren Hunt, from a variant area of winter active turf (probably 'Tifway' or 'Tifgreen') on a Hong Kong Golf Course in April 1996 (Roche and Loch, 2008a). A selection of this material was imported through vegetative quarantine via New Zealand for evaluation. Following a favourable assessment of its potential as a warm-season turfgrass variety under New Zealand conditions based on its superior comparative performance relative to other *Cynodon* accessions in glasshouse and field trials, the New Zealand registered (Plant Variety Rights grant number 1566, which is to expire on 14 July 2019) variety 'Grasslands AgRiDark' was released in New Zealand in 1999 (Roche and Loch, 2008a).

Material of 'AgRiDark' was sent from Grass Technology Ltd, New Zealand to the Australian Quarantine Inspection Service (AQIS) Post-Entry Quarantine facilities at Eastern Creek, New South Wales on 16 December 2004 to undergo testing. Following testing and clearance, vegetative material was released to DAFF Redlands Research Facility, Queensland for the purposes of conducting Plant Breeder's Rights testing. AGRD was granted PBR in Australia on 20 January 2009 (Roche and Loch, 2008a). Initially 500m² of AGRD was vegetatively (asexually) propagated and grown on as foundation stock at RRF beginning on 1 August 2008. It was of the opinion that DAFF would at the time multiply sufficient vegetative material to supply a licensee in each desired state and/or territory within Australia upon the request of the (now) owners Cervadon Ltd, New Zealand. First sale of AGRD in Australia was of 100m² to City of Casey, Melbourne on 27 January 2010.

BoskerTM (C3)

The accession number S-282 (M Robinson 2008, pers. comm., 16 September), later designated 'C3' [*Cynodon dactylon* (L.) Pers.] was selected from a series of trials initiated by The Victorian Department of Agriculture (Daratech) and the Melbourne Cricket Club (P Semos 2006, pers. comm., 18 August). The trials setup as the National Bentgrass and Couchgrass Evaluation Trials involved a selection and evaluation process which was carried out between 1986 and 1991 and was one of the first [earlier studies were undertaken at ATRI (P McMaugh 2009, pers. comm., 11 November)] of its kind to be formally undertaken within Australia (Robinson and Neylan, 1997). At the time, this was made possible with Turfgrass Technology (a division of Daratech) receiving funding from a succession of financial contributors including the Australian Golf Union, Victorian Golf Association, Victorian Golf Course Superintendents Association, the Melbourne Cricket Club

and the Horticultural Research and Development Corporation (Robinson and Neylan, 1993). A total of six couchgrass trial sites were setup across Australia with the majority of the sites being established between September and November 1991, but a later site was also established in Victoria during October 1994 (Robinson and Neylan, 1997). The six trial sites were located in Perth (Collier Park Golf Course, Como, Western Australia), Melbourne (Turfgrass Technology's Research Station at Frankston, but later transferred to the Peninsula Country Club, Frankston, Victoria in November 1992 due to the uncertainty of the future of the research station and a second site at the Greenacres Golf Club which was the last site to be established in 1994), Sydney (The Australian Golf Club, Kensington, New South Wales), Adelaide (Riverside Golf Club, West Lakes, SA) and the Gold Coast (The Gold Coast Burleigh Golf Club, Miami, Queensland), providing different environmental and climatic conditions in a variety of regions across Australia (Robinson and Neylan, 1993). The trial evaluated some 400 couchgrass accessions (P Semos 2006, pers. comm., 18 August). The tetraploid (2n = 36) variety C3 was one of the 400 vegetative accessions which had initially been collected from the Ladies 4th Tee of the Wonthaggi Golf Club, Victoria in November 1987 by John Nevlan (Ho, 1999; M Robinson 2008, pers. comm., 16 September) who was at the time, an agronomist with the Turf Research and Advisory Institute, Victorian Department of Agriculture.

C3 was selected from the trial and later trademarked as Bosker[™] (P Semos 2006, pers. comm., 18 August) by the Australian turf company StrathAyr Turf Systems Pty Ltd. Vegetative material of C3 from the National Couch-Grass Trial was later provided to StrathAyr for subsequent multiplication, yet the variety was never released for commercial production by StrathAyr. Lowlands Lawn Turf (which is now combined with Qualturf Pty Ltd and Saliba Turf Supplies Pty Ltd), Windsor, New South Wales has sold material of Bosker[™] some years ago, and M. Collins and Sons Pty Ltd, Revesby, New South Wales have a licence to sell Bosker[™] but have not cultivated any material as yet (G Beehag 2009, pers. comm., 6 October).

Champion Dwarf

'Champion Dwarf' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was selected in 1987 from a 'Tifdwarf' *Cynodon* hybrid golf green that had been planted in 1969 (Kaapro, 199a) by Richard Morris Brown in Walker Country, Texas (Miller and Edenfield, 2002). Work to develop the triploid (2n = 27) selection and conduct independent research of the turf performance was undertaken by Coastal Turf, Inc. of Bay City, Texas, USA (Brown et al., 1997; Kaapro, 199a).

The selection from the Tifdwarf patch was based on the advantageous characteristics of vertical leaf extension rate, lateral stem development, turf recuperative rate, shoot density, leaf blade width and

terminal height (Kaapro, 199a). Champion Dwarf was first sold in the United States in March 1996 (Brown et al., 1997).

Brown et al. (1997) referred to one of the more unique traits of Champion Dwarf as one that does not routinely form a seedhead and that no inflorescences of any kind had been observed in comparative trials conducted in Bay City, Texas, USA. This included testing Champion Dwarf with three other varieties ('Tifway', 'Tifgreen', and Tifdwarf) grown in test plots, glass-house and in large-acre production fields with varying management practices, over a minimum eight year period (Brown et al., 1997).

In addition to the side-by-side comparisons in Bay City, the four varieties have been grown in US locations with dissimilar climates, such as Palm Desert, California, and Auburn, Atlanta; yet no inflorescence development had been observed in the Champion Dwarf plots in these locations either (Brown et al., 1997).

However, in field trials conducted by the DAFF Turf Research team, Redlands Research Facility, Queensland, Australia seed heads were observed and recorded in two trials (spaced plant and sward). Both experiments were setup to obtain morphological-agronomic characteristics as listed by Roche (2009) for the purposes of a Plant Breeders Right's comparative testing. The first, a sward trial planted on 7 June 2002 tested the greens quality grasses Champion Dwarf, 'FloraDwarf', 'MS-Supreme', 'TifEagle', 'Novotek', Tifgreen and Tifdwarf. DAFF researches recorded Champion Dwarf as having produced inflorescences with a mean value of 0.67 per plot [1 plot measuring 1 x 0.9 m by 3 replicates (=3 plots)] with a minimum value of 0 and maximum value of 2 inflorescence counts at 343 to 344 DPP when using a 0.1225m² quadrat. In the second experiment, a spaced plant trial planted on 13 February 2006 tested Champion Dwarf, MiniVerdeTM, MS-Supreme and TifEagle. Champion Dwarf at 192 and 195 DPP was recorded as having produced inflorescences with a mean value of 11.73 per plant [5 plants measuring up to 1 x 1 m by 6 replicates (=30 plants)] with a minimum value of 61 inflorescence present within a 0.1225m² quadrat. Champion Dwarf was granted PBR in Australia 22 May 2002 (Kaapro, 1999a).

Material of Champion Dwarf which was sent by Coastal Turf, Texas to Australia, arrived and was planted in the turf demonstration plots at DAFF Redlands on 9 November 2001. The latter 7m² observational plot over the years has been vegetatively (asexually) multiplied to obtain 70m² of foundation turf which is also situated at Redlands and has over the years supplied planting material for numerous regional trial sites across Australia. One such trial was the Horticulture Australia (HAL) funded study 'Management of new warm-season greens grasses in Australia' (project code TU05001). All seven trial sites tested Champion Dwarf for the duration on the trial which ran

between 2006 and 2010. They included, Glenelg Golf Club (SA), Sanctuary Cove Golf Club (Qld), Horton Park Golf Club (Qld), Twin Water Golf Club (Qld) and the central test facility at DAFF Redlands Research Facility (Qld). In late 2012, Brett Morris, Superintendent at Brisbane Golf Club was in the process of converting their putting greens to this variety with the support of the members and the committee. Vegetative material was sourced from RRF.

ConquestTM (Riley's Evergreen)

ConquestTM was discovered by the late Rod J. Riley in 1991 growing in a *Cynodon dactylon* 'Wintergreen' bowling green at Homebush Bowling Club, Homebush, New South Wales, Australia (Kaapro, 1999). ConquestTM [*Cynodon dactylon* (L.) Pers.] which also goes by the name of 'Riley's Evergreen' was granted Plant Breeder's Rights on 13 June 2000 (Commonwealth of Australia, 2005d) and was released soon after. ConquestTM was selected as it displayed good low temperature leaf colour retention and a broad leaf width in comparison to the parent plant. ConquestTM also has reasonable wear recovery, low thatch and actively grows throughout the year showing good colour retention.

CT-2

^cCT-2' [*Cynodon dactylon* (L.) Pers.] was discovered by breeder Hughbert F. Whiting through a series of cross-pollination of selected varieties of *Cynodon dactylon* grass plants (Whiting, 1989) at Fallbrook, California, USA (Whiting, 1988). The parental grass plant being commonly known as 'Wintergreen' was the male grass plant and the female grass plant was commonly known as 'Greenlees Park' (Whiting, 1988). Following crossing, the desired plant was then selected and asexually repropagated as the new and distinct variety of *Cynodon dactylon* grass plant CT-2 (Whiting, 1989). CT-2 is trade marked by the Greg Norman Turf Company (GNTC) as GN-1TM in the USA. GNTC identifies the variety on their web site (www.sharl.com) as a medium textured hybrid bermudagrass; however the breeder identifies the variety as a straight *Cynodon dactylon* variety.

