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Review

Ethnobotanical Uses, Nutritional Composition, Phytochemicals, Biological Activities, and Propagation of the Genus *Brachystelma* (Apocynaceae)

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Abstract: The *Brachystelma* genus (family: Apocynaceae) consists of geophytes that are traditionally utilised among rural communities, especially in East Africa, southern Africa, West Africa, and northern and western India. Apart from being used as a food source, they are indicated as treatment for ailments such as colds, chest pains, and wounds. This review provides a critical appraisal on the ethnobotanical uses, nutritional value, phytochemical profiles, and biological activities of *Brachystelma* species. In addition, we assessed the potential of micropropagation as a means of ensuring the sustainability of *Brachystelma* species. An inventory of 34 *Brachystelma* species was reported as a source of wild food and traditional medicine (e.g., respiratory-related conditions, pains, and inflammation) across 13 countries, predominantly in Africa and Asia. *Brachystelma circinnatum* and *Brachystelma foetidum* were the most popular plants based on the high number of citations. Limited data for the nutritional content was only available for *Brachystelma edulis* and *Brachystelma naorojii*, as well as phytochemical profiles (based on qualitative and quantitative techniques) for five *Brachystelma* species. Likewise, a few *Brachystelma* species have evidence of biological activities such as antimicrobial, antioxidant, and acetyl cholinesterase (AChE) inhibitory effects. Extensive studies on *Brachystelma togoense* have resulted in the isolation of four compounds with therapeutic potential for managing different health conditions. As a means of contributing to the sustainability of *Brachystelma* species, micropropagation protocols have been devised for *Brachystelma glabrum*, *Brachystelma pygmaeum*, *Brachystelma ngomense*, and *Brachystelma pulchellum*. Nevertheless, continuous optimisation is required to enhance the efficiency of the micropropagation protocols for these aforementioned *Brachystelma* species. Despite the large number of *Brachystelma* with anecdotal evidence as food and medicine, a significant number currently lack empirical data on their nutritional and phytochemical profiles, as well as their biological activities. The need for new propagation protocols to mitigate the declining wild populations and ensure their sustainability remains pertinent. This is important should the potential of *Brachystelma* species as novel food and medicinal products be achieved.

Keywords: Apocynaceae; conservation; ethnobotany; food crop; micropropagation; phenolics

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1. Introduction

The genus *Brachystelma* R. Br. ex Sims is a member of the Ceropegieae tribe (sub-family Asclepiadoideae and family Apocynaceae) [1,2]. In tropical and arid regions, especially in Africa, Asia, and Australia [3–7], an estimated 141 members of the genus have been reported. As one of the centres of endemism, South Africa has a significant number of *Brachystelma* species (Figure 1), where they occur across all the nine provinces [8]. In terms of morphology, *Brachystelma* species are often geophytes; however, a few species have fusiform roots rather than a tuber [4,7,9]. Above ground, *Brachystelma* species are small and herbaceous, and often form a cluster of deciduous stem with well-developed leaves [4]. The flowers are small and short-lived, and are often an extremely small corona—the name *Brachystelma* translates to "short crown" [4,8,10].

Members of the Apocynaceae family are generally known to be one of the most dominant plant families in African traditional medicine [8,11] and as food plants in southern Africa [8,12]. Furthermore, many *Brachystelma* species are regarded as a source of valuable therapeutic compounds for the treatment of diverse health conditions and general well-being [13,14]. Particularly, *Brachystelma* are used among local communities for several ailments [10,15]. When compared to *Ceropegia* L., members of the *Brachystelma* genus have limited evidence in terms of their nutritional value, phytochemicals, and biological activities [8]. This is often attributed to their small size and inconspicuous nature as well as their general rarity. Many *Brachystelma* species are visible above ground, predominantly during the rainy season [2,6,10].

Currently, there is a paucity of information regarding the propagation and cultivation of members of the *Brachystelma* genus despite the continuous strains on the wild population in many countries where they are known to occur [2]. Furthermore, many *Brachystelma* species remain threatened in nature because of their slow growth and the encroachment on their natural habitats by external factors [2,8,16–20]. In this review, we provide a critical appraisal of the ethnobotanical uses, nutritional contents, phytochemical profiles, biological activities, and micropropagation endeavours for members of the *Brachystelma* genus. The presence of consolidated information will provide a platform that will help with the judicious utilisation of this valuable genus and evidence-based approaches for future research directions toward its conservation and sustainability.

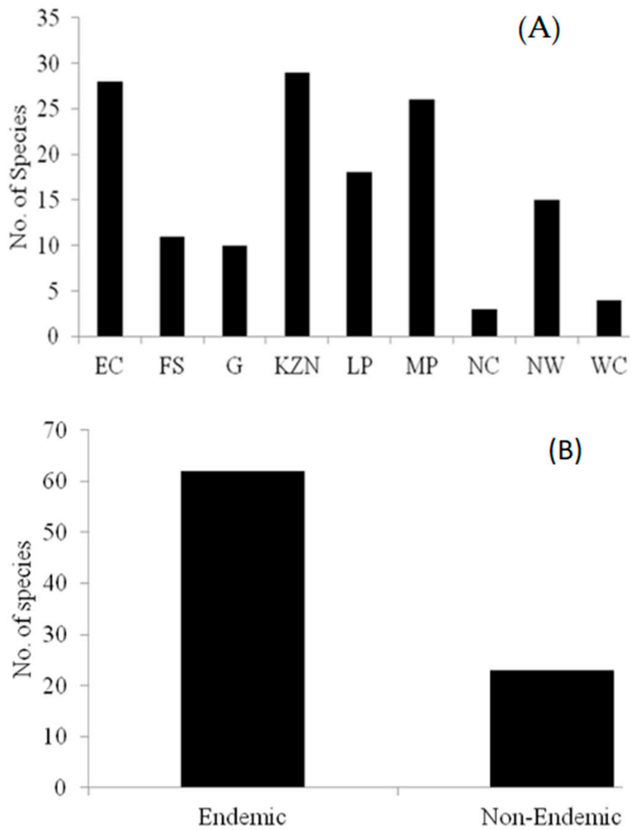

Figure 1. Overview of *Brachystelma* species diversity in South Africa. (**A**) Distribution across different provinces; (**B**) endemic versus non-endemic species based on the South African National Biodiversity Institute (SANBI) Red List data [21]. EC—Eastern Cape, FS—Free State, G—Gauteng, KZN—KwaZulu-Natal, LP—Limpopo, MP—Mpumalanga, NC—Northern Cape, NW—North West, WC—Western Cape.

