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Original Research Article

Ethnobotanical, phytochemical and pharmacological properties of Crinum bulbispermum (Burm f) Milne-Redh and Schweick (Amaryllidaceae)

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Abstract

Purpose: *To present an overview of the ethnobotany, phytochemistry and pharmacology of Crinum bulbispermum so as to understand its importance and potential in primary healthcare systems.*
Methods: *A review of the literature was undertaken and an in-depth analysis of previous research on ethnobotany, phytochemistry and pharmacology of C. bulbispermum. Literature sources included papers published in journals, reports from international, regional and national organizations, conference papers, books and theses. Electronic search engines such as Google, Google scholar, publishing sites such as Elsevier, scienceDirect, BioMed Central (BMC), PubMed and other scientific database sites such as ChemSpider, PubChem were used.*
Results: *Crinum bulbispermum is a popular medicinal plant in southern Africa used as remedy for aching joints, rheumatism, kidney or bladder infections, septic sores and wounds. The chemical composition of C. bulbispermum is dominated by various alkaloids and non-alkaloids isolated from the bulbs, flowers, flowering stalks, leaves and roots. Major biological activities demonstrated by C. bulbispermum include antimicrobial, antioxidant, actinociceptive, antiplasmodial activities as well as effects on the central nervous system.*
Conclusion: *The widespread usage of C. bulbispermum as herbal medicine is threatening wild populations, and this calls for conservation strategies and mechanisms for sustainable utilization of the species.*

Keywords: *Actinociceptive, Amaryllidaceae, Antiplasmodial, Crinum bulbispermum, Ethnobotany, Pharmacology, Phytochemistry*

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INTRODUCTION

Crinum bulbispermum (Burm. f.) Milne-Redh. & Schweick. is a deciduous bulbous plant which belongs to the Amaryllidaceae family. *Crinum bulbispermum* was initially described as *Amaryllis bulbispermum* Burm. f. and then mistakenly identified as *Amaryllis longifolia* L. which is now *Cybistetes longifolia* (L.) Milne-Redh. & Schweick. [1-3]. However, Milne-Redhead and Schweickerdt [2] simplified the matter in their revision of the genus *Ammocharis* by suggesting the new combination, *C. bulbispermum*, based on *A. bulbispermum* Burm. [3]. *Crinum bulbispermum* is a perennial herb growing to about 1 m tall, with a very large bulb, 10-11 cm in diameter covered with bases of older leaves and vermiform roots at the base [4]. *Crinum bulbispermum* has many, thick, stout, simple and glaucous green leaves, measuring 78-92 cm long, 6-6.7 cm broad, oblong to linear in shape, acuminate, flat and shining, margin

slightly scabrous and undulate [3]. The peduncle is 50-90 cm long and has an umbel with between 6 to 16 flowers, which are white in colour with a dark red keel or entirely suffused with red; the stamens are declinate, white or suffused with pink and the style deep pink in the upper portion with a light brown stigma [4]. *Crinum bulbispermum* occurs naturally along rivers and streams or in damp depressions in black clay or sandy soils in Lesotho, South Africa and Swaziland [4].

The plant is cultivated throughout the world for its beautiful and trumpet or bell-shaped flowers. Like most medicinal plants in Southern Africa, *C. bulbispermum* is collected from the wild. The unsustainable harvesting of *C. bulbispermum* as herbal medicine and ornamental plant is threatening its continued existence. Although *C. bulbispermum* is widespread in South Africa, its population is declining due to over-exploitation of its bulbs which are sold in the medicinal (muthi) markets in Durban [5,6], Johannesburg [6,7] and Polokwane [8] in South Africa. Raimondo *et al* [9] categorized *C. bulbispermum* as declining in South Africa based on the modified IUCN Red List Categories and Criteria version 3.1 of threatened species [10-12]. According to Victor and Keith [11] and von Staden *et al* [12], a species listed as Least Concern (LC) under the IUCN Red List Categories and Criteria version 3.1 [10] can additionally be categorized either as rare, critically rare or declining. The observed population decline of *C. bulbispermum* in South Africa [9] is due to over-exploitation as an ornamental plant, medicinal plant trade and popularity of the species in the medicinal plant (muthi) markets. It is within this context that the current study was carried out, aimed at comprehensively documenting the ethnobotany, phytochemistry and pharmacology of *C. bulbispermum* so as to highlight research gaps and provide a foundation for further investigations on the plant species.