Whiting (1989) described the *Cynodon dactylon* variety Wintergreen (C84-135) as the closest known variety to his variety CT-2, however the breeder of the former variety, Peter McMaugh explained that this was "incorrect" (P McMaugh 2009, pers. comm., 11 November). The results contained in this thesis support McMaugh's assertion. CT-2 was selected for improved colour,

winter colour retention and less thatch build-up (Whiting, 1988). The grass plant CT-2 is entirely public p

CT-2 was initially made available through Tyagarah Turf, Byron Bay, New South Wales in 1991 and was "widely" used on sports fields, golf courses and school playgrounds (Anonymous, 1995). PBR protection of the variety in Australia has since expired and a renewal was not applied for by the breeder Hugh Whiting following the 20 year term post the original filing date of 6 May 1988.

CynoMax^{тм} (LEG13A)

'LEG13A' [*Cynodon dactylon* (L.) Pers.] was a result of open pollination followed by seedling selection carried out by Todd Layt, Clarendon, New South Wales in 2008 (Pannanen, 2008b). The seed parent 'C1' (Legend®) is characterised by a large number of inflorescences produced on each plant. LEG13A is trademarked as CynoMaxTM and is characterised as having fast 'speed' of growth, strong sod strength, low seed head production and dark leaf colour (Pannanen, 2008b). LEG13A was granted Plant Breeder's Rights on 18 March 2009 (Pannanen, 2008b).

From initial observations made by Todd Layt, CynoMax[™] produces less seedheads than other common bermudagrasses, is fast growing and produces minimal thatch which is preferred for oversowing (Layt, 2009). CynoMax[™] was realised by Ozbreed in 2011.

CynoSportTM (WGP3)

'WGP3' [*Cynodon dactylon* (L.) Pers.] which is marked as CynoSport[™] was a result of open pollination followed by seedling selection carried out by Todd Layt, Clarendon, New South Wales, Australia in 2008. The seed parent was identified by Todd Layt as being *Cynodon dactylon* (varieties present included 'Wintergreen', 'Greenlees Park' and 'C1'). The seed parent is characterised by a large number of inflorescences produced on each plant. WGP3 is characterised as having fast 'speed' of growth, strong sod strength, low seed head production and dark leaf colour (Pannanen, 2008a). The characteristics identified by Pannanen (Pannanen, 2008a) are identical to that of 'LEG13A'. CynoSport[™] was released by Ozbreed in 2011. WGP3 was granted Plant Breeder's Rights on 18 March 2009 (Pannanen, 2008a).

FloraDwarfTM

The variety FloraDwarf[™] was assigned by Dudeck & Murdoch (1998) as being a *Cynodon* sp., but following morphological and agronomic studies by Roche & Loch (2005) the fine textured turfgrass would be better identified as a *Cynodon* hybrid [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy]. FloraDwarf[™] was one of 224 selections of couchgrass collected throughout the state of Hawaii, USA during 1988 (Dudeck & Murdoch, 1999). The accession was collected from the practice green on the Wailua Municipal Golf Course located on the island of Kauai, Lihue, Hawaii by Dr Albert E. Dudeck on 28 June 1988 (Dudeck & Murdoch 1998; Dudeck & Murdoch 1999; Miller and Edenfield, 2002). The 'Tifgreen' (Tifton 328) practice green from where the material was selected was thought to have been planted in 1977 (Dudeck & Murdoch 1999).

From 1989 to 1999, FloraDwarf[™] was tested as Florida Hawaii Bermudagrass accession 135 (FHB-135) (Dudeck & Murdoch 1999) and was characterised as a sterile, fine textured, inconspicuous flowering, low growing stoloniferous grass that does not produce rhizomes. FloraDwarf[™] was released by the Florida Agricultural Research Station on 24 January 1995 (Brown et al., 1997; Dudeck & Murdoch 1998; Dudeck & Murdoch 1999) and is suitable for use on golf course putting greens and lawn bowling greens.

Foundation stock of FloraDwarf[™] is grown under certification standards governed by the Southern Seed Certification Association, Auburn, Alabma, USA with stock obtainable upon request from Florida Foundation Seed Producers, Inc., Greenwood, Florida, USA (Dudeck et al., 1994).

Material of FloraDwarf[™] sent from the University of Florida arrived in Australia and was planted at DAFF Redlands Research Facility, Cleveland, Queensland in the facility's turf demonstration plots on 18 October 2001. The latter 7m² observational plot over the years has been vegetatively (asexually) multiplied to obtain 70m² of foundation turf which is also situated at Redlands and has over the years supplied planting material for numerous regional trial sites across Australia. Once such trial was the Horticulture Australia (HAL) funded study 'Management of new warm-season greens grasses in Australia' (project code TU05001). All seven trial sites tested FloraDwarf[™] for the duration on the trial which ran between 2006 and 2010. They included, Glenelg Golf Club (SA), Sanctuary Cove Golf Club (Qld), Horton Park Golf Club (Qld), Twin Water Golf Club (Qld) and the central test facility at DAFF Redlands Research Facility (Qld).

FLoraTeX®

FLoraTeX® [*Cynodon dactylon* (L.) Pers.] is thought to have been introduced into United States under the name 'Franklin' on 18 February 1954 by African Explosives and Chemical Industries Ltd, Johannesburg, Transvaal, South Africa (Juska and Hanson, 1964; Dudeck et al., 1994). The selection was assigned the plant introduction (PI) number 213385, by the USDA New Crops Research Branch, Crops Research Division. 'Franklin' was originally collected from a putting green that was severely damaged by mealybugs, *Antonina indica* Green, at Mount Edgecomb Golf Course, Natal, South Africa (Dudeck et al., 1994).

Between 1955 and 1962 experimental work was undertaken on the PI 213385 accession in Alabama, Arizona, California and Georgia. During this period breeder Arden A. Baltensperger noted that he had received contaminated plant material from the Southern Regional Plant Introduction Centre, New Orleans, Louisiana. Baltensperger continued to test three vegetative off-types from PI 213385, including the accession 119 (FB-119) which was later designated as FLoraTeX® (Dudeck et al., 1994).

FLoraTeX® was jointly released by the Florida Agricultural Experiment Station in 1992 and the Texas Agricultural Experiment Station (TAES) in 1993 (Dudeck et al., 1994; Juska and Hanson, 1964; Dudeck et al., 1995; Polomski and Shaughnessy, 2003) [Busey (Busey and Dudeck, 1999) says it was released in 1994]. FLoraTeX® is a prolific seed head producer that may produce viable seeds (Dudeck et al., 1994; Dudeck et al., 1995), deep rooted, excellent colour retention in the autumn and early spring 'greenup' with superior dehydration avoidance. The true origin of FLoraTeX®, however, was lost over time due to vegetative contamination and misplacement of original stock material of PI 213385 from the Southern Regional Plant Introduction Station, USDA-ARS-SAA, Griffin, USA (Dudeck et al., 1994).

Vegetative material of FLoraTeX® was brought into Australia from the United States and planted at DAFF Redlands Research Facility, Queensland in their turf demonstration plots on 9 November 2001. Rochedale Turf Pty Ltd was the sole Australian licensee of the turfgrass.

Gullygold

'Gullygold' was discovered in February 2006 by Thomas G. Parker, Sydney Cricket Ground Curator, as a chance seedling or mutant plant growing among "Common" bermudagrass (*Cynodon dactylon*) in a cricket wicket at Wisemans Ferry, NSW (Roche, 2010). A selected piece of sod was removed from the wicket block and grown in a pot to undertake initial observations. In Feb 2007 a

sample of this material was taken and provided to Dad and Dave's Turf farm, Pitt Town, NSW to grow-on, multiply and take further observations. By 2009, approximately 300m² of turf had been produced as nursery stock at the NSW farm. Observations undertaken from Thomas Parker and Graeme Colless of Dad and Dave's Turf reported initially that the plant was very quick to run across the surface, recoverd quickly after scalping and produced a dark green colour with (little to) no fertiliser input (Roche, 2010). Gullygold was granted PBR on 29 March 2011 following testing at Redlands Research Facility (Commonwealth of Australia, 2005j).

Grand Prix

'Grand Prix' [Cynodon dactylon (L.) Pers.] is a variety that was produced by control pollination followed by selection of 'Wintergreen' and 'Couch 5' (also designated 'C5') by breeder David Nickson. Couch 5 [accession number S-130 (Robinson and Neylan, 1993)] an experimental breeding line that was selected from seed provided to the breeder by Arden Battensperger (McMaugh, 2008) was a selection from an earlier series of crosses by the breeder between 'Wintergreen' and a number of Cynodon dactylon accessions, which were collected from the Peninsula Country Club Fairway 8B, Frankston, Victoria by Peter McMaugh in February 1987 (Neylan, 2005; M Robinson 2008, pers. comm., 16 September). A series of other Cynodon collections were carried out between 1986 and 1990 from the Peninsula area of Victoria, Australia. Random open pollination (P McMaugh 2009, pers. comm., 11 November) was carried out by the breeder in 1998 and subsequent seedlings and selected plants were observed between 1998 and 2000. In the spring of 2000, the remaining potted seedlings were planted into plots at the Evergreen Turf farm at Pakenham, Victoria, Australia and allowed to expand fully (Roche and Loch, 2006a). The final selection of Seedling 12 (later designated DN12) in late 2002 was based on shoot density, leaf colour, turf quality and reduced thatch accumulation (Roche and Loch, 2006a). Grand Prix was granted Planted Breeder's Rights on 12 September 2006 (Commonwealth of Australia, 2005e).

Grand Prix also showed superior signs of wear tolerance and recovery in a *Cynodon* spp. study conducted at DAFF Redlands Research Facility, Queensland between 2005 and 2007. Wear tolerance was associated with high shoot density, a dense stolon mat strongly rooted to the ground surface, high cell wall strength as indicated by high total cell wall content, and high levels of lignin and neutral detergent fibre (Roche et al., 2009). As a result of the study the variety was chosen to be planted as the playing surface at Skilled Park, Robina, Queensland which opened in February 2008.

A secondary wear study was conducted by DAFF as part of a Horticulture Australia (HAL) funded trial (project code TU08018) to investigate wear tolerance and recovery of a selection of C4 warm-

season turfgrass varieties. Throughout the duration of the study, 2008-2012, Grand Prix showed significant wear tolerance in the simulated and actual wear (in play on touch football fields) studies conducted at DAFF Redlands Research Facility and Redlands Touch Football fields respectively.