2. Methods

We explored different online sources such as Google Scholar, Science Direct, and PubMed to gather relevant literature, with an emphasis on peer-reviewed articles. In addition, dissertations/theses that are accessible online were assessed for eligibility. Some of the keywords/phrases used included "*Brachystelma*", "biological activities", "phytochemicals", "conservation" and "ethnobotanical uses". These aforementioned keywords/phrases were used individually and in various combinations to expand the number of hits. No time limit was added but the last search period was on 18 December 2021.

Given the significance of accurate scientific names [22], we cross-referenced the plant names for the different *Brachystelma* species with recognised databases such as The Plant List (http://www.theplantlist.org/ accessed on 18 January 2022) and World Flora Online (http://www.worldfloraonline.org/ accessed on 18 January 2022). A few examples of the synonyms associated with *Brachystelma* species were *Raphionacme splendens* subsp. *bingeri* (A. Chev. Venter), *Tenaris schultzei* (Schltr. E. Phillips), and *Brachystelma magicum*.

3. Distribution and Botanical Description of *Brachystelma* Species

3.1. Distribution of Brachystelma Species

In the Ceropegieae tribe, *Brachystelma* is regarded as the second largest genus [1,2]. It is distributed, in order of decreasing diversity, in sub-Saharan Africa, India, Southeast Asia, and Australia [8]. As indicated by Masinde [10], about 90% of *Brachystelma* species occur exclusively in sub-Saharan Africa. In South Africa, *Brachystelma* species occur across the country i.e., in all nine provinces. Relative to the 85 *Brachystelma* species in the South African Red List data, approximately 73% are endemic to South Africa (Figure 1; Supplementary Table S1). In India, *Brachystelma* species are still being discovered and identified [20,23,24], which is an indication of their expanding diversity.

Evident from their distribution, *Brachystelma* species are variable in habitat preference. Some are found on mountain slopes with up to 3000 mm of annual precipitation, whereas others are found among rocks in grasslands with up to 200 mm of annual precipitation [10,20,23]. However, a common factor is the requirement of well-drained soils [10]. The grassland habitat makes them difficult to locate among the grasses. Furthermore, their visibility is restricted because they are geophytic and generally appear above ground during the rainy season [6,25,26].

3.2. Overview of Botanical Aspects of Brachystelma Species

Morphologically, members of the *Brachystelma* genus are relatively uniform in terms of their vegetative parts (Figure 2A–C). They are considered small perennial geophytic herbs arising from depressed globose or discoid tubers with fibrous roots originating from the sides and the base [10,27]. The tubers range from 15 to 200 mm in diameter, and the tuber size is said to correlate to the age of the plant [9,10]. From the rootstock arises one or generally a cluster of deciduous stem (Figure 2B). Their leaves are simple opposite, with or without stalks, and may be extremely variable in shape, even on a single plant [4,10,15]. Unlike the majority of its milky sap-producing Apocynaceae members, *Brachystelma* leaves are known to produce a clear sap [10]. The leaves are always non-succulent, and usually finely haired [4]. The flowers are inconspicuous, especially in a field among other vegetation [4,15]. They are often flat to cylindrical, come in one to several, and are found together on the side of the stem or rarely terminal, with short to medium-length stalks.

Their corolla is rotate to tubular, divided to halfway or further down the base with five lobes (star-like), and free at the tips and variously spreading, or united at the tips, forming a cage-like structure [4,15]. The flowers are generally dark (brown to maroon, or pinkish purple) and accompanied by a slightly unpleasant odour, which is the primary attraction of pollinators (true flies) [4,5,9,15].

Figure 2. Morphology of some *Brachystelma* R. Br. ex Sims species. (**A**) Flowers (left to right—*Brachystelma gerrardii*; *Brachystelma pulchellum*; *Brachystelma pygmaeum* subsp. *flavidum*; *Brachystelma pygmaeum* subsp. *pygmaeum*); (**B**) leaves (left to right—*Brachystelma gerrardii*; *Brachystelma ngomense*; *Brachystelma pulchellum*; *Brachystelma pygmaeum* subsp. *flavidum*); (**C**) tubers and roots (*Brachystelma pygmaeum* subsp. *pygmaeum*). © Adam Shuttleworth.

For most *Brachystelma species,* the pollinia are usually ovoid and are more or less D-shaped, sub-erect to erect, and near or on top of the staminal column [8,10,15]. Generally, the fruits are erect, long, or short in fusiform pairs. Once dry, the fruit (now follicle) contains the seeds [3,9]. The seeds are dark brown or light black and the size range is 5–10 mm long by 1.5–7 mm wide [8]. One end of the seed has a coma comprising a tuft of fine, white, fluffy hairs up to 20 mm long, which allow for wind dispersal [10,15,27].

3.3. Taxonomy of Brachystelma Species

The importance of the accurate naming of plant species cannot be over-emphasised [22]. A few inconsistencies were evident in the naming of some of the *Brachystelma* species reported in the eligible literature included in the current review (Table 1). Some of the observations included the incorrect spelling and use of synonyms for a few *Brachystelma* species. This clearly supports the call to make reference to reference databases for the validation of plant names.

4. Ethnobotanical Applications of *Brachystelma* Species

Many *Brachystelma* species are known by diverse names among different ethnic groups (Table 1), which is an indication of their value and importance. The tubers are more widely utilised than other plant parts such as the leaves, roots, and stem. This is an indication that the tubers are highly valued in ethnobotany given their extensive utilisation across many countries. The ethnobotanical uses of approximately 34 *Brachystelma* species have been documented in 13 countries (e.g., Australia, Botswana, Burkina Faso, India, Lesotho, Namibia, Nigeria, South Africa, Tanzania, and Thailand) across the world (Table 1). Based on the number of citations, ranging from three to six mentions, *Brachystelma barberiae*, *Brachystelma dinteri*, *Brachystelma circinnatum*, and *Brachystelma foetidum* were identified as the most popular species. Even though the majority of the *Brachystelma* species common in Africa have diverse uses, limited applications were indicated for *Brachystelma* species (e.g., *Brachystelma glabriflorum* and *Brachystelma kerrii*) found in Asian countries such as Cambodia and Thailand [28–30].

In some cases, local uses were identified on a regional basis, such as for *Brachystelma johnstonii* in East Africa [10] and *Brachystelma circinnatum* in southern Africa [31–36]. Generally, the diverse uses recorded for the 34 *Brachystelma* species can be categorised into two major areas, as highlighted below.

