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journal homepage: www.elsevier.com/locate/sajbPhenology of honeybush (*Cyclopia genistoides* and *C. subternata*) genotypesMlamuli M. Motsa^{a,b,*}, Margaretha M. Slabbert^b, Cecilia Bester^a, Mzabalazo Z. Ngwenya^c^a Cultivar Development Division, Agriculture Research Council Infruitec-Nietvoorbij, Private Bag X5026, Stellenbosch 7599, South Africa^b Department of Horticulture, Tshwane University of Technology, Private Bag X680, Pretoria 0001, South Africa^c Biometry Unit, Agriculture Research Council, Private Bag X8783, Pretoria 0001, South Africa

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ABSTRACT

No information is available on the phenological phases between and within genotypes of *Cyclopia* (honeybush). This information is important to understand the timing of plant development and growth, species co-existence, and growth dynamics of the genus. The study spanning 2 years determined the monthly genotypic variation, the time of the start of a growth phase, and duration of budding, flowering, fruiting, and seed dispersal in *C. subternata* and *C. genistoides*. The results indicated differences in phenology between and within species and that initiation period among individuals of a phenological phase is inversely proportional to that duration of that phenophase. Budding, flowering, fruiting, and seed dispersal peaked in the months of July, September, October, and December, respectively. Compared to *C. genistoides*, *C. subternata* genotypes had a shorter time (days) from start of observation to start of flowering (25.9 versus 51.2), fruiting (46.1 versus 61.5), and seed dispersal (96.3 versus 110.0). However, the duration (days) of flowering (13.2 versus 24.3), fruiting (45.6 versus 52.2), and seed dispersal (4.3 versus 8.9) was shorter in *C. genistoides*. Using observational qualitative analysis, phenology of these *Cyclopia* species was categorised into three groups; early, intermediate, and late. The findings serve as a platform for investigating factors affecting the reproductive phases, morphology, and physiology in these and other *Cyclopia* species. It will assist farmers and researchers to time crop requirements and management practices, thus having practical implications in the cultivation of the species.

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1. Introduction

Phenology is the study of the timing of plant growth phases that are biologically important, including the causes of timing and interrelation between growth phases of the same or different species (Ruml and Vulić, 2005). Phenological research assists in the monitoring of plant developmental stages during the growing season, understanding species interactions, and making comparable observations (Fenner, 1998; Saska and Kuzovkina, 2010). Apart from phenology being important in agricultural practices (Zavalloni et al., 2006; Saska and Kuzovkina, 2010), these growth phases may determine a species' ability to establish and persist in favourable and avoid unfavourable climatic conditions, thus shaping their distribution (Chuine, 2010; Van Der Putten et al., 2010; Pau et al., 2011; Gratani, 2014). Furthermore, phenology helps differentiate populations from different altitudes and latitudes (Vitasse et al., 2009), which may lead to variations in the morphological traits of widely distributed plant species, thus indicating survival ability and resource acquisition in heterogeneous and variable

environments (Zhuang et al., 2011; Gratani, 2014). Species with wider geographical ranges can therefore exhibit a larger intraspecific variation in morphology, physiology, phenology, and growth rate (Gratani, 2014). In most cases, the timing of each phenological phase (phenophase) is regulated by mechanisms which act to ensure that each phase occurs in suitable conditions in its own period, although there is some interdependence between them. Phenological studies may include observation, recording, and interpretation of the timing of plants life history events (Fenner, 1998; Tooke and Battey, 2010; León-Ruiz et al., 2011).

Presently, information on the phenology of *Cyclopia* is scarce. *Cyclopia*, also known as "honeybush", is a largely unstudied leguminous genus of South African herbal teas restricted to the megadiverse Cape Floristic Region (Joubert et al., 2011). Many *Cyclopia* species (e.g. *C. genistoides*, *C. intermedia*, and *C. subternata*) flower in spring (September–October), although some species e.g. *C. sessiflora* flower in winter (May–June) (Van Der Walt, 2000; Stepanova et al., 2012). However, to date, no research has been conducted on the comparative phenology between and within *Cyclopia* species. Schröder et al. (2014) specified that plant phenophases can display inter-annual variability and large spatial differences attuned to environmental cues, individual characteristics such as genes, age, soil conditions, water supply, diseases, and competition. Phenological studies in ecosystems such as the Mediterranean climate of the Fynbos biome (Berjano et al., 2006) where *Cyclopia* naturally grows are important in order to identify

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phenological patterns (timing, frequency, and duration) within different areas. This paper assesses the phenophases of two commercially important *Cyclopia* species, namely, *C. genistoides* and *C. subternata*. The objective of the study was to determine the phenophases and the differences between and within these two *Cyclopia* genotypes in the Agriculture Research Council Infruitec-Nietvoorbij genebank collection. An understanding of the phenology of the two species will thus enhance farmers' ability to plan management practices in relation to the events occurring in the plant in order to schedule for pollination, irrigation, fertilisation, crop protection, harvesting, and other cultural practices at optimum times. In addition, this will assist in identifying suitable areas for commercial seed production purposes.