The first sale of the variety Grand Prix anywhere in the world was on 21 October 2006 by Evergreen Turf Pty Ltd to a homeowner on Dandenong Road, Frankston East, Victoria (Nickson, 2007).

Greenlees Park (Greenleaf ParkTM)

'Greenlees Park' [Cynodon dactylon (L.) Pers.] was selected from Greenlees Park Bowling Club (Ho, 1999) at Concord in New South Wales by the late Rod J. Riley in January of 1965 following the Australian Bowls Championship (McMaugh, 2008). Greenlees Park has been referred to as the first single-strain couch grass in Australia (McMaugh, 1988); following the initial planting of the accession at the latter Greenlees Park Bowling Club in late 1969 (Beehag, 2006) after subsequent propagation and growing-on of the grass. Multiplication and commercialisation of Greenlees Park was undertaken by Beverina Estate, Cobbitty and Qualturf, Windsor which were both located in New South Wales (McMaugh, 2002). Beverina Estate trademarked the variety as Greenleaf Park[™] (McMaugh, 1988). The selection and vegetative material from Greenlees Park Bowling Club was used as source for numerous bowling greens and golf courses throughout Sydney, other parts of Australia and even Indonesia from the early 1970s (McMaugh, 1988). Floreat Park Bowling Club, Floreat had the first Greenlees Park green in Western Australia, however this was acquired by accident; the latter club had arranged to obtain the C. dactylon variety 'National Park' from a club on the east coast, however when the time came to have it sent over the club concerned said they did not have material of 'National Park' and recommended that another club had a plentiful supply of Greenlees Park (Vernon n.d.). As a result, the Floreat Park Bowling Club established their green with Greenlees Park. Following the success of the variety for lawn bowling use in New South Wales, 'Greenlees Park' moved into commercial turf production with the first supply being developed by George Dukats at Theresa Park and Peter McMaugh at Qualturf in Windsor (McMaugh, 2008). Today, the purity and origin of guaranteed Greenlees Park is debateable. It is believed that a true to type supply of Greenlees Park is still present at Greenless Park Bowling Club, but clean material is also being grown on sportsfields in and around Cairns, Qeensland (i.e. Cairns Saints Australian Football Club Inc. and Gladstone Race Course, which are both located in Queensland).

Hardy TurfTM (JT1)

'JT1' [*Cynodon dactylon* (L.) Pers.] was a result of a spontaneous mutation or chance seedling discovered in the mid-1990s by Lynn Davidson as a superior plant growing in a commercial field of "Common" *Cynodon dactylon* on Jimboomba Turf Company's farm at Jimboomba, Queensland, Australia (Loch and Roche, 2003c). The selection by now Owner/Manager of Jimboomba Turf Company, Lynn Davidson, was taken from the "Common" couch field in 1999 after observing a superior turf quality, vigorous lateral spread, high shoot density, darker green colour and low presence of inflorescences of this mutant plant (Loch and Roche, 2003c). The variety was released on 13 September 2002 (L Davidson, pers. comm., 29 January 2010) and is now sold and trade marked as Hardy Turf[™] for commercial and home plantings through Jimboomba Turf Group Pty Ltd. JT1 was granted Plant Breeder's Rights on 24 February 2005 following testing at Redlands Research Facility, Cleveland, Qld, Australia (Commonwealth of Australia, 2005f).

Hatfield

A *Cynodon* selection was made by Graham Hatfield from a population growing in soil excavated from a building footing in 1983 at 43 Sheilds Street, Gympie, Queensland, Australia (Loch and Roche, 2003e). The breeding process involved a single cycle of selection to separate out material of the selected plant for vegetative propagation (Loch and Roche, 2003e). The selected plant was given the experimental name ES302 and later designated as 'Hatfield' [*Cynodon dactylon* (L.) Pers.] producing a dense, mat-forming, dark-green turf. Paul Nunn of Turfworld, Kilcoy, Queensland (at the time of this publication) the sole grower of Hatfield first sold sod of the variety on 6 September 2007 as 'cover grass' (because the sod was contaminated with another *Cynodon* spp. variety) to Tinamba Turf (W Scattini 2008, pers. comm., 31 August). Hatfield was next sold to Dig-It! Landscapes Pty Ltd for Kilcoy High School, Queensland on 22 February 2008 and was identified as the informal release date for the *Cynodon* variety (W Scattini 2008, pers. comm., 31 August). Hatfield was granted PBR in Australia on 20 August 2004 (Commonwealth of Australia, 2005h).

Legend® (C1)

The accession number S-49 (M Robinson 2008, pers. comm., 16 September), later designated 'C1' [*Cynodon dactylon* (L.) Pers.] was selected from a series of trials initiated by The Victorian Department of Agriculture (Daratech) and the Melbourne Cricket Club (P Semos 2006, pers.
comm., 18 August). The trials setup as the National Bentgrass and Couchgrass Evaluation Trials involved selection and evaluation processes which were carried out between 1986 and 1991 and were one of the first [earlier studies were undertaken at ATRI (P McMaugh 2009, pers. comm., 11 November)] of its kind to be formally undertaken within Australia (Robinson and Neylan, 1997). At the time, this was made possible with Turfgrass Technology (a division of Daratech) receiving funding from a succession of financial contributors including the Australian Golf Union, Victorian Golf Association, Victorian Golf Course Superintendents Association, the Melbourne Cricket Club and the Horticultural Research and Development Corporation (Robinson and Neylan, 1993). А total of six couchgrass trial sites were setup across Australia with the majority of the sites being established between September and November 1991, but a later site was also setup in Victoria in October 1994 (Robinson and Neylan, 1997). The six trial sites were located in Perth (Collier Park Golf Course, Como, WA), Melbourne (Turfgrass Technology's Research Station at Frankston, but later transferred to the Peninsula Country Club, Frankston, Victoria in November 1992 due to the uncertainty of the future of the research station and a second site at the Greenacres Golf Club which was the last site to be established in 1994), Sydney (The Australian Golf Club, Kensington, New South Wales), Adelaide (Riverside Golf Club, West Lakes, South Australia) and the Gold Coast (The Gold Coast Burleigh Golf Club, Miami, Queensland), providing different environmental conditions in a variety of climatic regions across Australia (Robinson and Neylan, 1993). The trial evaluated some 400 couchgrass accessions (P Semos 2006, pers. comm., 18 August). The C1 tetraploid (2n = 36) which was one of the 400 accessions was a vegetative collection taken from Shepparton Tennis Club (court nearest the road), Victoria in March 1986 made by John Neylan (Ho, 1999; Robinson and Neylan, 1993), who was at the time, an agronomist with the Turf Research and Advisory Institute, Victorian Department of Agriculture. Released in 1993, C1 was selected from the trial and in 1997 trademarked as Legend® (P Semos 2006, pers. comm., 18 August) by the Australian turf company StrathAyr Pty Ltd. Vegetative material of C1 selected from the National Couch-Grass Trial, was later provided to StrathAyr Pty Ltd for subsequent multiplication and later production. Today Legend® is used as a turf cover in a range of high and low input facilities, including major sporting stadia within Australia.

MiniVerdeTM (P18)

'P18' trademarked as MiniVerde[™] [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was first produced in 1992 by the late Howard Kaerwer as part of a breeding program designed to develop improved varieties of seed producing *Cynodon sp.* MiniVerde[™] is a mutant obtained from

a *Cynodon* line believed to be 'Tifdwarf' which was grown in a greenhouse owned by H&H Seed Company in Yuma, Arizona, USA (Kaerwer, 2001). MiniVerdeTM posees a high shoot density and tolerates continuous close mowing required for use in the golf and lawn bowling industry. The variety was selected for its extremely fine leaf texture, rapid growth rate, uniform dark green colour (Roche and Loch, 2008b) and excellent low temperature colour retention. MiniVerdeTM does not exhibit purple leaf coloration due to anthocyanin production typical of Tifdwarf exposed to low, non-freezing temperatures (White n.d.). Uncontaminated MiniVerdeTM was first sold in the United States on 19 July 2005 (Roche and Loch, 2008b).

Material of MiniVerde[™] which was sent by Turfgrass America arrived in Australia and was planted at DAFF Redlands Research Facility, Queensland on 8 June 2006 (Roche and Loch, 2008b). The variety was later trialled for the purposes of obtaining Plant Breeder's Rights certification. PBR of P18 was granted on 27 June 2008. Foundation material of P18 (approximately 100m²) was planted at DAFF Redlands on 10 November 2006 to multiply and supply to the Australian licensee(s). Evergreen Turf was in discussions with Turfgrass America to obtain a licence here in Australia, but negotiations ended in late 2012 and a deal was not reached. As the time of publication the P18 foundation plot was still at DAFF Redlands, but it would be sometime if the variety ever became developed commercially here in Australia.

The first enterprise in Australia to be supplied planting material of MiniVerde[™] was Coorparoo Bowls Club, Coorparoo, Queensland on 14 September 2009 to sprig their number two (eastern) bowling green. Following permission from John Holmes, Global Sales Manager of Phillip Jennings Turf Farm, Sandersville, Georgia, USA, DAFF supplied vegetative material to contract greenkeeper Darryl Bain (Professional Greens Management Pty Ltd) of the club following discussions with Queensland Bowls who wanted to upgrade one of their two greens. The purpose of this was not only to improve the problematic number two green at, but also to supply a potentially improved turf variety to the as of 2010 Queensland Bowls High Performance Training Centre as of 2010. The high performance program is part of Queensland Bowls 2008-2012 strategic initiative. The program is being run in collaboration with the Queensland Bowls to be acknowledged as the number one elite bowling state in Australia.

MiniVerde[™] had also been trialled by DAFF at various regional sites across Australia between 2006 and 2010. Once such trial included the Horticulture Australia Limited funded study 'Management of new warm-season greens grasses in Australia' (project code TU05001). A total of seven sites were constructed as part of the project, of which all seven trialled MiniVerde[™]. They

included Glenelg Golf Club (SA), Mornington Peninsula (Chisholm) TAFE College (VIC), Bermagui Golf Club (NSW), Sanctuary Cove Golf Club (Qld), Horton Park Golf Club (Qld), Twin Water Golf Club (Qld) and the central test facility at DAFF Redlands Research Facility, Cleveland, Qld, Australia.

