

Roscoea purpurea (Kakoli): Exploring the imperative for conservation of an endangered Ashtawarga plant through comprehensive review

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ABSTRACT

In COVID Era, achieving holistic health through Ayurveda is gaining popularity day by day. However, market fails to satisfy this ever-increasing demand due to unavailability and extinction of some of the major medicinal flora. Being an Ashtawarga plant, *Roscoea purpurea*, known for its multiple Ayurvedic formulations, medicinal and rejuvenating properties, is also available in very limited quantity and considered endangered. Traditionally, various parts of the plant like leaves, roots and flowers are used for treating diseases like diarrhoea, diabetes, inflammatory disorders, rheumatic pain and fever. On account of its rich chemical base consisting polyphenols and flavonoids, it is reported to have anti-cancerous and anti-oxidant activities. Howbeit, due to its endangered status, Department of AYUSH, Govt. of India has permitted to swap these kind of rare herbs with accessible substitutes. Adulteration with other plants opens the ground for exploitation in drug industry and degrades the quality and credibility of Ayurvedic medicine. So, the study aims to provide the importance of this endangered plant in lieu of its traditional uses, Ayurvedic properties and bioactivities. It also tries to emphasize that, owing to its rich medicinal record, not only the plant needs to be explored further but conservation is important as well. Hence, a strategic conservation plan has also been presented.

Keywords: Ashtawarga, Ayurveda, conservation, herbal drugs, *Roscoea purpurea*,

INTRODUCTION

Roscoea is an important genus belonging to the family Zingiberaceae which constitutes nearly 24 herbaceous tuberous geophytes, according to Royal Botanic Gardens, Kew having various medicinal uses (Dhyani et al., 2020; Zhao et al., 2017). *Roscoea purpurea*, known by the common name of *Kakoli*, is a large lush green perennial rhizomatous herb between 15-30 cm in height with light brown, thick, fleshy, and bundled roots (rhizomes). The leaves are fleshy and flowers are hooded expressing a purple tone shade (Sahu et al., 2010; Gopal et al., 2014). It is an important species of the "Ashtawarga" group with *Jivaka-Rishabhak*, *Mahameda-Meda*, *Riddhi-Vriddhi* and *Kakoli-Kshirakakolia* as clan members. These plants are claimed to be useful in healing and treating fractured bones, weakness, pyrexia, body overweight, and regulating diabetes; also renowned for their effect in balancing *vata*, *pitta*, and *raktadoshas* (Dhyani et al., 2010; Chinmay et al.,

2011; Virk et al., 2015; Rajashekhar et al., 2015; Misra et al., 2015). Due to lack of clarity, several different authors used the substituted species of *Roscoea procera* Wall., *Roscoea alpina* Royle and others but it is evident that the genuine species of *Kakoli* is *Roscoea purpurea* Smith, which can be characterized by purple colour stout stem and flowers as described in ancient literatures of *Shaligram* and *Bhav Prakash Nighantus* and also indicated by Balkrishna et al. (2012). Although, due to its rare nature, the Ministry of Ayush has now permitted to replace it with other available sources like *Aswagandhā* (*Withania somnifera* (L.) Dunal) and *Krsna-mūsalī* (*Curculigo orchoides* Gaertn) but the real nature of the plant should be conserved, for which its conservation and cultivation is mandatory (Virk et al., 2017).

Further, in order to correctly characterize the true identity of species, some compounds have been isolated and purified from different plant parts to be used as chemical markers. Kaur et al., (2020a)