4.1. Uses as Food and for Nutritional Needs

The underground parts (particularly the tubers) of many *Brachystelma* species are known to retain a large amount of water and nutrients, thereby making them highly desirable as a food source. The tubers of several *Brachystelma* species (e.g., *Brachystelma burchellii*, *Brachystelma dinteri*, *Brachystelma schultzei*, *Brachystelma thunbergii*, and *Brachystelma tuberosum*) are eaten raw or prepared by indigenous groups in Africa [10,34,37,38], Asia [17,27], and Australia [39]. This is particularly important when there is limited food supply or during hunting expeditions. In East Africa (Kambaland) and South Africa, some *Brachystelma* species (e.g., *Brachystelma gracile* and *Brachystelma johnstonii*) are reported to be particularly enjoyed by livestock herders [10,15]. In Lesotho, the local name '*Bohobe-ba-setsomi*' loosely translates to "bread of the hunters", clearly denoting the value and importance of *Brachystelma* species [33]. Other food-related applications for *Brachystelma* species include as vegetables, snacks, and sweet preserves (Table 1).

Given the high water content, the tubers of *Brachystelma* species are often considered an effective resource for quenching thirst in dry areas where fresh water is scarce [15,36,40]. In Burkina Faso, roots and tubers of *Brachystelma bingeri* are commonly used as a thirst quencher among the nomadic and hunting populations located in Nayala province [40]. Masinde [10] indicated similar uses for *Brachystelma simplex*, *Brachystelma rubellum*, and *Brachystelma plocamoides* in East Africa. The moisture content for the tubers is often high, reaching 80–97% (Table 2), which justifies their uses as thirst quenchers among many indigenous groups.

Some wild animals are believed to harvest and feed on *Brachystelma* tubers [10]. *Brachystelma* species are explored for their nutritional value by animals. On the basis of causal observations [41], animals such as porcupines, mole rats, and blesmols, as well as baboons, are often the major competitors for all underground storage organs, including the deeply buried ones. Based on available anecdotal data, there are no reports of any *Brachystelma* species being poisonous to humans or livestock [10].

4.2. Applications as Herbal Medicine

Some *Brachystelma* species have also been reported as medicinal herbs across several countries (Table 1). The East African species *Brachystelma buchananii* and *Brachystelma johnstonii* are regarded as medicinal herbs with diverse healing properties [10]. For instance, the tuber of *Brachystelma buchananii* is chewed and thereafter, the paste is placed on a wound. In northern Uganda, the tuber of *Brachystelma johnstonii* is dried, ground, and taken to relieve chest pains [10]. Evidence of the medicinal uses has been reported for

Brachystelma foetidum in Lesotho [37,42] and *Brachystelma togoense* in Nigeria [43,44]. In India, *Brachystelma edulis* is used to exert therapeutic benefits for a wide range of health conditions, including headaches and stomach-aches [45,46]. The medicinal effect of other species, including *Brachystelma kerrii*, has been recorded in Cambodia and Thailand [29,30].

Based on the number of mentions, *Brachystelma edulis*, *Brachystelma naorojii*, and *Brachystelma togoense* were identified as species with diverse medicinal applications (Table 1). Consensus on the uses of medicinal plants is an indication of their potential therapeutic values [47]. Some degree of similarity in uses was demonstrated with the application of *Brachystelma* for managing respiratory-related conditions such as colds, coughs, and chest pain in three countries, including China, India, and Lesotho (Table 1). However, different *Brachystelma* species were utilised for cold in these aforementioned countries. These include *Brachystelma edule* (China), *Brachystelma naorojii* (India), and *Brachystelma foetidum* (Lesotho).

Table 1. Distribution of *Brachystelma* species with their documented ethnobotanical uses. #Species = names provided in brackets are either as spelt in the original article or synonyms; ns = not specified.

#Species	Country/Region	Local Name(s)	Plant Part(s)	Uses	References
Brachystelma arnottii Baker (*Brachystelma arnotii*)	southern Africa	ns	Tuber	Snack, vegetable	[12,32]
Brachystelma barberiae Harv. ex Hook.f.	Botswana	ns	Tuber	Eaten raw, sometimes roasted	[48]
Brachystelma barberiae Harv. ex Hook.f. (*Brachystelma barberae*)	southern Africa	ns	Tuber	Snack, vegetable	[12,49]
Brachystelma bingeri A. Chev. (Synonym: *Raphionacme splendens* subsp. *bingeri* (A.Chev.) Venter)	Burkina Faso	*Sensenega*, *Daffio*	Roots, tuber	Appetite suppressant and thirst quencher	[40,50]
Brachystelma buchananii N.E.Br.(Synonym: *Brachystelma magicum*)	Tanzania	ns	Tuber	Wound healing, magic power	[10]
Brachystelma burchellii (Decne.) Peckover (Synonym: *Macropetalum burchellii* Decne.)	southern Africa	ns	Tuber	Wild food	[31,34]
Brachystelma circinnatum E.Mey (*Brachystelma circinatum*)	Lesotho	*Bohobe-ba-setsomi*	Tuber	Food for hunters	[33,37]
Brachystelma circinnatum E.Mey. (= *Brachystelma filiforme* Harv.)	southern Africa	ns	Tuber	Snack, vegetable, sweet preserve, wild food	[31–36]
Brachystelma cupulatum R.A.Dyer	Namibia	ns	Tuber	Snack, vegetable	[32]
Brachystelma dinteri Schltr.	southern Africa	ns	Tuber	Snack, vegetable	[31,32,35]
Brachystelma discoideum R.A.Dyer	Namibia	ns	Tuber	Snack, vegetable	[32]
Brachystelma edule Collett & Hemsl.	China	ns	Tuber	Wild food, cough, and reducing phlegm	[45]