Traditional medicinal usage

Crinum bulbispermum is a popular medicinal plant in both rural and urban communities in Southern Africa [6,7,13,14]. It is commonly known as Orange river lily in English in South Africa and it is also known by various vernacular and synonym names in different geographical regions in Lesotho, South Africa and Swaziland (see Table 1). In South Africa, the roasted bulbs of *C. bulbispermum* are applied to aching joints, rheumatism, varicose veins and backache, kidney or bladder infections and are used as poultices for septic sores and abscesses [13]. The southern Sotho in South Africa use the leaves and sliced or crushed bulbs to make a strong brew for treating colds, coughs, and as an external application or wash for wounds, scrofula and haemorrhoids (Table 2). According to Roberts [13], the leaves are used to bind dressings in place and flowers are placed over swollen joints and sprains to reduce swelling. Unspecified parts are used as infusions during pregnancy to ensure easy delivery [15]. The bulbs of *C. bulbispermum* are used for colds and to stimulate breast milk supplies by the Sotho in Lesotho [16]. Several tribes in Lesotho, South Africa and Swaziland are reported to use the juice squeezed from the base of the leaves to cure earache [13,17]. Sometimes pieces of roasted bulb are placed behind the ear or over the ear to ease the pain. Roberts [13] reports that some tribes in South Africa make a brew of the leaves which they believe to be an effective treatment for malaria. This same brew is drunk by the Zulu in South Africa as a treatment for rheumatic fever (usually half a cup chopped leaves in one cup boiling water and strained after standing for five minutes). The Tswana in South Africa drink a brew of crushed leaf bases and stalks to increase the flow of urine in bladder and kidney infections. The sliced bulb is also warmed and applied over the kidneys to ease discomfort [13]. Hutchings [18] records the use of *C. bulbispermum* bulb as a Zulu, Xhosa and Sotho gynaecological remedy and charm. *Crinum bulbispermum* leaves are browsed by cattle in South Africa [13].

Phytochemistry

Various reports on the phytochemical screening of *C. bulbispermum* bulbs, flowers, flowering stalks, leaves and roots confirm the presence of isoquinoline alkaloids, flavonoids, sterols, aldehydes, acids, esters, alcohols, esters, amines and amides, fatty acids and their esters (Table 3). Alkaloids are considered the major bioactive components of *C. bulbispermum* which exhibits various pharmacological effects [24,32,36-41]. Six distinct and structurally diverse isoquinoline alkaloids characteristic of the Amaryllidaceae family [42] have been isolated from *C. bulbispermum*, namely lycorine-type alkaloid (represented by eight alkaloids, alkaloids **1**-**8**); galanthamine-type alkaloid represented by galanthamine **9**; cherylline-type alkaloid represented by cherylline **10**; crinine-type alkaloid (represented by 29 alkaloids, alkaloids **11**-**39**), tazettine-type alkaloid (represented by 8α-ethoxyprecriwelline**39**, N-desmethyl-8α-ethoxypretazettine **40** and N-desmethyl-8β-ethoxypretazettine **41**) and two minor alkaloids namely augustamine **42** and trisphaeridine **43** (Table 3). Non-alkaloid compounds isolated from

C. bulbispermum bulbs, flowers and leaves are flavonoids represented by 15 compounds (compounds **44**-**58**); sterols represented by dihydrositosterol **59** and stigmasterol **60**; aldehydes, acids and esters represented by 4,5-methylenedioxy-4'-hydroxy-2-aldehyde-(1,1'-biphenyl) **61** and ρ-hydroxybenzene acetic acid ethyl ester **62**; alcohols, esters, amines and amides represented by β-(3,4-dimethoxyphenyl)-α,β-ethanediol **63** and choline **64**; and fatty acids and their esters represented by nine compounds, compounds **65**-**73** (see Table 3). Although very little pharmacological evaluation of non-alkaloid compounds isolated from *C. bulbispermum* has been done to date, flavonoids are known to exhibit antioxidant activity, free radical scavenging capacity, coronary heart disease prevention, hepatoprotective, anti-inflammatory, anticancer and antiviral activities [43]. While phytosterols are known to have bioactive prevention properties such as lowering of cholesterol levels [44] and cancer prevention [45] properties. Previous research by Li *et al* [46] showed that unsaturated fatty acids have antifungal potencies, low toxicities and good pharmaceutical properties.