Evergreen Turf of Victoria was originally listed as the Australian agent under the Australian Plant Breeder's Rights scheme. In 2012 Dean Holden of Evergreen Turf confirmed they were no longer acting as the agent for P18 (D Holden 2012, pers. comm., 24 July).

Mountain GreenTM (TL1)

Mountain Green[™] was observed in about 1989 by Barry McDonagh on the No. 8 green at the Townsville Golf Course, Townsville, Queensland, Australia. Designated by Tropical Lawns Pty Ltd, Cairns, Queensland and trialled as 'TL1', the dark green chance seedling was selected from a 'Tifgreen' putting green as a distinctly coarser, densely matted turfgrass. Although Mountain Green[™] was selected from a sward of the *Cvnodon* hybrid variety Tifgreen, its inflorescence structure (4, not 3, racemes per inflorescence), agronomic attributes (e.g. its tolerance to certain herbicides) and its DNA profile are consistent with a chance seedling of Cynodon dactylon rather than a mutant plant of a hybrid (C. dactylon x C. transvaalensis) origin (Loch and Roche, 2003d). Distinct characteristics of Mountain Green[™] [Cynodon dactylon (L.) Pers.] include having very short internodes, prostrate growth habit, dark green colour, and a deep, strong rhizome system. Mountain GreenTM is suited to moderate wear situations and tolerates shaded environments better than other warm-season Cynodon varieties, with the exception of 'Plateau'. Mountain Green[™] has been sold since about mid-2002 as a golf greens grass and is in play in areas of the wet tropics of North Queensland, namely the courses at El Arish, Dunk Island and Babinda (T Anderlini 2009, pers. comm., 30 November). When questioned on 30 November 2009, Terry Anderlini Anderlini from Tropical Lawns Pty Ltd stated that to date, at no stage has Mountain Green[™] been sold for home lawns or landscaping. However, this does not mean that the variety would not meet the requirements of a commercial or home lawn particularly in areas of far North Queensland. TL1 was granted PBR on 24 February 2005 (Commonwealth of Australia, 2005i).

MS-Choice

'MS-Choice' [*Cynodon dactylon* (L.) Pers.] (Krans et al, 1995c; Krans and Philley, 1998c; USDA et al., 2006c), released on 21 May 1991 was developed at the Plant Science Research Centre,

Mississippi Agricultural and Forestry Experiment Station, Mississippi, USA. MS-Choice originated from a single clone collected from the 13th fairway at the Shady Oaks Country Club, Jackson, MS on August 21 1980 (Krans et al, 1995c). Persons knowledgeable of Shady Oaks Country Club's history have said the fairway where the selected accession had been collected had not been intentionally replanted with couch grass seed, sprigs, plugs or sod since 1913 (Krans and Philley, 1998c). MS-Choice's origin may be from any one of the following sources: (a) a seed within the original seed lot; (b) a seed or plant introduced unintentionally to the site; or (c) a plant which developed as a result of an environmentally selected mutation(s) (Krans and Philley, 1998c).

The bermudagrass is characterised by a dark green colour, high-shoot density, low seed-head density, medium-coarse leaf texture, good autumn colour retention, average sod strength, good cold tolerance, excellent shade tolerance as compared to other bermudagrasses, some dollar spot, and average leaf-spot resistance (Krans and Philley, 1998c).

Breeding and foundation stock of MS-Choice is maintained by the Mississippi Agricultural and Forestry Experiment Station. Certified sod and sprigs are marketed by the Mississippi Sod Producers Association (Krans et al, 1995c).

Vegetative material of MS-Choice was obtained from Mississippi State University and later planted at DAFF Redlands Research Facility, Queensland on 11 January 2005.

MS-Express

'MS-Express' *Cynodon* x *magennisii* Hurc. (Krans et al., 1995a; USDA et al., 2006d), released on 21 October 1991 was developed by the Plant Science Research Centre, Mississippi Agricultural and Forestry Experiment Station, Mississippi. MS-Express originated from a single plant collected from the 10th fairway at the Shady Oaks Country Club, Jackson, Mississippi, on 21 August 1980. Fairways from where the collection was taken at Shady Oaks Country Club had been previously planted with bermudagrass seed between 1913 and 1933.

MS-Express origin may be from anyone of the following sources: (a) a seed within the original seed lot; (b) a seed or plant introduced unintentionally to the site; or (c) a plant which developed as a result of an environmentally selected mutation(s) (Krans and Philley, 1998a).

The identifying features of the original clone of MS-Express were characterised by a medium green colour, prostrate leaf growth, high shoot density, moderate seed-head density, very fine leaf texture, autumn colour retention, average sod strength, excellent cold tolerance, good shaded tolerance

compared to other bermudagrasses, good dollar spot and good leaf-spot resistance (Krans and Philley, 1998a).

Breeder and foundation stock of MS-Express is maintained by the Mississippi Agricultural and Forestry Experiment Station. Certified sod and sprigs are marketed by the Mississippi Sod Producers Association (Krans et al., 1995a). Vegetative material of MS-Express was obtained from Mississippi State University and later planted at DAFF Redlands Research Facility, Queensland on 11 January 2005.

MS-Pride

'MS-Pride' *Cynodon* x *magennisii* Hurc., (Krans et al., 1995b;USDA et al., 2006f)], released on 21 October 1991, was developed at the Plant Science Research Center, Mississippi Agricultural and Forestry Experiment Station, Mississippi, USA. MS-Pride originated from a single selection collected from the 5th fairway at the Vicksburg Country Club, Mississippi, on 20 August 1980. Persons knowledgeable of Vicksburg Country Club's history have said that the fairway was established with Bermudagrass seed in the mid-1960's and had not been intentionally replanted with bermudagrass seed, sprigs, plugs or sod since 1960 (Krans et al., 1998b). MS-Pride's origin may be from anyone of the following sources: (a) a seed within the original seed lot; (b) a seed or plant introduced unintentionally to the site; or (c) a plant which developed as a result of an environmentally selected mutation(s) (Krans et al., 1998b).

The identifying features of the original clone of MS-Pride were characterised by a medium to dark green colour, short-head density, low seed-head density, fine leaf texture, excellent autumn colour retention, excellent sod strength, average cold tolerance, good shade tolerance as compared to other couchgrass, excellent dollar spot and leaf-spot resistance (Krans et al., 1998b).

Breeder and foundation stock of MS-Pride is maintained by the Mississippi Agricultural and Forestry Experiment Station. Certified sod and sprigs are marketed by the Mississippi Sod Producers Association (Krans et al., 1995b). Supply of vegetative material of MS-Pride in Australia was obtained from Mississippi State University and later planted at DAFF Redlands Research Facility, Queensland on 11 January 2005.

MS-Supreme

The 'MS-Supreme' *Cynodon* x *magennisii* Hurc. (Krans et al., 1999; USDA et al., 2006f) hybrid was discovered in 1991 by Jeffrey V. Krans as a mutant plant in the No. 14 'Tifgreen' putting green

that had been planted in 1964 at the Gulf Shores Country Club, Gulf Shores, Alabama, USA (Krans et al., 1999). MS-Supreme developed in 1998 is a high-density, fine-textured, fast prostrate growing variety that can withstand a sustained 3.2 mm cutting height. Krans et al. (1999) made the selection as it maintained a darker green colour and higher shoot density than the surrounding Tifgreen during extended periods of wet, overcast weather. Other selection characteristics included its narrow leaves and prostrate growth habit. MS-Supreme was first sold in the United States on 9 June 1999 (Loch and Roche, 2003a).

Vegetative material of MS-Supreme was obtained from Mississippi State University, USA and later planted at DAFF Redlands Research Facility, Queensland in their turf demonstration plots on 13 February 2001. MS-Supreme was initially trialled for the purposes of obtaining Plant Breeder's Rights. The variety was granted PBR on 25 February 2005 (Loch and Roche, 2003a). MS-Supreme was subsequently trialled in a study to assess the 'Management of new warm-season greens grasses in Australia' as part of a Horticulture Australia Limited funded study (project code TU05001). Seven sites participated in the four year study (2006-2010) which saw a range of greens quality grasses being trialled at venues positioned predominantly down the eastern coast of Australia. They included Glenelg Golf Club (South Australia), Mornington Peninsula TAFE College (Victoria), Bermagui Golf Club (New South Wales) and the three Queensland venues at Horton Park Golf Club, Twin Water Golf Club and the central test facility at Redlands Research Facility, Cleveland, Qld, Australia.

NovotekTM (TL2)

Vegetative material was taken from a disease resistant mutant plant by Terry Anderlini on the No. 15 green at Novotel Palm Cove resort course near Cairns, Queensland, Australia in 1996 (Roche and Loch, 2003b). Later designed 'TL2' the triploid (2n = 27) (Roche and Loch, 2003b) interspecific hybrid [*Cynodon dactylon* (L.) Pers. X *C. transvaalensis* Burtt-Davy] was included an on-going program of selection and testing of promising 'Tifgreen' (Tifton 328) mutants by Tropical Lawns Pty Ltd. Novotek[™], first sold commercially in Australia in 2003, produces a healthy vigorous growth during the tropical wet season, dense fine-textured appearance under close mowing, and dark green leaves. In subsequent trials conducted by Terry Anderlini, Novotek[™] was identified as the outstanding plant among selections of mutant Tifgreen genotypes from other north Queensland sites in terms of colour, texture and density for greens use (Roche and Loch, 2003b).

TL2 was initially trialled for the purposes of obtaining Plant Breeder's Rights. PBR was granted on 24 February 2005 (Roche and Loch, 2003b). Novotek[™] was subsequently trialled in a study to

assess the 'Management of new warm-season greens grasses in Australia' as part of Horticulture Australia (HAL) funded initiative (project code TU05001). Out of the seven regional trial sites positioned predominantly down the eastern coast of Australia, three trialled the variety between 2006 and 2010. They included the Queensland venues at Horton Park Golf Club, Twin Water Golf Club and the central test facility at Redlands Research Facility, Cleveland, Qld, Australia.