Table 1. *Cont.*

#Species	Country/Region	Local Name(s)	Plant Part(s)	Uses	References
Brachystelma edulis Coll. and Helmls	India	*Galya, Hanuman batata*	Tuber	Famine food, vegetable, decoction used for bodily discomfort, cough and cold, stomachache, headache, dysentery, enhancing fertility, applied on skin inflammation	[45,46]
Brachystelma foetidum Schltr.	Lesotho	*Seru*	ns	For colds in children	[37,42]
Brachystelma foetidum Schltr.	southern Africa	ns	Tuber	Snack, vegetable, meal, yeast	[31,33,34,37]
Brachystelma gerrardii Harv.	Swaziland (Kingdom of Eswatini)	*Sidzendza*	Tuber	Wild food	[38,51]
Brachystelma glabriflorum (F.Muell) Schltr.	Australia	*Badju, Djalwak*	Tuber	Wild food	[39]
Brachystelma glabriflorum (F.Muell) Schltr.	Thailand	ns	ns	ns	[28]
Brachystelma glabrum Hook. f.	India	ns	Tuber	Wild food	[52]
Brachystelma gracile E.A.Bruce	East Africa	ns	Tuber	Food, thirst quencher	[10]
Brachystelma gymnopodum (Schltr.) Bruyns (Synonym: *Ceropegia pygmaea* Schinz)	southern Africa	ns	Tuber	Snack, vegetable, wild food	[31,35]
Brachystelma johnstonii N.E.Br.	East Africa	*Akurukuri, Naporokenyen*	Tuber	Food, thirst quencher, medicine for chest pain	[10]
Brachystelma kerrii Craib	Cambodia, Thailand	ns	Whole plant	Medicine: bodily discomfort	[29,30]
Brachystelma laevigatum Hook.f.	India	ns	Whole plant	Attractive foliage	[17]
Brachystelma mahajanii Kambale & S. R. Yadav	India	ns	ns	ns	[23]
Brachystelma nallamalayana sp. nov	India	ns	Tuber	Food	[24]
Brachystelma naorojii P.Tetali & al.	India	ns	Tuber	Food and medicine: stomach-ache, cough and colds	[45]
Brachystelma pauciflorum Duthie	India	ns	Tuber	Wild food	[17]
Brachystelma plocamoides Oliv.	East Africa	ns	Tuber	Food, thirst quencher	[10]
Brachystelma pullaiahi Rao et al.	India	*Nakshtralamokka, Nemithigadda*	Tuber	Wild food (eaten raw)	[53]
Brachystelma rubellum (E.Mey.) Peckover	East Africa	*Mkumbe, Muthunga*	Tuber	Food, thirst quencher	[10]

Table 1. *Cont.*

#Species	Country/Region	Local Name(s)	Plant Part(s)	Uses	References
Brachystelma schultzei (Schltr.) Bruyns (Synonym: *Tenaris schultzei* (Schltr.) E.Phillips)	southern Africa	ns	Tuber	Wild food	[31,36]
Brachystelma simplex Schltr.	East Africa	ns	Tuber	Food, thirst quencher	[10]
Brachystelma thunbergii N.E.Br (currently regarded as ambiguous)	South Africa	*Kamb(a)roo*	Tuber	Snack, vegetable, meal, yeast, sweet preserve	[34,41]
Brachystelma togoense Schltr.	Nigeria	ns	Whole plant	Medicine: dysentery, cough and cold, wounds, stomachache, typhoid, erectile dysfunction	[43,44]
Brachystelma tuberosum (Meerb.) R.Br. ex Sims	southern Africa	ns	Tuber	Wild food	[31]
Brachystelma vartakii Kambale & S. R. Yadav	India	ns	ns	ns	[23]
Brachystelma volubile Hook.f.	India	*Telugu*	Tuber	Wild food	[20,52]

5. Nutritional and Phytochemical Aspects

5.1. Nutritional Composition of Brachystelma Species

Despite the relatively large number of *Brachystelma* species with an ethobotanical record as wild food (Table 1), only *Brachystelma edulis* and *Brachystelma naorojii* currently have empirical data on their nutritional content (Table 2). Nutritional analysis for wild edible plants that serve as food is essential to justify their incorporation into the diet [12,41,54,55]. Based on the study conducted by Deshmukh and Rathod [46], the tubers of *Brachystelma edulis* could be a good supplement for nutrients such as protein, fibre, and carbohydrates. The recorded amount for these aforementioned nutrients is within the range for common foods, especially vegetables [56]. Particularly, the level of carbohydrates among *Brachystelma* species was comparable to African leafy vegetables such as *Bidens pilosa*, *Chenopodium album*, and *Portulaca oleracea* [57]. The fibre content in *Brachystelma edulis* (8%) tubers compared favourably to *Ceropegia hirsuta* (9.1%), a closely-related species [46]. A similar trend was observed with regards to the crude protein level between these two aforementioned plants.

For some of the content (e.g., ash and moisture), comparable levels were present in the leaves and tubers, which is an indication of plant-part substitution with respect to *Brachystelma edulis* [27,45,46] and *Brachystelma naorojii* [58]. However, in terms of the dry matter (% DW), the tubers were generally higher than the leaves for both *Brachystelma edulis* [45,46] and *Brachystelma naorojii* [58]. Existing evidence demonstrated some degree of variation in parameters, such as ash and dry matter between *Brachystelma edulis* and *Brachystelma naorojii* (Table 2).

Table 2. Proximate and mineral composition of different *Brachystelma* species.

Component	Content
Proximate (*Brachystelma edulis, BE* and *Brachystelma naorojii, BN*)	
Ash (% of fresh weight)	*BE* leaves: 12 [45] *BE* tuber: 11–11.5 [27,46] *BN* leaves: 7.8 [58] *BN* tuber: 6 [58]
Crude fat (% DW)	0.12 [46]

Table 2. *Cont.*

Component	Content
Crude fiber (% DW)	8.0 [46]
Crude protein (g/100 g DW)	3.93 [46]
Dry matter (% FW)	*BE* leaves: 8.61 [45] *BE* tuber: 19.3 [46] *BN* leaves: 5.8 [58] *BN* tuber: 7.17 [58]
Energy value (k cal/100 g DW)	302.4 [46]
Moisture (% FW)	*BE* leaves: 91.4 [45] *BE* tuber: 80.8–97.01 [27,46] *BN* leaves: 94 [58] *BN* tuber: 93 [58]
Total carbohydrates (% DW)	3.61 [46]
Mineral composition (mg/100 g DW)—*Brachystelma edulis* [46]	
Calcium	464.8
Copper	0.94
Iron	40.3
Magnesium	186.7
Manganese	3.27
Phosphorus	143.4
Potassium	416.3
Sodium	9.5
Zinc	1.07

FW = fresh weight, DW = dry weight.

Mineral analysis of *Brachystelma edulis* tubers revealed the presence of 9 elements with high concentrations of potassium, phosphorus, and magnesium [46]. These minerals are essential for maintaining good health and well-being [56,57,59]. Based on the concentrations of these minerals found in *Brachystelma edulis* (Table 2), it certainly competes favourably with many indigenous vegetables (e.g., *Amaranthus spinosus*, *Bidens pilosa*, *Chenopodium album*, and *Momordica involucrata*), which are often sourced by local communities to meet their daily requirements [55]. However, the absence of nutritional and mineral content for the majority of *Brachystelma* species remains a major gap requiring empirical data to support their overwhelming use as a food source among different ethnic groups. Particularly, the potential development of novel food products from indigenous plants, including *Brachystelma* species, will be driven by the availability of basic and in-depth data on their nutritional value and sensory attributes [60].