Pharmacological activities

A number of pharmacological activities of *C. bulbispermum* have been reported in literature justifying some of its ethnomedicinal uses. Some of the listed pharmacological activities may not relate directly to the ethnomedicinal uses of *C. bulbispermum*, but may provide some insight into its potential therapeutic value and bioactive properties. A wide range of biological activities have been reported including antimicrobial [36], antinociceptive [37], antiplasmodial [38], antioxidant [37,39], cytotoxicity and anti-apoptotic [24,37,40,41] as well as effects on the central nervous system (CNS) [37].

Effects on the central nervous system (NCS)

In recent years, members of the Amaryllidaceae family have been shown to contain alkaloids with promising acetylcholinesterace (AChE) inhibiting properties [47,48]. Adewusi and Steenkamp [39] found ethyl acetate extracts of *C. bulbispermum* bulbs and roots as well as methanol bulb extract to have some level of inhibitory activity against AChE with IC_{50} values of 0.0021 ± 0.007, 0.0393 ± 0.014 and 0.0148 ± 0.039 mg/mL, respectively.

Table 1: Vernacular names and synonyms of *Crinum bulbispermum*

Vernacular name(s) with ethnic group/geographical region in brackets	Country	Reference
Lelutla, mototse (Sesotho)	Lesotho	[20]
Orange rivierle lie, Vaalrivierle lie, Vleilelie (Afrikaans), Orange river lily, wild amaryllis (English), lelutla, mototse (Sotho), mduze, umduze, umduzi, umnduze (Zulu)	South Africa	[5,6,8,14]
Umnduze (Swazi)	Swaziland	[19]
Synonym		
Amaryllis bulbispermum Burm. f.		[4]
Amaryllis longifoliasensu Jacq.		[4]
Amaryllis longifolia var. *riparia* Ker-Gawl.		[4]
Crinum capense sensu Herb.		[4]
Crinum longifolium (L.) Thunb.		[4]
Crinum riparium Herb.		[4]

Table 2: Ethnomedicinal uses of *Crinum bulbispermum* in Southern Africa

Use	Country practised	References
Abscesses and sores	South Africa	[17]
Aching joints	South Africa	[13]
Backache	South Africa	[17]
Binding for dressings	South Africa	[17]
Charm	Lesotho; South Africa	[16,18]
Colds	Lesotho; South Africa	[13]
Coughs	Lesotho; South Africa	[13]
Earache	South Africa	[17]
Gynaecological remedy	South Africa	[18]
Haemorrhoids	South Africa	[13,17]
Kidney and bladder infections (increase urine flow)	South Africa	[17]
Malaria	South Africa	[17]
Reduce swelling of swollen joints and sprains	South Africa	[17]
Scrofula	South Africa	[13]
Varicosities	South Africa	[17]
Wounds	South Africa	[13]

Table 3: Alkaloids and non-alkaloids isolated and characterized from *Crinum bulbispermum*