has reported the presence of a compound, named Sitostanol caffeoate in the roots of plant, which can be used as chemo-marker. Several other studies reported the presence of flavonoids, steroids, terpenoids, alkaloids and phenolic acids. Rhizomes of *R. purpurea* are reported to possess significant antidiabetic and hypolipidemic activity. In other studies, immunomodulatory, spermoprotective, and anti-tubercular potency have been revealed (Bairwa *et al.*, 2012; Subramoniam *et al.*, 2013; Kumar, 2014). Traditionally, roots (rhizomes), leaves, and flowers are used in the treatment of diabetes, diarrhoea, high blood pressure, hyperthermia, haematemesis, polydipsia, rheumatic pain and inflammation etc. Immuno-stimulating properties of rhizomes, extracted in ethanolic extract have also been demonstrated (Handa, 1980; Singh and Rawat, 2011; Gopal *et al.*, 2014). The rhizomes and tubers are essential ingredients of number of herbal formulations prepared like tonic and Chyawanprash (Handa, 1980; Singh and Rawat, 2011; Kaure *et al.*, 2020a). In Nepal, boiled tubers of the plant are eaten and traditionally used in veterinary medicines (Handa, 1980; Singh and Rawat, 2011). So, with an objective to present a comprehensive and upgraded review of this highly valuable Ayurvedic species, this study has been drafted to include traditional uses, major phytochemicals and biological studies conducted till now. The present review summarizes the updated information regarding the chemical constituents, pharmacological study, medicinal uses and ayurvedic significance along with its various formulations as well as some mentioned the different ways to conserve and cultivate it which is a pressing need.

Botanical description

Kakoli is a stout perennial herb, 25–38 cm tall. Leaves are 4–8, elliptic, lance-shaped to oblong-ovate, 14–20 cm long, sometimes sickle-shaped, tip tapering and slightly eared at a base on lower leaves, side veins parallel; leaf eared fringed with hairs. Flowers are borne in a cluster at the top, the cluster stalks are enclosed by upper leaf sheaths, only the upper part of bracts and flowers visible. Typical structured flowers are slightly purple, mauve, lilac, pink or white with purple markings where only one or two flowers open at a time. Outer calyx is tubular-

shaped, three petals arising from the calyx form a slightly longer tube than the calyx and terminates with trilobular structure raised vertically having one hooded central lobe surrounded by two slightly smaller side lobes. Staminodes, constituting four sterile stamens, are reformed inside the petals where the two lateral staminodes appeared as small straight petals and the other two central ones got partially fused at the base and forms a lip. Lip is non-deflexed, 4.5–6.5×2–5 cm (Flowers of India).

Geographical distribution

As a native species of Nepal, *R. purpurea* is distributed and scattered in India and Bhutan also, where it prefers to grow along green hillsides, alpine grassland, stony ridges and slopes of Central to Eastern Himalaya extending from Uttarakhand to Assam and Sikkim, upto an elevation of 3300 m (Dhyani *et al.*, 2010; Gopal *et al.*, 2014).

Ayurvedic properties and formulations of *Kakoli*

Kakoli is one of the most profligate plants of the nature. It is grouped among Ashtawarga plants, *Jivanîya*, *Sukrajanana* and *Brahmhaniyagana* in different Ayurvedic scriptures (Mishra, 2020). The plant is sweet in taste, heavy and mucilaginous in attribute and cold in potency. In Ayurveda, it is considered *Vata pittasamaka*, *slesmataka*; *roghanatâ* and is a part of different Ayurvedic preparations namely, *Chyavanprashrasayan* (a class of rejuvenating tonics), *Mahakalyan* and *Jivaniyaghrita* (Herbal medicines with ghee as a base), *Vachadi* and *Chitrakadi tala* (herbal oil, having properties to treat enlarged lymph nodes, fistula and sinus), *Vrahini gutika* (useful against erectile dysfunction) and *Astawarga* and *Jivaniya gunachurna* (Marde and Mishra, 2019).

Phytoconstituents of *Roscoea purpurea*

Several bioactive compounds from different chemical categories such as diterpenoids, flavonoids, and phenolic acids have been reported from *R. purpurea* till now (Table 1). Sitostanol caffeoate, a compound claimed to lower the risk of coronary heart diseases has been isolated from the roots by Kaure *et al.*, (2020a), which can be further utilized as a marker to identify and differentiate the species from its homotypes. In Nepal, the boiled tubers are eaten and used in veterinary medicine. A research conducted by Misra *et al.*, (2015) have

proved the nutraceutical value of the set tubers and showed that they are highly rich in fibre (28%), proteins (3.5%) and oil (3.5%). Total phenolic and flavonoid content were found to be ranged between 7.10 to 6.10 %, respectively. Phytochemical screening of the powdered tubers showed the presence of alkaloids, carbohydrates, flavonoids, glycosides, phenolics, proteins, saponins, and tannins (Misra et al., 2015; Devkota and Timalsina, 2021). The nutritional components of the rhizomes constitute fiber, oil, protein, sugar, and starch (Owolabi et al., 2012; Misra et al., 2015). Rawat et al. (2014) reported the presence of riboflavin, thiamine, minerals, fat and fibers in rhizomes. The methanolic extract having blackish-brown composition displayed the presence of two compounds belonging to catechin-type of class such as epigallocatechin and epicatechin along with other compounds in the roots (Kauret al., 2020a).