5.2. Phytochemical Profile of Brachystelma Species

Among the Ceropegieae, *Brachystelma* have not received much attention in terms of the quantification of their phytochemicals. Currently, the phytochemical profiles of five *Brachystelma* species have been investigated using different analytical techniques, including qualitative and quantitative tests (Table 3). Most studies search first for alkaloids, as they are known among the Apocynaceae to possess a range of potent pharmacological activities, including antibacterial, anticancer, and antimalarial activities [13,61]. Phytochemical analysis of *Brachystelma edulis*, based on qualitative tests, indicated the presence of alkaloids, cardiac glycosides, phenols, saponins, and tannins (Table 3). Similarly, *Brachystelma bingeri* contains saponins, triterpenes, and sterols [62]. Furthermore, a spectrophotometric test of *Brachystelma bingeri* revealed an estimated 1.7 mg EGA/g (equivalent of gallic acid) of total phenolics, whereas the total flavonoids and tannins were only found in trace quantities.

Table 3. Phytochemical analysis of *Brachystelma* species.

	***Brachystelma* Species**	**Content**	**Reference**
Qualitative Tests			
Alkaloid	*Brachystelma edulis*	+	[27,45]
	Brachystelma naorojii	+	[58]
Coumarins	*Brachystelma bingeri*	+	[62]
	Brachystelma naorojii	+	[58]
Flavones	*Brachystelma edulis*	+	[45]
Glycosides	*Brachystelma edulis*	+	[27,45]
	Brachystelma naorojii	+	[58]
Phenol	*Brachystelma edulis*	+	[27]
	Brachystelma naorojii	+	[58]
Reducing sugars	*Brachystelma edulis*	+	[45]
	Brachystelma naorojii	+	[58]
Saponosides	*Brachystelma bingeri*	+	[62]
Saponin	*Brachystelma edulis*	+	[27]
Steroids and triterpenes	*Brachystelma bingeri*	+	[62]
Tannins	*Brachystelma edulis*	+	[27,45]
	Brachystelma naorojii	+	[58]
Spectrophotometric Technique			
Total peholic	*Brachystelma bingeri*	1.7 mg EGA/g	[62]
Ultra-High-Performance Liquid Chromatography–Tandem Mass Spectrometry (UHPLC-MS/MS) Analysis			
Caffeic acid	*Brachystelma pulchellum*	0.331–1.476 µg/g	[63]
Caffeic acid	*Brachystelma pygmaeum*	0.445–1.282 µg/g	[63]
Ferulic acid	*Brachystelma pulchellum*	10–55 µg/g	[63]
Ferulic acid	*Brachystelma pygmaeum*	9–43 µg/g	[63]
Gallic acid	*Brachystelma pulchellum*	0.119–0.40 µg/g	[63]
Gallic acid	*Brachystelma pygmaeum*	0.263–0.565 µg/g	[63]
m-Hydroxybenzoic acid	*Brachystelma pulchellum*	0.206–1.167 µg/g	[63]
m-Hydroxybenzoic acid	*Brachystelma pygmaeum*	0.167–1.595 µg/g	[63]
p-Coumaric acid	*Brachystelma pulchellum*	2–7 µg/g	[63]
p-Coumaric acid	*Brachystelma pygmaeum*	1.352–2.783 µg/g	[63]
p-Hydroxybenzoic acid	*Brachystelma pulchellum*	0.496–1.374 µg/g	[63]
p-Hydroxybenzoic acid	*Brachystelma pygmaeum*	0.385–1.060 µg/g	[63]
Protocatechuic acid	*Brachystelma pulchellum*	0.450–1.488 µg/g	[63]
Protocatechuic acid	*Brachystelma pygmaeum*	0.223–0.955 µg/g	[63]
Salicylic acid	*Brachystelma pulchellum*	0.839–2.464 µg/g	[63]
Salicylic acid	*Brachystelma pygmaeum*	0.307–1.086 µg/g	[63]
Sinapic acid	*Brachystelma pulchellum*	14–58 µg/g	[63]
Sinapic acid	*Brachystelma pygmaeum*	30–98 µg/g	[63]
Syringic acid	*Brachystelma pulchellum*	0.672–3.107 µg/g	[63]

Table 3. *Cont.*

	Brachystelma **Species**	**Content**	**Reference**
Syringic acid	*Brachystelma pygmaeum*	0.167–0.910 µg/g	[63]
Vanillic acid	*Brachystelma pulchellum*	0.506–1.804 µg/g	[63]
Vanillic acid	*Brachystelma pygmaeum*	0.663–2.834 µg/g	[63]

EGA = equivalents of gallic acid.

Brachystelma pulchellum and *Brachystelma pygmaeum* also demonstrated the presence of phenolic acids, including both hydroxybenzoic and hydroxcinnamic acids [63]. The phenolic acid profile of in vitro grown *Brachystelma pulchellum* and *Brachystelma pygmaeum* showed similarities in their phenolic profiles, but quantities varied among species. Given the importance of hydroxybenzoic and hydroxcinnamic acids [64,65], their presence in these aforementioned *Brachystelma* species provide evidence on the potential health benefits associated with their consumption.

Some *Brachystelma* species, including *Brachystelma ngomense* and *Brachystelma pulchellum*, have been observed to have a fibrous stem covered by bark that appears red-violet in colour. Even though not well characterised in *Brachystelma* (or in Apocynaceae), red bark has generally been reported to be characteristic of chromoalkaloids (betacyanins) located in the cellular vacuoles of sub-epidermal tissue [66–68]. Certainly for *Brachystelma* species, this is an area that still requires stringent research [8].