S/No.	Alkaloid	Plant part	References
	Lycorine-type alkaloid		
1	Lycorine	Bulbs, flowering stalks, roots	[21,22]
2	1, 2-di-O-acetyllycorine	Bulbs	[23]
3	8-hydroxylycorine-7-one	Bulbs	[24]
4	Hippacine	Bulbs	[25]
5	Hippadine (pratorine)	Bulbs	[21]
6	Hippamine	Bulbs	[24]
7	Pratorinine	Bulbs	[26,27]
8	Protorimine	Bulbs	[28]
	Galanthamine-type alkaloid		
9	Galanthamine	Bulbs	[26,27]
	Cherylline-type alkaloid		
10	Cherylline	Bulbs	[23]
	Crinine-type alkaloid		
11	Crinine	Bulbs	[22,23]
11	3-O-Acetyl-crinine (krepowine)	Bulbs	[23,28]
12	3-O-acetylhamayne	Bulbs, flowering stalks, roots	[29,30]
13	3-O-acetyl-powelline	Bulbs	[28]
14	3,4-anhydropowelline	Bulbs	[28]
15	6-hydroxycrinamine	Bulbs, flowering stalks, roots	[30]
16	11-hydroxyvittatine	Bulbs	[24]
17	Bowdensine	Bulbs	[23]
18	Bulbisine	Bulbs	[28]
19	Bulbispermine	Bulbs, flowering stalks, roots	[29,31]
20	buphanamine	Bulbs	[26,27]
21	Buphanidrine-6-β-ethoxy	Bulbs	[28]
22	Buphanisine	Bulbs	[28]
23	Buphanisine-6-α-hydroxy	Bulbs	[28,32]
24	Buphanisine-6-β-hydroxy	Bulbs	[28]
25	Buphanisine-6-α-ethoxy	Bulbs	[28]
26	Crinalbine	Bulbs	[21]
27	Crinamidine	Bulbs	[23]
28	Crinamine	Bulbs, flowering stalks, roots	[21-23,30]
29	Crinamine-6-α-hydroxy	Bulbs, flowering stalks, roots	[22,29]
30	Crinamine-6-β-hydroxy	Bulbs, flowering stalks, roots	[22,29]
31	Crinine-6-α-hydroxy	Bulbs	[28]
32	Crinine-6-β-hydroxy	Bulbs	[28]
33	Crinine-6-α-ethoxy	Bulbs	[28]
34	deacetylbowdensine	Bulbs	[23]
35	Hamayne	Bulbs	[21,31]
36	Powelline	Bulbs	[22,23]
37	Powelline-6-α-ethoxy	Bulbs	[28]
38	Vittatine	Bulbs	[26,27]
	Tazettine type alkaloid		
39	8α-ethoxyprecriwelline	Bulbs, flowering stalks, roots	[29]
40	N-desmethyl-8α-ethoxypretazettine	Bulbs, flowering stalks, roots	[29]
41	N-desmethyl-8β-ethoxypretazettine	Bulbs, flowering stalks, roots	[29]
	Other minor alkaloids		
42	Augustamine	Bulbs	[28,32]
43	Trisphaeridine	Bulbs	[28]
	Non alkaloid compounds		
	Flavonoids		
44	4'-hydroxy-7-methoxy-flavan	Bulbs	[28]
45	2(S)3',4'-dihydroxy-7-methoxy-flavan	Bulbs	[25]
46	4'-dihydroxy-7-methoxy-flavan-3-ol	Bulbs	[25]
47	7,4'-dihydroxy-flavanone[(-)-liquiritigenin]	Bulbs	[25]
48	7'-hydroxy-8-methoxy-flavanone[Isolarrien]	Bulbs	[25]
49	4'-hydroxy-7-methoxy-flavone	Bulbs	[33]
50	4,4'-dihydroxy-2-methoxy-chalcone	Bulbs	[33]
51	2',4,4'-trihydroxy-chalcone(Isoliquiritigenin)	Bulbs	[25]
52	4-hydroxy-2',4'-dimethoxy-dihydrochalcone	Bulbs	[25]

Table 3: Alkaloids and non-alkaloids isolated and characterized from *Crinum bulbispermum* ***(Continued)***

S/No.	Alkaloid	Plant part	References
53	Isorhamnetin-3-O-glucoside(3'-methyl-quercetin 3-O-glucoside)	Flowers	[34]
54	Kaempferol-3-O-glucoside	Flowers	[34]
55	Kaempferol-3-xyloside	Leaves	[31]
56	Kaempferol-3-O-β-D-xylopyranosyl(1→3)β-D-glucopyranoside	Flowers	[34]
57	Quercetin-3-O-glucoside	Flowers	[34]
58	Quercetin-3-O-β-D-(6-O-acetylglucopyranosyl)(1→3)β-D-glucopyranoside	Flowers	[34]
	Sterols		
59	Dihydrositosterol	Bulbs, leaves	[35]
60	Stigmasterol	Bulbs, leaves	[35]
	Aldehydes, acids and esters		
61	4,5-methylenedioxy-4'-hydroxy-2-aldehyde-(1,1'-biphenyl)	Bulbs	[25]
62	ρ-hydroxybenzene acetic acid ethyl ester	Bulbs	[33]
	Alcohols, esters, amines and amides		
63	β-(3,4-dimethoxyphenyl)-α,β-ethanediol	Bulbs	[33]
64	Choline	Bulbs	[21]
	Fatty acids and their esters		
65	Linoleic acid	Bulbs	[35]
66	Linoleic acid methyl ester	Bulbs, leaves	[35]
67	Oleic acid	Bulbs, leaves	[35]
68	Palmitic acid	Bulbs, leaves	[35]
69	Palmitic acid methyl ester	Bulbs, leaves	[35]
70	n-Hexacosane	Bulbs, leaves	[35]
71	n-Heptacosane	Bulbs, leaves	[35]
72	n-Nonacosane	Bulbs, leaves	[35]
73	n-Pentacosane	Bulbs, leaves	[35]