Singamaneni et al. (2021) reported two new compounds coronarin K and L from the class labdanediterpenes along with other known compounds as coronarin A, kaempferol 3-O-methyl ether, kaempferol, fenozan acid, 3-(3-methoxy, 4-hydroxyphenyl)-2-propenoic acid ferulic acid, caffeic acid, bisdemethoxy curcumin and gallic acid from its rhizomes. Coronarin K demonstrated to have anti-cancer potency when evaluated against lung cancer cell line (A-549).

Miyazaki et al., (2014) isolated flavonoids as kaempferide, kaempferide 3-O- β -D-glucuronopyranoside, kaempferol 3-O- β -D-glucuronopyranoside and (Z)-3-hexen-1-ol- β -D-glucopyranoside from aerial parts and kaempferide, kaempferol 3-O-methyl ether from the rhizomes of *R. purpurea*. Kaur et al. (2020b) reported the triterpenoid, lupenone for the first time from *Kakoli* roots. Similarly, stigmasterol was also obtained from powdered leaves of *R. purpurea* by Barai and Bag (2019) which is known to possess various pharmacological activities including anti-osteoarthritic, antioxidant, anti-inflammatory, anti-mutagenic, hypoglycemic, anti-genotoxicity and anticancerous (Kauret al., 2011; Ali et al., 2015). They have also reported the presence of monoterpenoids in ethanolic extract of leaves by GC-MS study.

Medicinal uses

Roscoea purpurea is an important species contributing to the Ayurvedic members of *Ashtawarga* plants, considered as prime *rasayana* group possessing health rejuvenating properties. Due to its anti-oxidant, anti-aging and cell regenerative properties, it is an essential ingredient of several poly herbal formulations, specially 'Chayawanprash', which is widely used in India as an energy and immunity booster supplement (Misra et al., 2015; Ravalet al., 2015). Its regular use also enhances the intellect, long life and memory (Tripathi, 2013). Traditionally, its roots and other parts in the form of various preparations are used medicinally (Table 2). Roots and rhizomes are considered as good appetizer, aphrodisiac, and tonic (Miyazaki et al., 2014). Therapeutically, they are reported to be anti-rheumatic, diuretic, expectorant, febrifuge, galactagogue, haemostatic and *Sukrajanana* (semen count enhancer) in nature (Balkrishna et al., 2012; Sharma and Sharma 2008). In Ayurveda, its roots are considered beneficial in *Vishamajvara* (high fever), *Pittaroga*, *Raktapitta* (Bleeding disorders), *Netraroga* (Eye disorders), *Hridayaroga* (Heart disorders), *Daha* (Burning sensation) and *Swasaroga* (Respiratory disorders) (Ayurvedic Pharmacopoeia of India, 2001). According to Saheli et al. (2019), the plant is known to possess anti-diabetic and hypoglycemic effects. The various medicinal properties of the plant along with its parts have been summarized (Fig. 2).

Anti-oxidant activity

Tubers and rhizomes of *R. purpurea* are commonly used for various Ayurvedic formulations, so some of the researchers have tried to summarize pharmacological aspects of these parts. The methanolic extract derived from the tubers of *R. purpurea* was tested for anti-oxidant activity by using ferric reducing power, DPPH and α -carotene linoleate assays. The extract has shown to regulate the reducing power with an increase in concentration, similar to positive control (ascorbic acid, quercetin, rutin and BHT). In DPPH and carotene bleaching assay, the extract showed IC_{50} 810.66 \pm 1.154 & 600.66 \pm 1.154 μ g/ml, which can be justified by the presence of polyphenolic content of the tubers which are well validated for their potent bioactivities (Misra, et al., 2015). In a

similar study, the ethanolic extract and its petroleum ether, chloroform, acetone, ethanolic and aqueous fractions prepared from tubers of *R. purpurea* were evaluated for anti-oxidant activity by using DPPH assay. The extract and petroleum ether, chloroform, acetone, ethanolic and aqueous fraction exhibited anti-oxidant effects with IC₅₀ of 0.925±0.005, 1.25±0.005, 0.25±0.005, 0.48±0.005, 0.77±0.011 & 1.04±0.005 mg/ml, respectively (Srivastava *et al.*, 2015).