6. Biological Activities of *Brachystelma* Species Extracts and Isolated Compounds

Based on indigenous knowledge, several *Brachystelma* species are known for their therapeutic effects for different health conditions, including the treatment of coughs and colds, wounds, and bodily discomfort, and for enhancing fertility (Table 1). The rich phytochemical pools in a few *Brachystelma* species have been established from the existing limited studies (Table 3). The presence of therapeutic phytochemicals are generally known to be responsible for the biological effect of medicinal plants [69]. Despite insufficient data on the biological activities of *Brachystelma* species, the tubers and leaves of a few species have been evaluated for antioxidant and acetylcholinesterase inhibitory effects as well as acute toxicity (Table 4). In terms of the antioxidant effects, four *Brachystelma* species (*Brachystelma bingeri*, *Brachystelma edulis*, *Brachystelma pulchellum*, and *Brachystelma pygmaeum*) have been assessed using diverse assays, including ABTS, DPPH, FRAP, and ORAC methods. The use of different antioxidant assays is beneficial, as it has the potential to effectively reveal all the mechanisms associated with the evaluated plant [70]. However, no noteworthy antioxidant effects were observed among the evaluated *Brachystelma* species (Table 4). Likewise, *Brachystelma bingeri* had moderate (50%) acetylcholine esterase (AChE) inhibition at a tested concentration of 100 µg/mL [62]. Using an in vivo model, a methanol extract of *Brachystelma binger* tubers caused no mortality to the tested mice and was considered safe at a concentration of up to 3000 mg/kg body weight [62].

The biological activity of isolated compounds from the aerial parts of *Brachystelma togoense* has predominantly targeted different microbes (e.g., *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus typhi*, *Staphylococcus pneumoniae*, and *Candida albicans*) as a means of establishing their antimicrobial properties (Table 5). As revealed by Ekalu et al. [43], phaeophytin A had the most promising antimicrobial effect (MIC = 0.09 mg/mL) against *Candida albicans*. Furthermore, the MIC value ranged from 0.18 to 0.37 mg/mL against the four tested bacterial strains (*Escherichia coli*, *Staphylococcus aureus*, *Streptococcus typhi*, and *Staphylococcus pneumoniae*). Relative to the recommended MIC (<100 µg/mL), the antimicrobial property of the majority of compounds isolated from *Brachystelma togoense* are classified as weak to moderate effects, thereby suggesting limited clinical significance [71].

Table 4. Examples of biological activities from different parts of *Brachystelma* species. EAA-equivalents ascorbic acid, TE-Trolox equivalents.

Biological Activity	Species	Plant Part Extraction/Solvent	Method/Assay Positive Control	Key Results	Reference
Acetylcholine esterase (AChE) inhibition	*Brachystelma bingeri*	Tuber methanol	AChE inhibition effect Galanthamine (10 µg/mL, 100%)	Extract (tested at 100 µg/mL) had 50% inhibition of enzyme.	[62]
Acute toxicity	*Brachystelma bingeri*	Tuber methanol	In vivo testing using six (6) mice to determine the lethal dose (LD_{50}) for 72 h	Up to 3000 mg/kg of body weight extracts had no mortality. An indication that the LD_{50} value is greater than 3000 mg/kg of body weight.	[62]
Antioxidant	*Brachystelma bingeri*	Tuber methanol	2,2 diphenyl-1-picrylhydrazyl (DPPH) quercetin (82%)	Extract had low (6%) radical scavenging activity.	[62]
Antioxidant	*Brachystelma bingeri*	Tuber methanol	Ferric-reducing antioxidant power (FRAP) Quercetin (4.69 mmol EAA/g)	Low ferric-reducing power, 0.013 mmol equivalents ascorbic acid per gram of extract (mmol EAA/g extract).	[62]
Antioxidant	*Brachystelma bingeri*	Tuber methanol	2,2′-azinobis—(3-ethylbenzothiazoline-6-sulfonic acid (ABTS) Quercetin (70 mmET/g)	Moderate activity, 25 mmol Trolox equivalents (mmTE)/g of dry extract.	[62]
Antioxidant	*Brachystelma edulis*	Tuber and leaves	Enzyme (peroxidase) activity	A total of 8.43 and 4.07 (unit/min/mg protein) for tubers and leaves, respectively.	[72]
Antioxidant	*Brachystelma edulis*	Tuber and leaves	Enzyme (catalase) activity	A total of 0.3 and 0.39 (unit/min/mg protein) for tubers and leaves, respectively.	[72]
Antioxidant	*Brachystelma edulis*	Tuber and leaves	Enzyme (superoxide dismutase) activity	A total of 0.21 and 0.35 (unit/min/mg protein) for tubers and leaves, respectively.	[72]
Antioxidant	*Brachystelma pulchellum*	Whole plant methanol	Oxygen radical absorbance capacity (ORAC)	Cytokinin-treated in vitro-regenerants had approximately 35–72 µmol/g Trolox equivalents (TE).	[63]
Antioxidant	*Brachystelma pygmaeum*	Whole plant methanol	Oxygen radical absorbance capacity (ORAC)	Cytokinin-treated in vitro regenerants had approximately 45–76 µmol/g Trolox equivalents (TE).	[63]

Table 5. Examples of chemicals isolated from *Brachystelma* species and their biological effects.

Compound	Species	Plant Part	Bioactivity Tested	Reference
(2R, 3R)-dihydroflavonol-3-O-a-L-rhamnoside	*Brachystelma togoense*	Aerial parts	Antimicrobial (significant antimicrobial effects) against *Escherichia coli*, *Streptococcus typhi*, and *Candida albicans*	[44]
1-Methylcyclopentene	*Brachystelma togoense*	Aerial parts	Antimicrobial (noteworthy MIC) against *Escherichia coli* (0.1875 mg/mL) and *Candida albicans* (0.1875 g/mL)	[73]
Lupeol	*Brachystelma togoense*	Aerial parts	Antimicrobial (MIC was 0.18–0.37 mg/mL) against *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus pneumoniae*, *Streptococcus typhi*, and *Candida albicans*	[43,74]
Phaeophytin A	*Brachystelma togoense*	Aerial parts	Antimicrobial (MIC was 0.09–0.18 mg/mL) against *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus pneumoniae*, *Streptococcus typhi*, and *Candida albicans*	[43,74]
α-Amyrin	*Brachystelma togoense*	Aerial parts	Antimicrobial (MIC was 0.37 mg/mL) against *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus pneumoniae*, *Streptococcus typhi*, and *Candida albicans*	[43,74]

7. Conservation Status of *Brachystelma* Species and Sustainability

7.1. Conservation Status of Brachystelma Species

In South Africa, many members (54%) of the genus *Brachystelma* remain of conservation concern (Figure 3; Supplementary Table S1). Furthermore, the threats on members of the *Brachystelma* genus have been reported in India [8]. Findings by Singh [17] revealed that *Brachystelma laevigatum* and *Brachystelma pauciflorum* are currently vulnerable and endangered due to their use by local people as food and for their attractive foliage. According to Rajakullayiswamy et al. [20], *Brachystelma volubile* is currently indicated as being critically endangered. In India, the grasslands in which *Brachystelma* thrive are often considered "wasted and unproductive" when left in the hands of nature and thus left unprotected [16]. In addition, these grasslands are often encroached on for agriculture and fragmented by urbanisation and invasive species.