In an earlier research, orally administered aqueous leaf extract of *C. bulbispermum* doses of 1, 1.5 and 3 g/kg showed central inhibitory activity and markedly impaired the four parameters of rat hold-board test indicating its sedative properties [37]. The ability of *C. bulbispermum* to inhibit acetylcholinesterase may be ascribed to several alkaloids which have been isolated from the plant species (Table 3) indicating its potential for use in treatment of neurodegenerative diseases. Recently, the alkaloid galanthamine **9** isolated from *C. bulbispermum* (Table 3) was approved in the United States, many European countries and many Asian countries for the treatment of Alzheimer's disease (AD) [49]. Alzheimer's disease is characterized by a progressive impairment of cognitive functions including loss of memory and the inability to perform basic daily life activities [48]. Based on the cholinergic hypothesis, these symptoms are the results of the reduction in brain acetylcholine activity due to the catabolism of acetylcholine by ACHE [48]. Other alkaloids isolated from *C. bulbispermum* which have been screened for AChE inhibition activity include crinine **11** which exhibited inhibitory activity with IC_{50} of 461 µM, crinamine **28** (IC_{50} = 300 µM and lycorine **1** (IC_{50} = 213 µM) [48].

Antioxidant activity

A methanol bulb extract of *C. bulbispermum* showed some radical scavenging activity in ABTS assays with IC_{50} value of 0.0685 ± 0.041 mg/mL [39]. The total phenolic, flavonol and flavonoid contents of *C. bulbispermum* roots were relatively high for both solvents tested with total phenol of 202.38 ± 0.50 mg tannic acid/g, flavonol (20.79 ± 0.10 mg quercetin/g) and flavonoid (9.18 ± 0.50 mg quercetin/g) [39]. The levels of these phenolic compounds are an indication of the potential antioxidant activity of the plant extracts as phenolic compounds are well known as radical scavengers, metal chelators, reducing agents, hydrogen donors and singlet oxygen quenchers [50,51]. Additionally, the leaf extracts of *C. bulbispermum* showed modest antioxidant activity with EC_{50} value of 203.76 µg/mL which was assessed by the thiobarbituric acid reactive substances assay [37]. These findings support the traditional use of the plant species for treating neurological disorders especially those involving cholinesterase mechanism and reactive oxygen species.

Antinociceptive activity

Ratnasooriya *et al*. [37] evaluated the antinociceptive activity of *C. bulbispermum* leaves using three models of nociception namely tail flick, hot plate and formalin tests in male rats. The results showed that the leaf extracts had marked antinociceptive potential, particularly, when evaluated in the formalin test. According to Ratnasooriya *et al* [37], the obtained results suggest that the antinociception is mediated both spinally and supraspinally and is effective against phasic and continuous non-inflammatory or inflammatory pain. Ratnasooriya *et al* [37] attributed the antinociception of the leaf extract of *C. bulbispermum* to the results of opioid mechanisms, sedation and antioxidant activities of the species. These results support the traditional use of the species in various inflammatory ailments and diseases ranging from microbial infection to injury that result in swelling, cell injury and death.

Antiplasmodial activity

Van Dyk *et al* [38] screened *C. bulbispermum* for antiplasmodial activity using the [3H]-hypoxanthine incorporation assay against the chloroquine-resistant *Plasmodium falciparum*. Extracts of *C. bulbispermum* had IC_{50} values ≤ 1 µg/mL with the ethyl acetate extracts of the roots and bulbs having values comparable to chloroquine (0.04 µg/mL). Van Dyk *et al* [38] identified lycorine **1** as a potent antiplasmodial compound with an IC_{50} value of 0.03 µg/mL against the chloroquine-resistant strain (FCR-3) of *P. falciparum*, which is comparable to the activity of the crude extract and chloroquine. The most promising extract was the ethyl acetate bulb extract of *C. bulbispermum* with an IC_{50} value of 0.08 µg/mL and a security index of 2203.13. Likhitwitaywuid *et al* [52] reported an IC_{50} value of 0.3 µg/mL for lycorine **1** isolated from *C. amabile* against the chloroquine-resistant strain (W-2).