Anti-cancer activity

According to WHO, cancer is the leading source of death globally with a record 10 million deaths in 2020. Lung cancer and breast cancer are among the most common cancers and the search for plant bioactives as anticancerous aid is progressing. In his research study, Singamaneni, *et al.* (2021) has evaluated the potential of methanolic extract, its chloroform fraction and compounds, coronarin K and L, firstly isolated from rhizomes of *R. purpurea*, along with other known compounds; coronarin A, bisdemethoxycurcumin and kaempferol 3-O-methyl for anti-cancer activity against human lung (A549), colon (HCT-116), breast (MCF-7) and pancreas (Bxpc-3) cancer cell lines by using MTT assay. Paclitaxel was used as standard (IC₅₀ 6.2±0.20, 8.6±0.045, 46±0.74 & 3.81±0.32 μM, respectively for A549, HCT-116, Bxpc-3 & MCF-7 cells). It was found that extracts and fraction exhibited anti-cancer effects against all tested cell lines with IC₅₀ 21.35±0.83 to >100 μM when compared with paclitaxel. Moreover, coronarin K showed potent anti-cancer effect against A549, HCT-116, Bxpc-3 & MCF-7 cells with IC₅₀ 13.49±0.62, 26.03±1.46, 56.70±2.17 & 56.24±0.83 μM, respectively when compared with paclitaxel. Furthermore, coronarin L and coronarin A displayed anti-cancer effects against A549 cells with IC₅₀ 33.78±1.37 & 61.80±2.82 μM, respectively while coronarin L against MCF-7 cells with IC₅₀ 49.84±2.61 μM when compared with paclitaxel. Additionally, coronarin L, coronarin A and bisdemethoxycurcumin showed effects against Bxpc-3 cells with IC₅₀ 56.83±1.92, 22.83±1.47 & 68.15±2.41 μM, respectively when compared with paclitaxel. These results revealed that *R. purpurea* possessed prominent anti-cancer effects. (Singamaneni *et al.*, 2021). In a previous study, the ethanolic extract and its petroleum ether,

chloroform, acetone, ethanolic and aqueous fractions from rhizomes of *R. purpurea* were also evaluated for anti-cancer potential against human lung carcinoma (A549), cervical cancer (SiHa), Chinese hamster ovary cells (CHOK1) and rat glioma (C6) cell lines using SRB assay where vinblastine was established as control. It is noticed that the extract and fractions exhibited cytotoxic effect against all cell lines with IC₅₀ <10 to >150 μg/ml (Srivastava *et al.*, 2015).

Immunomodulatory activity

The ethanolic extract (300 & 600 mg/kg, p.o.) from the rhizomes was investigated for immunomodulatory activity by analyzing delayed type hypersensitivity (DTH) response and macrophage phagocytosis using carbon clearance assay in Swiss Albino mice. Cyclophosphamide (30 mg/kg) utilized as standard. It was observed that, the extract at both doses significantly (p<0.05) increased the foot pad thickness, WBC and total platelet count of anti-genically (SRBC suspension) challenged mice when compared with control group. Moreover, extract at both doses significantly (p<0.05) increased the phagocytic index using carbon clearance method when compared with control group. Therefore, these results justified that the extract possessed immunostimulant properties (Sahu *et al.*, 2010).

Why do we need to conserve *Roscoea purpurea* (Kakoli)?