OZ TUFFTM (Oz-E-Green)

'Oz-E-Green' [*Cynodon dactylon* (L.) Pers.] was discovered by Robert W. Morrow in 2001 as a superior plant growing among "Common" bermudagrass (*Cynodon dactylon*) on his turf farm, Oz Tuff Turf at Berries Road, Childers, Queensland, Australia (Loch and Roche, 2004). Registered as OZ TUFF®, the variety was granted Plant Breeder's Rights on 22 August 2005 (Commonwealth of Australia, 2005g) and was later released on 13 April 2006 (W Morrow 2006, pers. comm., 10 August). The selection criteria for Oz-E-Green included a dense prostrate growth habit and limited inflorescence production, high turf quality, and a dark green leaf colour (Loch and Roche, 2004).

OZ TUFF® was included in a four year Horticulture Australia (HAL) funded trial (project code TU08018) conducted by DAFF investigating wear tolerance and recovery of a selection of C4 warm-season turfgrass varieties. Throughout the duration of the study, 2008-2012, OZ TUFF® showed significant wear tolerance in the simulated and actual wear (in play on touch football fields) studies conducted at DAFF Redlands Research Facility and Redlands Touch Football fields respectively.

As of March 2013 OZ TUFF® was being sold commercially by Rosemount Turf, Tropical Lawns, Bay Turf, Glenview Turf, Australian Lawn Concepts, All Turf Solutions, Cobbitty Turf, Pakenham Turf and Oz Tuff Turf.

Patriot

'Patriot' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was commercially released in June 2002 by the Oklahoma State University/Oklahoma Agricultural Experiment Station, USA (Anonymous, 2004). Patriot is a tetraploid (2n = 36) having received 27 chromosomes (three basic genomes) from the 'Tifton 10' *Cynodon dactylon* var. *dactylon* hexaploid (2n=6x=54) parent and 9 chromosomes (one basic genome) from the *C. transvaalensis* (parent) (Taliaferro et al., 2004a). Although Patriot has the same chromosome number as most *C. dactylon* var. *dactylon* plants, it is highly sterile only rarely producing seed (Taliaferro et al., 2004a). Patriot previously designated OKC 18-4 [OKC represents Oklahoma State University Clonal (vegetative) Type (Martin, 2002)] was developed and extensively tested between 1997 and 2001 by the Oklahoma State University Bermudagrass Breeding and Development Team. The interspecific F_1 hybrid Patriot offers a dark blue-green colour, high shoot density, medium-fine texture, improved cold hardiness, a level of resistance to spring dead spot, rapid establishment and speedy divot recovery.

Patriot is available only as certified sprigs or certified sod and is a proprietary variety, exclusively licensed for production in Oklahoma to Easton Sod Farms (ESF), Inc. (Anonymous, 2004).

Material of Patriot which was sent by Oklahoma State University arrived in Australia and was planted at DAFF Redlands Research Facility, Queensland in their turf demonstration facility on 11 January 2005.

Plateau

'Plateau' is the result of a "spontaneous mutation" of an unnamed and unpatented common *Cynodon dactylon* growing on the property of the inventor, Peter Brown, at Collaroy Plateau, New South Wales, Australia in 1975 (Brown, 2002). Plateau [*Cynodon dactylon* (L.) Pers.] is described by Brown (2002) as being infertile, however studies undertaken by Professor Peter Martin of the University of Sydney concluded the variety is fertile (P McMaugh 2009, pers. comm., 11 November). Plateau, released in 1996, exhibits low growing height, prostrate spreading habit, prostrate shoot growth, short internode length, high sward density, wide stolon to rhizome width ratio, and medium seed head frequency (Brown, 2002). Due to the low compact growth and broad leaf of Plateau the variety is one of the more favourable selections of *Cynodon* currently available for shaded environments (>30% sun).

Brown (2002) acknowledges in the PBR application that the most similar known variety of *Cynodon dactylon* is 'Riley's Super Sport' that forms a dense turf of low growing height; the next variety of morphological comparison is the variety 'Greenlees Park'. However, morphological and developmental studies undertaken by Loch & Roche (2003d) confirm that this assertion is erroneous. The most similar variety, today, is Mountain GreenTM ('TL1'); however 8 out of the 19 characteristics measured in a spaced plant experiment conducted at DAFF Redlands, Qld show Plateau and Mountain GreenTM are significantly different (P≤0.01) (Loch and Roche, 2003d). PBR was granted on 25 February 2000 (Kaapro, 1999c).

Premier

⁶Premier' was discovered by Donald La Verne Parsons and Virginia Gail Lehman under cultivated conditions in a golf course fairway near Seal Beach, California, USA (Parsons and Lehman, 2007). Since Premier was introduced into Australia by Virginia Leman, Oregan, USA, in 2007, observations made by Dr Donald Loch (former Principal Scientist, DPI&F) and Matthew Roche (former Senior Scientist, DAFF) believed the variety was not a *Cynodon dactylon* (L.) Pers. as first identified by Parsons and Lehman (2007). Following morphological and agronomic studies undertaken between 4 October 2007 and 6 February 2008 at DAFF Redlands Research Facility, Queensland, the results obtained from this work strengthened Loch and Roche's claim. Communication between Matthew Roche and Dr Milton C. Engelke, turf breeder at Texas A&M University, identified that the variety was in fact a hybrid bermudagrass (*C. dactylon* x *C. transvaalensis*) (M Engelke 2009, pers. comm., 20 November 2009). Dr Engelke is married to Dr Virginia Gail Lehman who's turf farm, Blue Moon Farm, Lebanon, Oregon, USA, has ownership of Premier.

Premier was identified as a distinctly different vegetative patch or segregated clonal plant differing by darker green leaf colour from the suspected parental variety 'Tifgreen' (Tifton 328) (Parsons and Lehman, 2007).

Vegetative material of Premier obtained from Dr Lehman was planted at DAFF Redlands Research Facility, Queensland in their turf demonstration facility on 26 August 2010.

Riley's Super Sport

'Riley's Super Sport' [*Cynodon dactylon* (L.) Pers.] was granted Plant Breeder's Rights in Australia on 28 February 1997 (Commonwealth of Australia, 2005a) and was released soon after. The variety which is marketed as CelebrationTM in the USA, was also released in the latter country by Sod Solutions Inc. Riley's Super Sport was a result of "spontaneous mutation" from the *Cynodon dactylon* variety 'Greenlees Park' and selected by the late Rod J. Riley, Guilford, New South Wales in 1988 (Kaapro, 1996). The infertile (McMaugh, 2008) Riley's Super Sport exhibits a very prostrate growth habit thereby having minimal vertical growth (that is, very prostrate growth habit), extensive leaf production, short internode length, very low seed head production, and deep green coloration (Riley, 2000).

Royal Cape

'Royal Cape' [*Cynodon dactylon* (L.) Pers.] was discovered [in about 1930 (Taliaferro, 2003)] on the Royal Cape Golf Course at Wynberg, Cape Town, South Africa (Taliaferro, 2003). The turfgrass PI 213387 was selected by C. M. Murray of South Africa (Busey, 1989) and released by the University of California, Los Angeles, and the Crops Research Division, ARS, USDA in 1960 (Younger et al., 1972). Royal Cape was chosen for use along the lower Colorado River Basin (Younger et al., 1972) and was released on the basis of late-autumn and early spring growth, good colour, texture, tolerance to saline soils, and limited production of seed heads (Hanson and Juska, 1969).

A selection of Royal Cape was introduced into Australia prior to 1956 by the Royal Botanic Gardens in Sydney (McMaugh, 2008). Vegetative material of a strain of Royal Cape was obtained from R. J. Weppner, Toowoomba, Queensland by DAFF and planted at Redlands Research Facility, Queensland, in their turf demonstration plots on 25 May 2000. A proportion of this material was taken and trialed in a Plant Breeder's Rights spaced plant trial at RRS in 2003. Post planting on 4 March 2003 significant variation within the variety was observed and a collection of the two genotypes were made, highlighting the inconsistency within the variety. A selection of superior quality 'Royal Cape' was made by M.B. Roche and Dr D.S. Loch of DAFF and later named 'RCII'. 'RCII' was vegetatively propagated to obtain sufficient material to trial in further PBR spaced plant trials (10 June to 15 December 2004) undertaken by the former DAFF turf research team. 'RCII' has not been commercially protected nor released. Vegetative material of 'RCII' is maintained in a pure state at DAFF Redlands Research Facility for subsequent trialling and development.

Santa Ana

'Santa Ana' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was developed by researchers at the University of California, Riverside, Los Angeles, USA for parks, playground, sport fields (Augsdofer, 1995) and general home use. Named after the Californian city, the variety was a result of a deliberate cross (McMaugh, 1987) with grasses from South Africa and Iran (Augsdofer, 1995) by the late Dr Victor B. Younger. The parentage was from crossing the South African *C. dactylon* variety PI 213387 (Anderson and Sharp, 1995), otherwise known as 'Royal Cape' and a fine leaved, dark green seedling selection of *C. transvaalensis* which was originally introduced from the Orange Free State in 1983 (V Younger 1993, pers. comm. to P Leroy, 20 April) [McMaugh (P McMaugh 2009, pers. comm., 11 November) said Iran in 1987]. PI 213387 which

was released in 1960 was selected by C. M. Murry (Anderson and Sharp, 1995) from the Royal Cape Golf Course near Mowbray, Cape Province, Union of South Africa in 1930 (Taliaferro, 2000).

The result was a seedling that was selected from the University of California agronomy program in 1956 and was identified as RC145 (Beehag 1987). A source of RC145 was planted into field plots at the Santa Ana Research Station in 1958 (G Beehag 2009, pers. comm., 6 October) and observations were undertaken at locations throughout Chaffer and in several other states as RC145 (Anonymous n.d.). RC145, later designated Santa Ana was released in the USA in 1966 (Hanson, 1959; Hanson and Juska, 1969; Younger et al., 1972) [Beehag (1987) and Augsdorfer (1995) said it was released in 1967] by the California Agricultural Experiment Station.