7.2. Propagation of Brachystelma Species

Natural cultivation of *Brachystelma* species has remained neglected and sparingly documented [2,8]. The most prevalent constraints, with natural propagation of species belonging to the subfamily Asclepiadoideae, are often attributed to the scarcity of pollinators together with low seed set and viability [75–77]. Propagation via conventional methods i.e., cuttings and seedlings, is also either absent or not well-documented among *Brachystelma*. Following pollination, the rate (%) of successful fertilisation varies among species, but is generally low when compared to many bee-pollinated species [15]. The pollination is affected by the deposit of pollinia on the flowers' stigmatic surfaces fused within the top of the staminal column [8,15]. In terms of the arrangement of the male and female reproductive parts, they are combined into a small columnar structure within the base of the corolla, increasing the chances of self-pollination [15]. This inevitably affects the fertilisation due to the failure of producing viable seeds [76,78]. Upon successful fertilisation, fruit development is rapid but takes several weeks for the seeds to mature, and during this time, the fruit may be attacked by pests, thereby preventing seed maturation [15]. On this basis, efficient ex situ conservation measures are of importance for the future of the *Brachystelma* genus, especially when their nutritional and medicinal potential has not been fully assessed [8].

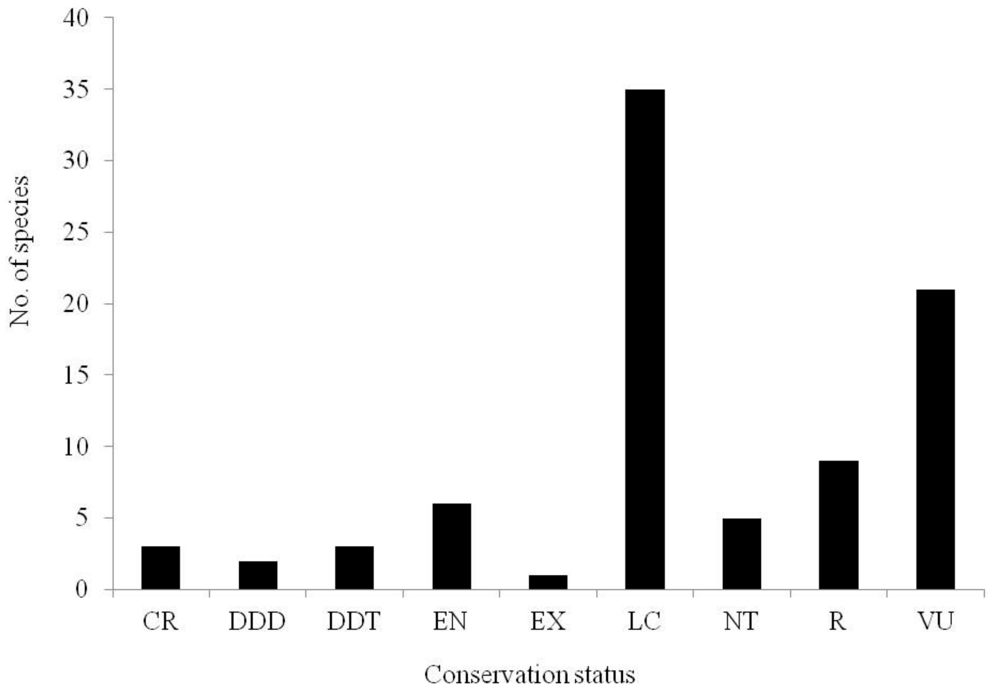

Figure 3. An overview of the conservation status of 85 *Brachystelma* species recorded in South Africa based on the South African National Biodiversity Institute (SANBI) Red List [21]. CR—Critically rare, DDD—Data deficient (insufficient information), DDT—Data deficient (taxonomically problematic), EN—Endangered, EX—Extinct, LC—Least concern, NT—Near threatened, R—Rare, VU—Vulnerable.

7.3. Micropropagation of Brachystelma Species

Globally, plant biotechnology, in particular, micropropagation, has remained valuable based on its beneficial role in ex situ conservation and the mass production of plants and plant-based resources [75,79–83]. Conventional methods alone are not sufficient to achieve optimum yield and production rates, especially in plants such as *Brachystelma*, which require long periods of time for bulb maturation [77,84]. The micropropagation process starts with the selection of appropriate explants till the successful establishment of the in vitro regenerants under ex vitro conditions [83]. The different stages need to be carefully implemented to achieve the overall goal of conserving the selected plant species.

Micropropagation attempts for four *Brachystelma* species have been conducted by different researchers (Table 6). Even though some degree of success was achieved for *Brachystelma glabrum* and *Brachystelma ngomense* [84,85], the regeneration frequencies for *Brachystelma pulchellum* and *Brachystelma pygmaeum* were quite low [84]. Following the application of thidiazuron (TDZ) in various combinations with three auxins (indole-3-acetic acid, IAA; indole-3-butyric acid, IBA; and naphthalene acetic acid, NAA), shoot proliferation of *Brachystelma glabrum* was significantly enhanced with the use of TDZ (1 mg/mL) and NAA (0.5 mg/mL) relative to the control and other treatments [85]. The vital role of cytokinins was evident with the diverse regeneration and proliferation responses recorded among the three *Brachystelma* species [84]. Particularly for shoot proliferation of *Brachystelma ngomense* (a *Brachystelma* species that is categorised as an endangered plant), the use of *meta*-topolin riboside (*m*TR, 25 µM) was the most effective (producing an estimated four in vitro shoots) of the three cytokinins tested. On the other hand, BA (25 µM) and iP (25 µM) were more suitable for the micropropagation of *Brachystelma pygmaeum* and *Brachystelma pulchellum*, respectively. Furthermore, various combinations (types and concentrations) of cytokinins and auxins were applied for the in vitro flowering and tuberisation of *Brachystelma glabrum* [85]. Given the importance of plant growth regulators (especially cytokinins and auxins) in micropropagation protocols [79,81,86,87], it is important to continuously explore how the different classes of plant growth regulators can be used to improve the regeneration of *Brachystelma* species.