India has ample resource of medicinal plants comprises of 8,000 diverse species. In the 21st century, there is a remarkably focus among individualson health and environmental preservation, resulting in a significant increase in the utilization of medicinal plants. The substantial amount of raw material is derived from forest area. Therefore, the forest area has been exacerbated by the pharmaceutical and associated industries. Consequently, myriads number of plants have been threatened and some of them have been enlisted in Red Data Book. *Roscoea purpurea* (Kakoli) is also among one of them, which is needed to be conserved and cultivated due to its excessive utilization (Kumar and Jnanesha, 2016; Virk *et al.*, 2017).

There are two main strategies for the conservation: *in situ* and *ex situ* conservation. In-

Table1: Chemical constituents of *Roscoea purpurea* in different plant parts

Chemical Constituents	Plant Parts	Extract	References
1. Phenolic compounds			
Protocatechuic acid	Tubers	Methanol/Ethanol	(Mishra et al., 2015; Srivastava et al., 2015)
Ferulic acid	Tubers	Methanol/ Ethanol	(Mishra et al., 2015; Srivastava et al., 2015)
Syringic acid	Tubers	Methanol/Ethanol	(Mishra et al., 2015; Srivastava et al., 2015)
Apigenin	Tubers	Ethanol	(Srivastava et al., 2015)
Vanillic acid	Tubers	Methanol	(Mishra et al., 2015)
Gallic acid	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
Caffeic acid	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
Ellagic acid	Rhizomes	Methanol-Chloroform fraction (Giri et al., 2017)	
Ferulic acid	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
Fenozan acid	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
3-(3-methoxy,4-hydroxyphenyl)-2-propenoic acid	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
ferulic acid			
2. Flavonoids	Aerial parts/Methanol		(Miyazaki et al., 2014)
Kaempferide	Rhizomes		
Kaempferol	Rhizomes/		
Kaempferide 3-O- β -D-glucuronopyranoside	Tubers	Methanol	(Singamaneni et al., 2021; Mishra et al., 2015)
Kaempferol 3-O- β -D-glucuronopyranoside	Aerial parts	Methanol	(Miyazaki et al., 2014)
Kaempferol 3-O-methyl ether	Rhizomes	Methanol	(Miyazaki et al., 2014; Singamaneni et al., 2021)
Quercetin	Rhizomes	Methanol	(Misra et al., 2015; Devkota and Timalsina, 2021)
Epicatechin	Roots	Methanol	(Kaure et al., 2020a)
Epigallocatechin	Roots	Methanol	(Kaure et al., 2020a)
Rutin	Rhizomes/	Ethanol	(Giri et al., 2017; Srivastava et al., 2015)
6-methylpinocembrin	Tubers	Ethanol	(Chaudhari, 1988)
3. Curcuminoid			
Bisdemethoxycurcumin	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	
4. Terpenoids			
Coronarin A (Labdanediterpenes)	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	Coronarin K
		Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)
Coronarin L	Rhizomes	Methanol-Chloroform fraction (Singamaneni et al., 2021)	Sterols
Stigmasterol	Leaves	Ethyl acetate	(Barai and Bag, 2019)
Beta-sitosterol	Tubers	Ethanol	(Chaudhari, 1988)
Beta-sitosterol-beta-D-glucoside	Tubers	Ethanol	(Chaudhari, 1988)
Triterpenoids			
Lupenone	Roots	Methanol	(Kaure et al., 2020b)
Ursolic acid	Tubers	Ethanol	(Chaudhari, 1988)
5. Miscellaneous			
Adenosine	Rhizomes	Methanol	(Miyazaki et al., 2014)
(Z)-3-hexen-1-ol- α -D-glucopyranoside	Aerial parts	Methanol	(Miyazaki et al., 2014)
n-hentriacontane	Tubers	Ethanol	(Chaudhari, 1988)