Santa Ana a sterile triploid (2n = 27) hybrid (Anderson and Sharp, 1995) is characterised by a deep blue-green colour, medium-fine texture, good colour retention, early green-up of the turf following winter (Anderson and Sharp, 1995), high wear tolerance and *eriophyid* mite (*Eriophyes cynodoniensis* Sayed) resistance (Hanson and Juska, 1969). Observations made by Dr Younger was that Santa Ana also possessed good tolerance to smog in Los Angeles and above average salinity tolerance (Beehag, 1987). Even with these positives, Santa Ana, at first, was not well accepted by the sod growers in the USA with the growers arguing that they didn't want it. This was because the growers at the time were already growing the hybrid bermudagrasses 'Tifgreen' and 'Tifway', and it was thought to be a huge investment to plant additional acres of an additional hybrid turf to make it worth their while (Augsdofer, 1995). This mindset was soon forgotten and the turf was widely used and popular for recreational and sports turf use within the USA.

In Australia, Santa Ana was released by the Plant Quarantine Service of the Commonwealth Department of Health (Australian Capital Territory) in late 1976. The variety had been introduced by City Parks Research in Canberra to compliment the work being undertaken by Dick Powell, John Mortimer and Peter Semos (McMaugh, 1988). Soon after, sod farms were producing commercial supplies of Santa Ana in Adelaide, Sydney and Perth (McMaugh, 1987). The variety was widely accepted for use on bowling greens with the most concentrated use at the time being in Adelaide, where over half the bowling clubs decided to adopt Santa Ana as their preferred turf for their greens (Beehag, 1987). Santa Ana was also a preferred choice for use on cricket wicket blocks. One example of its use is when it was planted at the Melbourne Cricket Ground following a problem with their wicket table. This problem led to the subsequent rotary hoeing of the wicket table in late 1981 under the direction of the then secretary, Ian Johnson (McMaugh, 2008). Santa Ana is still widely used as a suitable turfgrass for wicket blocks because of its fine texture and wear tolerance. Santa Ana performs well in cooler climatic zones like that of Melbourne and Sydney. Further north,

in Queensland Santa Ana does reasonably well during cooler periods; however in the warmer months, the turfgrass undergoes higher stress levels which results in a significant reduction in turfgrass quality. However, Goddard (1999) commented that Santa Ana was well suited to Western Austalia's hot dry summers and cool, wet winters when the variety was introduced into that state in the early 1980s. Santa Ana also produces high thatch levels all year round and often results in management problems.

SS-2

The *Cynodon dactylon* variety was selected by Max Stephenson of Twin View Turf, Qld. The variety identified as 'SS-2' or "secret stuff two" was made from a natural selection at undisclosed location. The variety to date has not yet been released.

Tifdwarf

There were two sources of vegetative material produced of 'Tifdwarf' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy], being for a test plot at the Georgia Coastal Plain Experiment Station and for later commercial production.

In 1962 the test plot selection was found occupying an area of about 457.2 mm (18 inches) in diameter by T. M. Baumgardner and Marion McKendree on the No. 2 green of the Plantation Course (Moncief, 1967), Sea Island Country Club, Sea Island, Georgia and by James Moncrief on the No. 12 green (Moncief, 1967) at Florence Country Club, Florence, S.C. (Anderson and Sharp, 1995). The source for a commercial nursery came from the No. 6 green at Glen Arven Country Club, Thomsaville, Georgia also in 1962 (Moncief, 1967).

A careful evaluation of all evidence indicates that Tifdwarf is a vegetative mutant that occurred in 'Tifgreen' (Tifton 328) at Tifton, Georgia, USA, before the first planting stock was sent out in 1954 for preliminary evaluation (Anderson and Sharp, 1995). It is believed that the golf courses at Florence and Sea Island Country Club, each received a sprig or two of this original natural mutation that occurred at Tifton Experimental Station. Tifdwarf underwent three years of research under the late Dr Glen W. Burton and graduate students, and two years of field testing against its comparator Tifgreen (Moncief, 1967) and was later officially released by the Georgia AES, Tifton, and Crops Research Division, U.S. Department of Agriculture in April, 1965 (Moncief, 1967; Anderson and Sharp, 1995; Hanna and Elsner, 1999). Tifdwarf is a dwarf type with small, short leaves, stems, internodes, and seedheads and provides a dark green colour throughout the warmer months.

However like other "ultradwarfs" Tifdwarf is assisted by its basic purple plant colour in the warmer months and becomes very noticeable when temperatures drop in winter. As a consequence, Tifdwarf takes on a purplish cast that is aesthetically objectionable to some.

Tifdwarf was mistakenly identified as 'Tiffany Grass' by bowling greenkeepers in northern New South Wales, but their Queensland counterparts were quick to recognise its unique growth habits and limited seed head production (Beehag, 2006). Ocean Shores Golf Club, Ocean Shores, New South Wales was an early residential golf course in Australia and Beehag (2006) suggests the golf club can lay claim to having the oldest Tifdwarf golf greens in Australia planted from 1970 (G Beehag 2009, pers. comm., 16 September). The material imported by greenkeeper Cliff Meredith (P McMaugh 2009, pers. comm., 11 November) was provided to Vic Phelps in 1968 to establish a nursery at his home for use at the then development Wendell West, which is now called Ocean Shores Golf Course; Vic at the time was the greenkeeper at the Byron Bay Golf Club, Byron Bay, New South Wales (G Beehag 2009, pers. comm., 16 September). Broadwater Bowling Club, New South Wales first planted the Tifdwarf on a full sized bowling green in 1973 using the same source of material as Ocean Shores (G Beehag 2009, pers. comm., 16 September). Roy Hulbert (who at the time was the greenkeeper at Bangalow Bowling Club, Bangalow, NSW assisted Ron Mathews (greenkeeper at Byron Bay Bowling Club) to plant Tifdwarf into the worn corner of the number one green which was then bentgrass (Agrostis spp.) of the Byron Bay Bowling Club in May 1969; The latter green was re-grassed completely to Tifdwarf on 13 September 1973 using material from the same source as was Broadwater Bowling Club (G Beehag 2009, pers. comm., 16 September). The green was in play on 21 November 1973 (G Beehag 2009, pers. comm., 6 October). Broadwater Bowling Club was the first full-sized green to be planted on the New South Wales North coast in April/May 1973 which had been planted by Rex Gulding before the Byron Bay Bowling Club, but was not in-play until after the latter club (G Beehag 2009, pers. comm., 6 October).

Commercial Tifdwarf planting material in the early days came from Banora Lawn Turf, Banora Point, New South Wales which was operated by Ray Jarred, Roy Hulbert's brother-in-law (G Beehag 2009, pers. comm., 6 October).

Pennant Hills Golf Club, Sydney, New South Wales planted a practice putting green (which has now been removed) as early as 1975 (Beehag, 2006). Vegetative material to source planting stock for this green was allegedly brought into Sydney ex Tifton, USA in May 1966 through the mail (G Beehag 2009, pers. comm., 6 October).

Tifdwarf was used on a 9-hole Darwin golf course in 1978; In South Australia, Tifdwarf was first planted at the Holdfast Bowling Club in 1975 (G Beehag 2009, pers. comm., 6 October).

Tifdwarf was used in far north Queensland from the late 1960s at Edmonton Bowling Club, Edmonton and has been the source of the variety in this region (information ex Rod Cade); it was also planted at the Gold Coast Ladies Bowling Club, the first on the Gold Coast to use the hybrid bermudagrass in the early 1970s (G Beehag 2009, pers. comm., 16 September). In Brisbane, an early club that had changed to Tifdwarf was Salsbury Bowling Club, Salsbury, Queensland (information ex Les Rowan) (G Beehag 2009, pers. comm., 6 October).

Foundation plant material of Tifdwarf is maintained by the Georgia Coastal Plain Experiment Station, Tifton, Georgia, USA. Mr Ray Jensen of Tifton, Georgia claimed in 1993 that he had the only true supply of Tifdwarf which he maintained as uncut plant material to prevent it from mutating; Mr Jensen believed that if the highly unstable Tifdwarf plant was cut short in the field it had a greater chance of producing new forms [off-types] than that by irradiation, or hybridisation, in the laboratories (G W Burton 1993, pers. comm. with G Beehag, 11 August).

TifEagle

'TifEagle' is a fine-textured Cynodon dactylon (L.) Pers. x C. transvaalensis Burtt-Davy (Hanna and Elsner, 1999; Hanna, 1997) variety suited for golf and bowling greens and other applications requiring regular close mowing. The "off-type" (Mutant No. 2) was selected from 48 such mutant plants (Hanna, 1998) in 1990 from a plot established from dormant stolons of the 'Tifway II' variety (C. dactylon x C. transvaalensis) previously treated with gamma radiation on January 12 1988 (Hanna and Elsner, 1999). TifEagle is a dense, fine-textured triploid (2n = 27) (Hanna n.d.2; Hanna, 1999a; Loch and Hanna, 2001a) that produces more shoots per unit area that are shorter with narrower leaves, better turf quality and colour and greater resistance to the tawny mole cricket (Scapteriscus Vicinus) than 'Tifdwarf' (Hanna, 1999a). Test plantings on experimental plots and putting greens since 1991 indicate that TifEagle can withstand routine cutting height of 3 mm and due to its canopy, can ensure a golfer's ball rolls quickly in the direction it was putted (Hanna, 1998). TifEagle was cooperatively released by United States Department of Agriculture -Agricultural Research Services (USDA-ARS) and the University of Georgia Coastal Plain Experiment Station in August 1997 (Miller and Edenfield, 2002). TifEagle was first sold in the USA in May 1999 (Loch and Hanna, 2001a) and foundation material is maintained by the USDA-ARS, Coastal Plain Experiment Station, Tifton, Georgia.

Material of TifEagle arrived in Australia and was planted at DAFF Redlands Research Facility, Queensland in their turf demonstration plots on 13 February 2001. Plant Breeder's Rights in Australia was granted on 22 May 2002 following testing at Redlands (Loch and Hanna, 2001a). The greens quality TifEagle for use on lawn bowling and golf greens is handled by the sole Australian licensee Twin View Turf Pty Ltd located at Wamuran, Queensland. Twin View Turf Pty Ltd first sold TifEagle to the North Lakes Golf Club, Mango Hill, Queensland, Australia to establish their 18 hole golf course which opened in 2002.