The type of explant used for culture initiation often contributes to the success or failure of such protocols as well as the determination of the quantity and quality of the in vitro regenerants [88]. Shoot-tip and nodal parts are the two types of explants currently being explored for the micropropagation of *Brachystelma* species (Table 6). For the micropropagation of *Brachystelma glabrum*, nodal explants were more responsive relative to the shoot tips [85]. As a means of enhancing the success of acclimatisation of in vitro regenerants, approaches that stimulant high rooting and survival percentage as well as ex vitro rooting are crucial final stages of micropropagation protocols [83]. Varying rooting and acclimatisation rates were demonstrated for the four evaluated *Brachystelma* species (Table 6). The limited survival rate, under ex vitro conditions, for the majority of the *Brachystelma* species justifies the need for more research focusing on this important micropropagation step. The use of biostimulants (e.g., seaweed extract) has been indicated as an economic and affordable means to potentially enhance the different stages of micropropagation, especially the acclimatisation phase [89].

Given the high number of factors that affect micropropagation endeavours, further research aimed at optimising the existing protocols for *Brachystelma* species remains pertinent. In particular, somatic embryogenesis is known to be an efficient approach for plant regeneration [90,91], and its exploration for *Brachystelma* species may result in enhanced regeneration and proliferation.

Table 6. In vitro micropropagation approaches for some *Brachystelma* species.

Species	Parameter Tested	Explant	Most Optimal Media Composition	Response	Reference
Brachystelma glabrum	Effect cytokinins (TDZ, BA) and auxins (IBA, NAA) on in vitro tuberisation	Shoot tip and nodal	TDZ + IBA	60% response in the production of aerial tubers	[85]
Brachystelma glabrum	Effect of auxins (IAA, IB, NAA) on in vitro rooting	Shoot tip and nodal	NAA (0.5 mg/L)	80% root induction and an average of 5.3 roots/ micro-shoot	[85]
Brachystelma glabrum	Effect of BA with different auxins (NAA, IBA) on in vitro flowering	Shoot tip and nodal	BA + NAA (2 mg/L)	60% of culture produced flower buds	[85]
Brachystelma glabrum	Effect of different cytokinins (BA, KIN, TDZ) on shoot induction	Shoot tip	TDZ (1 mg/L)	90% shoot induction and an average of 4.7 shoots/ shoot-tip explant	[85]
Brachystelma glabrum	Effect of different cytokinins (BA, KIN, TDZ) on shoot induction	Nodal	TDZ (1 mg/L)	100% shoot induction and an average of 5.5 shoots/ nodal explants	[85]

Table 6. *Cont.*

Species	Parameter Tested	Explant	Most Optimal Media Composition	Response	Reference
Brachystelma glabrum	Effect of TDZ (1 mg/L) with different auxins (IAA, IBA, NAA) on shoot proliferation	Shoot tip and nodal	TDZ + NAA (0.5 mg/L)	85% shoot induction and an average of 10.6 shoots/shoot-tip explant 90% shoot induction and an average of 11.4 shoots/ nodal explants	[85]
Brachystelma ngomense	Effect of auxin (IBA) on ex vitro rooting and acclimatisation	Nodal	100 mg/L IBA pulsing for 3 min	Survival rate: 42% (4 weeks) and 5% (10 weeks)	[84]
Brachystelma ngomense	Effect of cytokinins (BA, iP, *m*TR) on shoot proliferation	Nodal	*m*TR (25 µM)	Average of 4.4 shoots/ nodal explant	[84]
Brachystelma pulchellum	Effect of auxin (IBA) on ex vitro rooting and acclimatisation	Nodal	100 mg/L IBA pulsing for 3 min	Survival rate: 35% (4 weeks) and 0% (10 weeks)	[84]
Brachystelma pulchellum	Effect of cytokinins (BA, iP, *m*TR) on shoot proliferation	Nodal	iP (25 µM)	Average of 2.04 shoots/ nodal explant	[84]
Brachystelma pygmaeum	Effect of auxin (IBA) on ex vitro rooting and acclimatisation	Nodal	100 mg/L IBA pulsing for 3 min	Survival rate: 30% (4 weeks) and 3% (10 weeks)	[84]
Brachystelma pygmaeum	Effect of cytokinins (BA, iP, *m*TR) on shoot proliferation	Nodal	BA (25 µM)	Average of 2.57 shoots/ nodal ex-plant	[84]
Brachystelma pygmaeum	Effect of pulse dipping of IBA (100 mg/L) for different time intervals (3, 12 and 21 min) on ex vitro rooting and acclimatisation	Nodal	Similar response was observed across all treatments.	Survival rate: 5% for the three-time internal, whereas control had no survival	[84]

BA = N6-benzyladenine, IAA = indole-3-acetic acid, IBA = indole-3-butyric acid, iP = isopentenyladenine, KIN = kinetin, TDZ = thidiazuron, *m*TR = *meta*-topolin riboside, NAA = α-naphthalene acetic acid.

8. Conclusions

The current review highlights the ethnobotanical value of several *Brachystelma* species among different ethnic groups. The majority of these species are highly prized for their value as food and medicine. As indicated by Dyer [15], *Brachystelma* tubers might well make the difference between life and death for anyone cut off from their normal food and water supply. Therefore, the establishment of a modern cultivation technique would be useful as a means of the long-term conservation of these potential lifesavers. Currently, there is limited evidence on the biological efficacies of many *Brachystelma* species. Extracts from four *Brachystelma* species have been evaluated for AChE inhibition and antioxidant activities, and a few isolated compounds from *Brachystelma togoense* were assessed for antimicrobial effects. It is important to study their phytochemistry and pharmacological profile to scientifically support and secure the existing ethnobotanical uses. The lack of scientific research on this group is no reflection on their low significance. The scarcity of information could be attributed to the plant size and the areas in which they occur, i.e., in the vicinity of rural areas. Studies conducted on the phytochemistry of *Brachystelma* have only been able to detect the presence of a few important compounds but have not subjected them to rigorous scientific evaluation. Further exploration of the potential of *Brachystelma* species may require the development of rapid and efficient propagation protocols that guarantee the availability of sufficient plant materials and ensure their sustainability.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/horticulturae8020122/s1, Table S1: List of *Brachystelma* species in South Africa based on the South African National Biodiversity Institute (SANBI) Red List (http://redlist.sanbi.org/genus.php?genus=2640, accessed on 18 January 2022). CR—Critically rare, CR PE—Critically Endangered, Possibly Extinct, DDD—Data deficient (insufficient information), DDT—Data deficient (taxonomically problematic), EN—Endangered, EX—Extinct, LC—Least concern, NT—Near threatened, R—Rare, VU—Vulnerable.

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