Table 2: Medicinal uses and formulations of Roscoea purpurea

Traditional Uses	Preparation/Ayurvedic formulations	References
Abdominal disorders	Whole plant: Processed with <i>Vacādio</i> oil and other herbs is used as an <i>anuvāsanavasti</i> to treat distension, dyspepsia and <i>gulma</i> .	(Rao, 2011)
Cardio-vascular disorders	Roots: Powder with other herbs as <i>Crepidium acuminatum</i> , <i>Malaxis muscifera</i> ,	(Balkrishna, 2014)
Cardiac diseases	<i>Pueraria tuberosa</i> , <i>Crepidium acuminatum</i> ,	
Chest injury	<i>Malaxis muscifera</i> , <i>Polygonatum verticillatum</i> , <i>Pueraria tuberosa</i> , <i>Medā</i> , <i>Mahāmedā</i> mixed with honey. <i>Nāgabalāsarpi</i> processed with <i>Roscoea purpurea</i> and other herbs are useful in chest injury	
Cuts and wounds	Leaves and roots: Dried powder	(Pande, Tiwari, and Pande, 2006; Bisht, 2017).
Diabetes	Leaves, roots, rhizomes and flowers: Infusion/ Singamaneni, 2021; Seth	(Misra et al., 2015; paste. and Kondal, 2020).
Diarrhea and dysentry	Leaves, roots, rhizomes and flowers: Infusion/ Decoction.	(Seth and Kondal, 2020; Misra et al., 2015; Singamaneni, et al., 2021)
Gout	Whole plant: Decoction with <i>Prunus cerasoides</i> , <i>Glycyrrhiza glabra</i> , <i>Curcumalonga</i> , <i>Vetiveria zizanioides</i> , <i>Rubia cordifolia</i> , <i>Lilium polyphyllum</i> , is prepared as a paste, processed with oil and made into a tailak known as <i>Khuākapadmataila</i> , useful in treating gout and burning sensation.	(Tripathi and Pandey, 2012)
Hemorrhage	Roots: Powder helps to treat bleeding disorders. Whole plant: Powder processed with <i>Nāgabalāsarpi</i> and other herbs are useful in intrinsic hemorrhage.	(Balkrishna, 2014)
Paediatric diseases	Whole plant: powder, processed with <i>Ghrta</i> (clarified butter) and <i>Cedrus deodara</i> , <i>Lilium polyphyllum</i> , <i>Crepidium acuminatum</i> , <i>Malaxis muscifera</i> , <i>Vigna trilobata</i> and <i>Teramnus labialis</i> is useful in appropriate doses in case of emaciation in children.	(Balkrishna, 2014)
Respiratory disorders	Tubers: Paste.	(Swar, 2014)
Cold and cough	Whole Plant: Powder with other herbs and honey.	(Balkrishna, 2014)
Bronchitis and asthma	Rhizome: Powder mixed with orange rind powder (Balkrishna, 2014)	
Reproductive disorders	Roots and Rhizomes: Decoction	(Kaur et al., 2017; Seth and Kondal, 2020)
Impotency		
Sinusitis	Whole plant: Powder processed with <i>Ghrta</i> and herbs as <i>Glycyrrhiza glabra</i> , <i>Curcumalonga</i> , <i>Polygonatum verticillatum</i> , <i>Nelumbo nucifera</i> , used as <i>parishechana</i> (irrigative therapy) in <i>pittaja</i> associated with <i>drysipelas</i> and sinus.	(Rao, 2011).
Skin disorders	Roots: powder, mixed with black pepper and rat dropping, applied to boils.	(Singh, 2009; Quattrocchi, 2012).
Tuberculosis	Whole plant: Powder processed with <i>Nāgabalāsarpi</i> and other herbs. Bulbs are used to treat phthisis.	(Balkrishna, 2014; Saroya, 2013).
Urinary disorders	Roots: Extract as a tonic; ingested to treat urinary infection and polyuria	(Misra et al., 2015; Miyazaki et al., 2014)