Tiffine

Hybridisation of *Cynodon dactylon* types with South African Bermuda (*Cynodon transvaalensis*) produced 8 (Hanson, 1959) [Hein (1953) said 8 or 9; and Robinson & Latham (1956) mistakenly wrote 89, which is likely to be a result of a possible typographic error from Robertson and Burton (1953) writing eight-nine at the start of a paragraph insteasd of using numerical identifiers] hybrid plants from which Tifton 127 later named 'Tiffine'. The *Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy variety was selected and released by the Georgia AES, Tifton, and Crops Research Division, ARS, USDA in 1953 (Robinson and Latham, 1956). An F₁ hybrid and tested as Tifton 127, Tiffine is lighter green, more disease resistant, and much finer than common couchgrass (Hanson and Juska, 1969). The variety was introduced into Australia by Doug Corbett, former teacher of Greenkeeping at the Ryde School of Horticulture, New South Wales, following an informal visit to Sports Turf Research Institute (STRI) in Palmerston North, New Zealand. However, the material was mishandled and subsequently its identity lost by the staff at the Ryde School of Horticulture following building extensions in 1970 where the variety had been established in the field (McMaugh, 2008).

Tifgreen (Tifton 328)

During 1946, W. G. Thomas, Chairman of the Green Committee, and Walter Harkey, Superintendent of the Charlotte Country Club, North Carolina, USA, observed a fine-textured bermudagrass growing in their No. 4 green (Robinson and Latham, 1956). The selection was collected and planted in the turf plots at Tifton, Georgia, for further observation. In the spring of 1951 (Robinson and Latham, 1956), the "common", fine-textured, superior Charlotte Country Club, North Carolina strain (*C. dactylon*) was hybridised with a fine-leafed South African bermudagrass (*C. transvaalensis*) [Hanson (1959) says it was from East Lakes Golf Course, Atlanta, Georgia; while Hanna & Anderson (2008) says it was from Egypt]. The result was a completely sterile F^1 hybrid [triploid (2n = 27) (Hein, 1961; Anderson and Sharp, 1995)] tested as 'Tifton 328' and later registered as 'Tifgreen'. Tifgreen [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] was developed and released by the Georgia AES, Tifton, and Crops Research Division, ARS, USDA, in 1965 (Anderson and Sharp, 1995); Hein, 1961). Twelve years after release Hanna & Anderson (2008) reported that more than 8,000 greens had been converted to Tifgreen in the USA.

Vegetative material of Tifgreen was first reported to be introduced into Australia in 1956 by Doug Corbett, former teacher of Greenkeeping at the Ryde School of Horticulture, New South Wales (McMaugh, 2008). However the variety suffered its ill-fated demise following mishandling and poor labeling similar to that of the introduction of 'Tiffine' into Australia. A undisclosed golf course architect informed Gary Beehag (G Beehag 2009, pers. comm., 6 October) many years ago that he had brought vegetative material of Tifgreen from Hawaii into Sydney in 1969; some of this material was then used as planting material at the Pennant Hills Golf Club, Sydney in the practice green (which has since been removed) (G Beehag 2009, pers. comm., 6 October).

Material of Tifgreen was planted at the Gold Coast Burleigh Golf Club, Burleigh Heads, Queensland in 1974 (G Beehag 2009, pers. comm., 16 September), replacing the endemic species Queensland blue couch (*Digitaria didactyla*) as was used on up to 50% of the golf putting greens at the time (Beehag, 2006). One year later Tifgreen had become widely used on putting greens on the Gold Coast and in Brisbane (Beehag, 1992).

Foundation or breeder material of Tifgreen is maintained the Georgia Coastal Plain Experiment Station, USA.

Tifgreen-II

'Tifgreen-II', a sterile triploid (2n = 27) [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] is an improved mutant of 'Tifgreen' (Tifton 328) developed cooperatively by the US Department of Agriculture, ARS, the Georgia Costal Experiment Station, the US Golf Association Green Section, the Golf Course Superintendents Association of America (GCSAA), and the Department of Energy (Burton, 1985b).

Beginning in 1970 Powell et al. (1974) irradiated to 7000 rads of gamma irrigation (Burton, 1985b) thousands of rhizomes of [numerous] Tifton bermudagrass varieties, including Tifgreen. The treated dormant sprigs were then grown on as spaced plants and selections were made on the basis of plants or sectors of plants that appeared different (Burton, 1985b). In 1971 (Burton, 1983), one year following irradiation, Tifgreen-II was selected for having many desirable traits of Tifgreen but has a

lighter green colour and usually develops less of the undesirable purple colour when temperatures are low, is more vigorous, denser and exhibits much better spring recovery (Burton, 1985b).

One of the two clonal varieties to be released from this work included a Tifgreen-II in 1983 (Busey, 1989; Burton, 1983).

Tifgreen-II was included in the appendix as there was one report of the variety being introduced into Adelaide. However, insufficient evidence has been made available and no further information has come to light as to its use here in Australia.

Tiflawn

'Tiflawn' is a hybrid [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] between two selections of *Cynodon* that was released by the Georgia AES, Tifton, and Crops Research Division, ARS, USDA, in 1952 (Hanson and Juska, 1969); tested as Tifton 57 was developed in cooperative investigations between the latter station and the Division of Forage Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering (BPISAE) (Hein, 1953). Tiflawn was released in the US in 1956 (Hanson, 1959) [Robinson & Latham (1956) say it was released in 1950]. An F₁ hybrid that requires less fertiliser and is more wear resistant than common couchgrass (Hanson and Juska, 1969).

Tiflawn was included in the appendix; however no information has come to light on its introduction or use here in Australia.

TifSport[™] (Tift 94)

'Tift 94' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] (Hanna et al., 1997) was developed by United States Department of Agriculture-Agricultural Research Service (USDA/ARS) geneticist Dr Wayne Hanna at the University of Georgia Coastal Plain Experiment Station in Tifton, Georgia. The fine textured bermudagrass released in 1994 (Hanna et al., 1997) was a result of a mutant selected from sixty-six plants established from dormant stolons (each with two nodes) of the 'Midiron' variety [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] treated by gamma radiation on January 20, 1983 (Hanna, 1997; W W Hanna 2006, pers. comm., 10 July). After 12 years of multi-location testing, mutant number 40 was released as Tift 94 (Hanna, 1999b) and was first sold in the USA in June 1998 (Loch and Hanna, 2001b).

Trademarked as TifSportTM, the variety is a vigorous triploid (2n = 27) (Hanna n.d.1; Hanna, 1997; Hanna, 1999b; Loch and Hanna, 2001b) selected for close mowing, texture, density, resistance to southern mole cricket (*Scapteriscus borellii* Giglio-Tos; syn. *S. acletus* Rehn & Hebard), non-preference green-up characteristics and in particular its wear tolerance and recovery (Roche et al., 2009).

TifSport[™] patented in 1997 (White, 2006), is licensed exclusively to the University of Georgia Research Foundation for commercialisation and is protected by a US plant patent. TifSport[™] was brought into Australia from the USA and planted at DAFF Redlands Research Facility, Queensland in their turf demonstration plots on 3 February 2000. Tift 94 was trialled at Redlands for the purposes of conducting a Plant Breeder's Rights experiment and following successful testing, Tift 94 was granted PBR on 22 May 2002 (Loch and Hanna, 2001b). Twin View Turf Pty Ltd of Wamuran, Queensland is the only Australian licence holder of the variety TifSport[™]. TifSport[™] can only be sold as genetically certified turf or sprigs.

Tifway

'Tifway' [Cynodon dactylon (L.) Pers. x C. transvaalensis Burtt-Davy] released collaboratively in 1960 by the USDA-ARS and the Georgia Coastal Experiment Station (Hanna, 1997; Anderson and Sharp, 1995), was selected from a presumed chance hybrid between C. transvaalensis and C. dactylon that appeared in a seed lot shipped by D. Meredith, Johannesburg (Anderson and Sharp, 1995), South Africa to the USA in 1954 (Burton, 1966b). Tifway is a triploid (2n = 27) (Burton, 1960) bermudagrass that produced a darker green colour and stiff leaves, earlier spring growth, greater resistance to frost and to sod webworm [Herpetogramma licarsisalis (Walker)] and mole cricket [Gryllotalpa orientalis (= africana) Burmeister], better herbicide tolerance; the variety does not shed pollen and produces stiffer leaf blades than that of 'Tiffine' or 'Tifgreen' (Anderson and Sharp, 1995). Hanson and Juska (1969) rated 'Tifway' as equal or superior to Tiffine and 'Tifgreen' in disease resistance, density, weed resistance, seed head production, and rate of spread. Vegetative material of Tifway was introduced into Australia via New Zealand in 1956 by Doug Corbett, former teacher of Greenkeeping at the Ryde School of Horticulture, New South Wales, (McMaugh, 2008) and material was later planted (around 1965) at Beverina Estates, Cobbitty, New South Wales under the trademark SportswayTM (McMaugh, 2002). Horsfall Turf also had extensive plantings at about the same time (McMaugh, 1988). McMaugh (2002) suggested the variety suffered its ill-fated demise following mishandling and poor labeling similar to that of the introduction of Tiffine into Australia. However, Beehag (G Beehag 2009, pers. comm., 6 October) noted that Tifway had been

planted at Campbelltown Golf Club, Campbelltown and Cranbrook School, Bellevue Hill which are both located in New South Wales and still exist at the time of publication). Vegetative material of Tifway was also supplied to DAFF Redlands Research Facility in Queensland by Col Shiller of Walsh's Seeds Garden Centre, Toowoomba City, Queensland and later planted in their turf demonstration plots on 25 May 2000.