2. Materials and methods

2.1. Study populations

The study was carried out on 2-year-old plants, at Elsenburg Research Farm, situated in the Stellenbosch area of the Western Cape (S 33.502687, E 018.504760). Twenty-two *C. subternata* genotypes were replicated 60 times in a completely randomised block design, while 15 *C. genistoides* genotypes were replicated 90 times. However, mortality of cuttings resulted in a usable study population of approximately 651 and 156 individuals, respectively.

Clonal genotypes were selected and identified based on the area of collection (Fig. 1). *C. genistoides* genotypes were prefixed "G" denoting the species name, whereas *C. subternata* genotypes were prefixed "S". The subsequent alphabetical letter represented the site at which the genotype was initially collected before being rooted. *Cyclopia genistoides* genotypes were collected from Gouriquau (GG) near Gouritsmond, Koksrivier (GK) near Pearly Beach and Toekomst (GT) near Bredasdorp. *Cyclopia subternata* genotypes were collected from Tolbos (STB) near Napier, Kanetberg (SKB) near Barrydale, Groendal (SGD) near Loutewater, and Haarlem (SHL). Rooted cuttings of 12 months old were planted during 2011 in a sandy loam soil at a spacing for both species of 1 × 1 m (within rows and between rows). Both species were irrigated using drip irrigation in the dry summer seasons. No fertilisation or disease and pest control was applied since *Cyclopia*

species are most frequently organically grown, and weeds were manually kept at minimal levels.

2.2. Sampling

Five randomly selected plants of each genotype were sampled. Where the number of plants was limited, other plants were sampled as in the case of STB101 (1), GG3 (3), GG34 (4), GK8 (2), and GG9 (1). To distinguish between plants and ensure correct data recording, sampled plants were marked with chevron tape. Data comprised weekly observation of plants using visual estimates of phenophases. Observations were scored on a 0–100 scale with 10 increments. The criterion applied in all the phenophases was a threshold value of 10% per individual plant modified from Pudas et al. (2008), where a 10% budding and 90% flowering meant that 10% of the buds on the individual plant had developed into flowers (90%) by the observation date. Data were collected in 2013 and 2014. The different stages within a phenophase were not recorded. Data from local weather stations' were used to describe the climate near the observed species, for both years and also that of the provenances where clones were initially collected (Table 1). Four phenophases were observed: budding, flowering, fruiting (pod formation), and seed dispersal (harvesting).

2.2.1. Budding phase

The budding phase was determined when buds visually appeared (Fig. 2A). Bud set was followed by the bud development phase characterised by swelling of buds (Fig. 2B) before bud maturation and where the bud was completely opened with petals still clustered together (Fig. 2C). The time to start of budding was not calculated because the study started after buds had already formed, especially in early genotypes. However, the duration of budding from first day of observation until first flowers appeared could be quantified.

2.2.2. Flowering phase

Flowering was defined as the stage when the reproductive parts (male stamen and female pistil) were visible between unfolded or open flower parts. This stage marked the period in which pollinators would be attracted, leading to pollen dispersal and thus pollination (Fig. 3A–C).

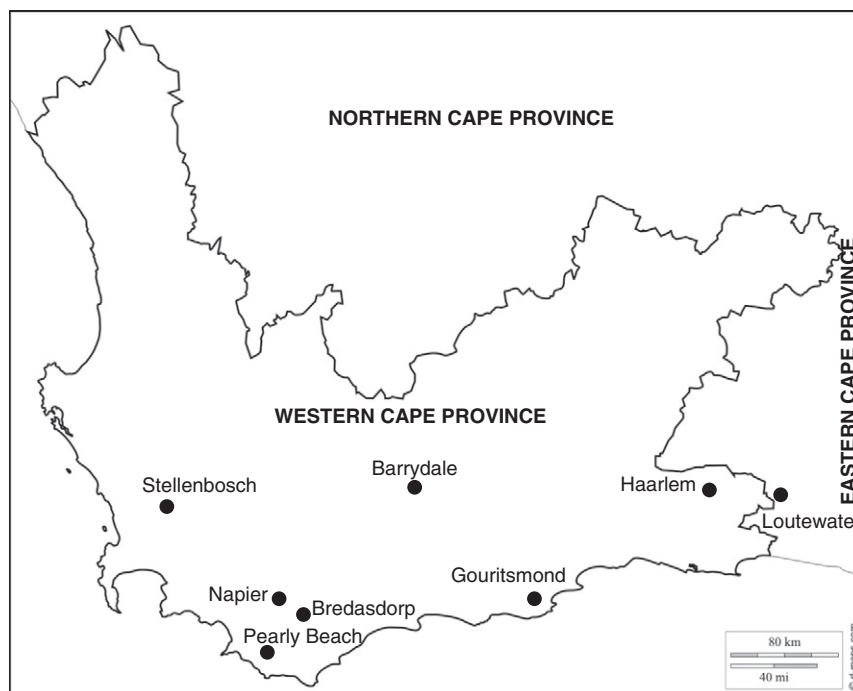


Fig. 1. Map of the geographical positions of the different *Cyclopia* species genotypes where they were initially collected before being grown in Elsenburg, Stellenbosch area, South Africa.

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