Antimicrobial activity

Griffiths [36] assessed antimicrobial activity of *C. bulbispermum* using the direct plate method and minimum inhibitory concentration values were determined. The best activity was observed for the alkaloid lycorine **1** against *Bacillussubtilis* [36]. One of the most common ethnomedicinal uses of *C. bulbispermum* is in the treatment of a wide range of infectious diseases caused by microorganisms such diseases or ailments include symptoms such as sores [17] and wounds [13]. Previous research showed that some of the alkaloids that have been isolated from *C. bulbispermum* have antibacterial activity. For example, the alkaloid crinamine **28** is known to have antibacterial activity, as it showed some strong activity against *Bacillus subtilis* and *Staphylococcus aureus* [53].

Cytotoxicity and anti-apoptotic activity

Seoposengwe *et al* [41] evaluated the cytoprotective potential of *C. bulbispermum*, after induction of toxicity using rotenone, in SH-SY5Y neuroblastoma cells. Rotenone reduced intracellular reactive oxygen species (ROS) levels after 24 h exposure. Pre-treating cells with *C. bulbispermum* extracts reversed the effects of rotenone on intracellular ROS levels. Rotenone exposure also decreased intracellular glutathione levels, which was counteracted by pre-treatment with any one of the extracts. MMP was reduced by rotenone, which was neutralized by pre-treatment with *C. bulbispermum* ethyl acetate extract. All extracts inhibited rotenone-induced activation of caspase-3. *Crinum bulbispermum* demonstrated anti-apoptotic activity and restored intracellular glutathione content following rotenone treatment, suggesting that they may possess neuroprotective properties.

Ratnasooriya *et al* [37] evaluated the sub-chronic toxicity of the aqueous leaf extract of *C. bulbispermum*. The extract induced mild to moderate toxicity in rats which developed diarrhoea and postural abnormalities on the second day, and two rats died by the fourth day. Liver and renal toxicities (increase of serum SGOT, SGPT, creatinine and urea) were also reported and the authors attributed this toxicity to the lycorine- and crinine-types of alkaloids present in *C. bulbispermum* (see Table 3). According to van Wyk *et al* [54] the major toxic compound in *C. bulbispermum* is the alkaloid crinamine **28**, which is regarded as highly lethal with an oral lethal dose LD_{50} at a concentration of 10 mg/kg body weight in dogs. Crinamine **28** is regarded as a powerful transient hypotensive in dogs and also shows respiratory depressant activity [55]. Similarly, Aboul-Ela *et al* [24] tested cytotoxicity of *C. bulbispermum* bulbs using the brine shrimp bioassay. The most effective one were the butanol fraction of the acidic extract of the non-flowering bulbs with LD_{50} of 63.1 µg/mL followed by the ether fraction of the alkaline extract of the flowering bulbs with LD_{50} of 73 µg/mL.

Van Dyk *et al* [38] evaluated the cytotoxicity of *C. bulbispermum* compounds as determined by *in vitro* cellular toxicity assay, reporting the IC_{50} value of 445.47 µg/mL for lycorine **1** against human kidney epithelial cells and toxicity index of > 15 000. Other researchers like Campbell *et al* [56] evaluated cytotoxicity activities of alkaloids

associated with *C. bulbispermum* reporting IC_{50} values of 0.6 and 0.7 µg/mL for lycorine **1** against the strains D-10 and FAC8, respectively. Likhitwitaywuid *et al* [52] reported IC_{50} values for ranging from 0.3 to 1.6 µg/mL against a series of human cancer cells. The anti-cancer activity of lycorine **1** was also reported by Li *et al* [57] as an effect due to the apoptosis inducing effect of lycorine **1** and it has been found to inhibit protein synthesis. Abd El Hafiz *et al* [32] examined the constituents of *C. bulbispermum* for activity against human leukemic Molt 4 cells. Of the flavan, 4'-hydroxy-7-methoxyflavan **44** and three crinanes namely powelline **36**, crinine-6-α-hydroxy **31** and buphanisine-6-α-hydroxy **23** tested, only buphanisine-6-α-hydroxy **23** and 4'-hydroxy-7-methoxyflavan **44** were moderately active, causing a steady decline (up to about 20 %) in the viability of leukemia cells over the three-day treatment period at a dosage of 71 µg/mL.