Tifway II

'Tifway II' [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] originated by exposing gamma irradiation to dormant 'Tifway' sprigs in 1971 and then selecting plants or sectors that appeared to be different (Burton, 1981). Tifway II was released cooperatively on 13 April 1984 [There are numerous variances of the date of release of which include: 1984 (Polomski and Shaughnessy, 2003); April 1981 (Anderson and Sharp, 1995); 1991 (Hanna, 1997); 1981 (Busey, 1989); 1999, (Busey and Dudeck, 1999); the article by Burton (1981) titled 'Tifway II Bermudagrass Released' was published in 1981 but no specific date was published], by the USDA-ARS, the Georgia Coastal Plain Experiment Station, the U.S. Golf Association Green Section, and the U.S. Department of Energy. Burton (1995a) noted that the sterile triploid (2n = 27) variety Tifway II looked like Tifway and had the same desirable characteristics but made a denser, more weed-free turf, more resistant to root knot, ring and sting nematodes, is more frost tolerant, establishes faster from sprigs, exhibits a little better quality, and often greens up slightly earlier in the spring.

The Georgia Coastal Plain Experiment Station, Tifton, has maintained a breeding stock of Tifway II material.

Tifway-II was included in the appendix; however no information has come to light on its introduction or use here in Australia.

Windsor Green

'Windsor Green' [*Cynodon dactylon* (L.) Pers.] was the first variety in Australia to be put through the Plant Breeder's Rights process (McMaugh, 2008). Windsor Green was granted PBR on 17 January 2004 (Commonwealth of Australia, 2005c). Released in 1993, the variety was a induced mutant from 'Wintergreen' through radiation. Windsor Green, a tetraploid (2n = 36) (Ho, 1999) was selected from 22 other mutants post radiation and screening showing superior density, colour and

wear tolerance as well as lower seed head production (McMaugh, 1993). Additional attributes included low temperature growth and high fibre (P McMaugh 2009, pers. comm., 11 November).

Winter Gem

'Winter Gem' [Cynodon dactylon (L.) Pers.] was a variety produced by control pollination followed by selection of Cynodon dactylon varieties 'Wintergreen' and 'Couch 5' (also designated C5) by breeder David Nickson. Couch 5 [accession number S-130 (Robinson and Neylan, 1993)] was an experimental breeding line (that was selected from seed provided to the breeder by Arden Battensperger (McMaugh, 2008) that was a selection from an earlier series of crosses by the breeder between Wintergreen and a number of Cynodon dactylon accessions which had been collected by the breeder from the Peninsula Country Club Fairway 8B, Frankston, Victoria, Australia with mentor Peter E. McMaugh in February 1987 (Neylan, 2005; M Robinson 2008, pers. comm., 16 September). A series of other Cynodon collections were carried out between 1986 and 1990 from the Peninsula area of Victoria. Crossing was carried out by the breeder in 1998 and subsequent seedlings and selected plants were observed between 1998 and 2000. In the spring of 2000, the remaining potted seedlings were planted into plots at the Evergreen Turf farm at Pakenham, Victoria, Australia and allowed to expand fully. The final selection of Seedling 9 (later designated DN9) in late 2002 was based on shoot density, leaf texture and retention of winter colour (Roche and Loch, 2006b). Winter Gem was granted PBR on 11 September 2006 (Commonwealth of Australia, 2005b) and was first sold to Victorian Parks Constructions in February 2007 for use at the Grand Prix racetrack, Albert Park, Melbourne, Victoria, Australia (D Holden 2009, pers. comm., 17 June).

Wintergreen (C84-135)

'C84-135' or more commonly known as 'Wintergreen' [*Cynodon dactylon* (L.) Pers.] was discovered by Peter E. McMaugh in 1969 (Ho et al., 1997; Ho, 1999) growing on a small bowling green surface at Nyngan, New South Wales, Australia. Wintergreen was selected for its vigorous growth, dark green olive colour and colour retention compared to other *Cynodon dactylon* varieties in Australia at the time. Following 10 years of experimenting under test and development (McMaugh, 1988), Wintergreen was released in 1983 (McMaugh, 2008). The tetraploid (2n = 36) variety which was described by John Neylan, then AGCSA Technical Manager 'as an oldie but a goodie' (McMaugh, 2005; McMaugh, 2008) is still widely used today within sub-tropical and warm

temperate zones within Australia. However, being able to identify a true-to-type source of "Wintergreen" through years of sod production and variation through either (i) seed within the original seed lot, (ii) seed or plant introduced unintentionally, or (iii) a mutation, is difficult. The truest to type available of 'Wintergreen' had been planted since 1998 or 1999 in a commercial nursery block at the breeders Windsor farm, New South Wales, Australia (P McMaugh 2006, pers. comm., 26 May). The latter sod farm was sold in July 2005.

Hubert F. Whiting was not involved in the collection and/or breeding work of Wintergreen as listed in the US Patent (McMaugh and Whiting, 1988). McMaugh states that Whiting was given shared patent rights of the cultivar in America in the USA (P McMaugh 2009, pers. comm., 11 November).

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Appendix II: Additional Morphological-Agronomic Data

This appendix contains informative data relevant to Chapters 2 to 4 of the thesis.

Leaf and Stolon Colour

RHS colour observations were recorded at the following times: experiment 1, 29 October 2002 (spring); experiment 3, 16 March 2004 (autumn) and experiment 4, 1 December 2004 (summer). Collected data is shown for the *C. dactylon* varieties assessed in Table II.1 and for the hybrid bermudagrass varieties measured in Table II.2.

Taxon	Variety	Stolon Colour	Leaf Colour
C. dactylon	Common	N199A (Exp.1)	137B (Exp.1)
(bermudagrass)	Hardi Turf	N199A (Exp.1)	137B (Exp.1)
	FLoraTeX	N199A (Exp.1)	137B (Exp.1)
	Conquest	N199A (Exp.1)	137A (Exp.1)
	Wintergreen	N199A (Exp.1), 200C (Exp.4)	137B (Exp.1&4)
	Windsor Green	N199A (Exp.1)	137B (Exp.1)
	Hatfield	N199A (Exp.1)	137B (Exp.1)
	SS2	N199A (Exp.1)	137B (Exp.1)
	Legend	N199A (Exp.1)	137B (Exp.1)
	CT-2	N187A & N77A (Exp.1)	147A (Exp.1)
	Mountain Green	N199A (Exp.1&3)	147A (Exp.1&3)
	Oz Tuff	N199B (Exp.3)	138B (Exp.3)
	Plateau	199B (Exp.1), N199B (Exp.3)	147A (Exp.1&3)
	Riley's Super	N199A	146A (Exp.1),
	Sport	(Exp.1&3)	N138B (Exp.3)
	Winter Gem	146B (Exp.4)	137B (Exp.4)

Table II.1 Visual observations of stolon and leaf colour of *Cynodon dactylon* varieties using theRHS colour chart.

Note: Data was collected during spring for Experiment 1, whereas Experiments 3 and 4 were collected in summer.

Table II.2 Visual observations of stolon and leaf colour of hybrid bermudagrass greens quality varieties using the RHS colour chart.

Taxon	Variety	Stolon Colour	Leaf Colour
C. dactylon x C.	Novotek	N1991 (Exp.1)	147A (Exp.1)
	Tifgreen	N1991 (Exp.1)	146A (Exp.1)
transvaalensis	Tifdwarf	N1991 (Exp.1)	137A (Exp.1)
(hybrid	TifEagle	N1991 (Exp.1)	>137A (Exp.1)
bermudagrass)	Champion Dwarf	N1991 (Exp.1)	137B (Exp.1)
	MS-Supreme	1991 (Exp.1)	137B (Exp.1)
	FloraDwarf	N1991 (Exp.1)	137A (Exp.1)

Note: Data was collected during spring.

Lateral Spread Measurements

Table II.3 identifies the dates where cumulative spread measurements (four measurements per plant)

were collected of the hybrid bermudagrass and bermudagrass varieties in experiments 1 to 5.

Table II.3 Diameter of spread measurements were collected over a number of test dates within spaced plant experiments 1 to 5 as identified. However, only the data collected on the final date was utilised within the present study.

Exp.	C. dactylon x C. transvaalensis (hybrid bermudagrass)	<i>C. dactylon</i> (bermudagrass)
1	8 Aug, 23 Aug, 5 Sep and 19 Sep 2012	8 Aug, 23 Aug, 5 Sep and 19 Sep 2012
2	8 Apr, 29 Apr, 19 May, 3 Jun, 17 Jun, 8 Jul, 22 Jul, 5 Aug and 21 Aug 2003	8 Apr, 29 Apr, 19 May, 3 Jun and 17 Jun
3	23 Oct, 6 Nov and 21 Nov 2003	23 Oct, 6 Nov and 21 Nov 2003
4	NA	10 Aug, 24 Aug, 7 Sep, 21 Sep, 5 Oct, 19 Oct and 2 Nov 2012.
5	18 May, 31 May, 15 Jun, 28 Jun, 14 Jul, 27 Jul, 10 Aug, 24 Aug, 7 Sep, 21 Sep, 5 Oct, 19 Oct and 2 Nov 2012.	NA

Note: NA, not applicable.

Appendix III: Recommended Cynodon Characteristics to be measured by Australia's Plant Breeder's Rights Office

Table III.1 Australia's Plant Breeder's Rights Office (PBRO) technical descriptor listing recommended characteristics to be measured of *Cynodon* spp. varieties under the Australian PBR scheme. The descriptor was updated 2 November 2012 by the PBRO.

Organ/Plant Part	Context	State of Expression
Plant	ploidy	
Plant	habit	
Plant	type	
Plant	height	
Plant	longevity	
Plant	spreading	
Stolon	nodes	
Stolon	internode length	
Stolon	internode thickness	
Stolon	colour when exposed to sunlight	
Culms	length	
Leaf blade	shape	
Leaf blade	length	
Leaf blade	width	
Leaf blade	colour	
Ligule	appearance	
Inflorescence	type	
Inflorescence	length of peduncle	
Inflorescence	maximum number of spikes	
Inflorescence	minimum number of spikes	
Culms	habit	
Leaf sheath	appearance	
Leaf blade	presentation	
Leaf blade	apex	
Inflorescence	anthers	
Plant	reproductive behaviour	

<u>Note</u>: The column 'state of expression' has descriptions entered into each cell by the Qualified Person (QP) conducting the PBR trial.