Seoposengwe *et al* [41] found the ethyl acetate extract of *C. bulbispermum* to have the least cytotoxic with an LC_{50} value of > 100 µg/mL. Contrary to this, the methanol extract produced an LC_{50} value of 46.18 ± 0.91 µg/mL. Adewusi *et al* [40] evaluated cytotoxicity of *C. bulbispermum* ethanol extracts against SH-SY5Y (human neuroblastoma) cells as well as toxically induced with Aβ, using the MTT and neutral red uptake assays. The extracts of the root and bulb of *C. bulbispermum* were the most toxic with IC_{50} values < 50 µg/mL in both assays. However, despite the observed toxicity, the roots and bulbs of *C. bulbispermum* still reduced the cell death induced by Aβ at less toxic doses. These results show that *C. bulbispermum* may contain several alkaloids with possible neuroprotective activities.

FINAL REMARKS

The widespread usage of *C. bulbispermum* as an ornamental plant and in traditional medicine resulting in negative impact on wild populations calls for conservation strategies and mechanisms for sustainable utilization of the species. The estimated declining rate of *C. bulbispermum* [9] is based on the decreasing population numbers at known habitat sites and reduction in the bulb sizes of the species sold in medicinal muthi markets [7]. Therefore, propagation protocols for *C. bulbispermum* should be developed as an alternative and viable means to provide sufficient plants to meet the demand for ornamental and medicinal purposes and at the same time protecting the natural populations. There is also need for comparative evaluation of the phytochemistry and biological activities of the various plant parts in an attempt to suggest plant part substitution as a means of conserving this highly collected plant species. If phytochemical and biological analyses prove that the leaves, flower stalks, flowers and fruits have comparable properties and activities to those of the bulbs and roots, then plant part substitution can be used to curtail the destructive harvesting of bulbs that has led to a continuous decimation of the wild populations.

The present review summarizes ethnomedicinal uses, phytochemistry, biological activities and cytotoxicity of different extracts and compounds of *C. bulbispermum*. *Crinum bulbispermum* has been traditionally used as herbal medicine throughout its distributional range in southern Africa, used for the treatment of common diseases and ailments like colds, cough, earache, haemorrhoids, malaria, wounds and others medical complications like gynaecological, kidney and bladder problems. Recent research on *C. bulbispermum* focused primarily on evaluating the antimicrobial, antioxidant, antiplasimodial, cytotoxicity activities of the species as well as the effects on the central nervous system. Alkaloids appear to be the major ingredients in *C. bulbispermum* bulbs, flowers, flower stalks, leaves and roots and these compounds appear to be responsible for the pharmacological properties of the species. Other important phytochemical constituents isolated from *C. bulbispermum* bulbs, flowers and leaves are flavonoids, sterols, aldehydes, acids and esters, alcohols, esters, amines, amides and fatty acids and their esters. Surprisingly, there is no systematic data linking the ethnomedicinal uses of *C. bulbispermum* to the phytochemical and pharmacological properties of these non-alkaloid compounds. Future studies should therefore, try to establish a link between the phytochemical and pharmacological properties of the non-alkaloid compounds and the ethnomedicinal uses of *C. bulbispermum*.

Although contemporary research involving *C. bulbispermum* is promising, it is too preliminary and sometimes too general to be used to explain and support its ethnomedicinal uses. Most of the mentioned phytochemical constituents and pharmacological studies have provided some suggestive scientific evidence for the various ethnomedicinal uses of *C. bulbispermum* in the treatment of parasitic diseases such as control and management of malaria, inflammatory ailments and wounds; there is need for extensive phytochemical, pharmacological, preclinical and clinical research. There is yet not enough

systematic data regarding the pharmacokinetics and clinical research of *C. bulbispermum* products and compounds. There are very few to nil experimental animal studies, randomized clinical trials and target-organ toxicity studies involving *C. bulbispermum* and its derivatives that have been carried out so far. Therefore, studies should identify the bioactive components, details of the molecular modes or mechanisms of action, pharmacokinetics and physiological pathways for specific bioactives of *C. bulbispermum*. Future studies should include the identification of any side effects and/or toxicity.

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Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

The authors declare that this work was done by the authors named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by them.

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