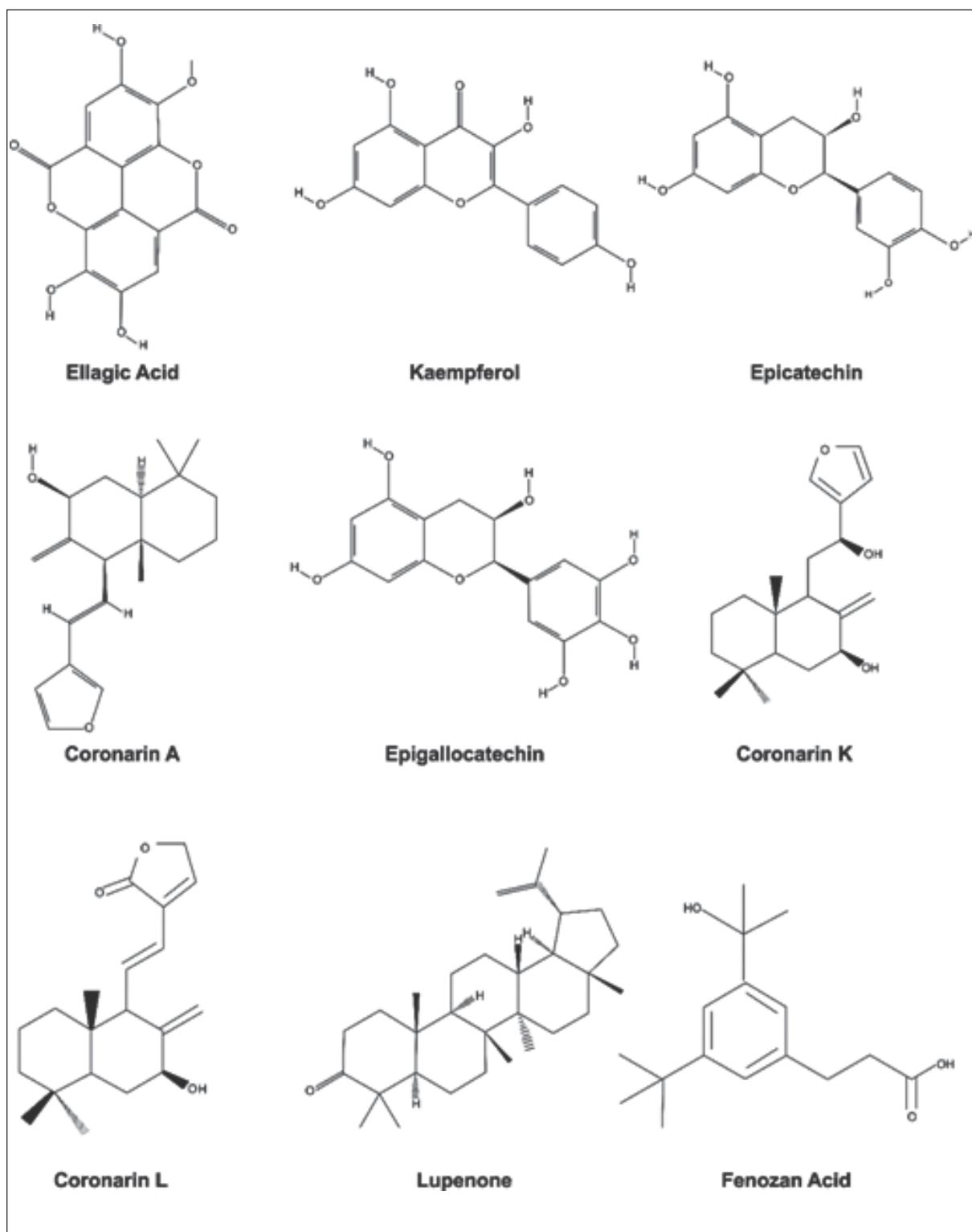


Fig.1: Chemical structures of *R.purpurea* compounds

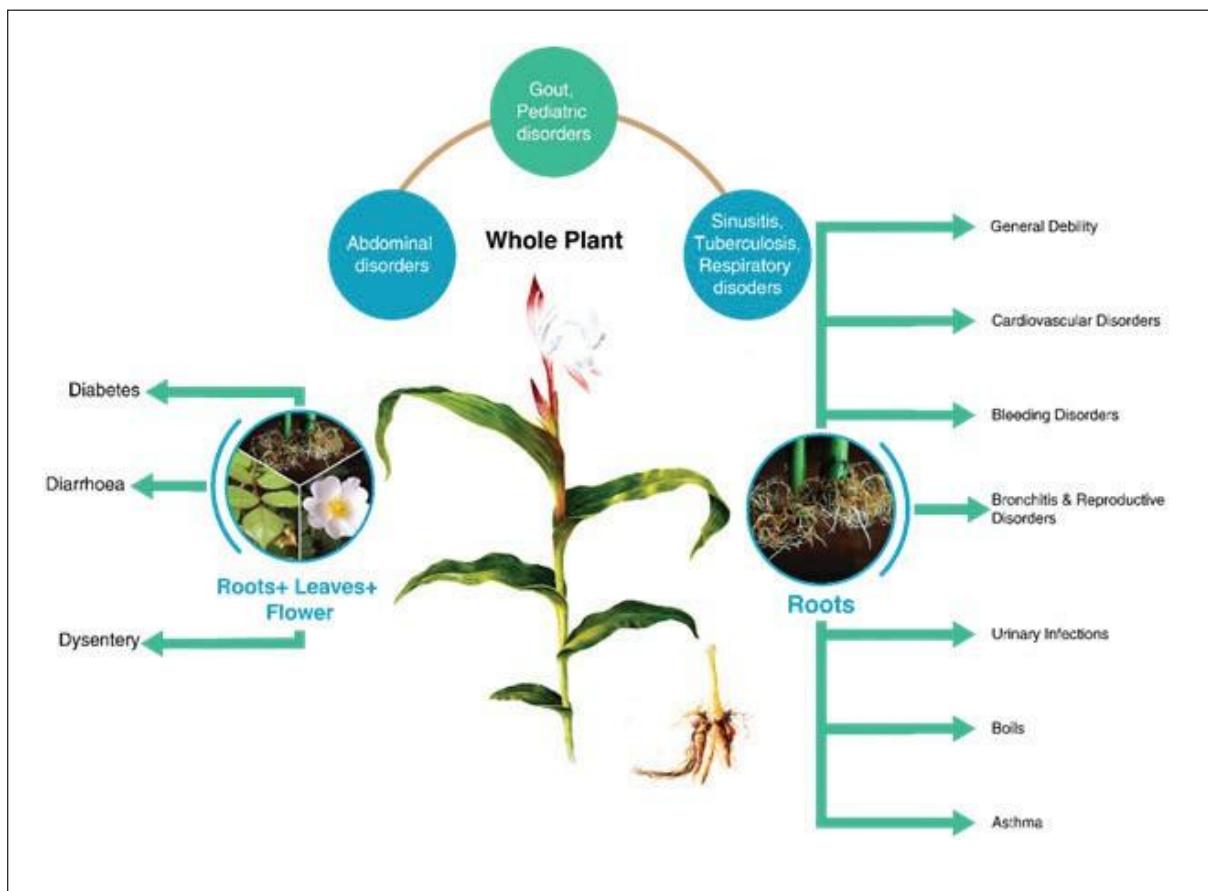


Fig.2 :Medicinal Properties ofKakoli (*R.purpurea*).

situ strategy of conservation focuses on the “on-site conservation” of the wild genetic diversity in natural environment. It is a methodological technique that entails to conserve such endangered plants species with indispensable therapeutic value (Chandra, 2016). The involvement of community is to preserve both threatened plant species as well as their natural communities. This procedure adopts an eco-centric strategy for conservation, where preserving endangered plant species is prioritized while minimizing human intervention (Akshay et al., 2014). There are two essential segments of this technique, firstly, is to explore for the geographical location of these plant species based on their therapeutic properties and industrial value. Secondly, to monitor, manage, and preserve such plant species by forming a network and zone of forest habitat of a specified size that must be established (Dhama et al., 2018). In addition to this, the significant aspect of this method is to investigate and survey of such plants after a regular interval of

time that may lead to better perception of natural biodiversity and various methods of preservation of such plants. This type of well-organised conservation strategy can evoke recovery of endangered plant species.

There are several *in-situ* conservation methods for medicinal plants such as biosphere reserve, national park, sacred groves and others. The Ministry of Environment and Forest, Government of India has identified 13 biosphere reserves for the conservation of medicinal plants in different states of India (Kadam and Pawar, 2020). In West Bengal and Tamil Nadu have established biospheres named Sunderban and Gulf of Mannar respectively for the protection of medicinal plant species (Chandra, 2016). In India, there are 91 national parks, 2 out of them namely, Pin Valley National Park and Great Himalayan National Park in Himachal Pradesh have been established. Unluckily, a substantial population live in proximity to these forests (Chandra, 2016). The establishment of national park

create the involvement of local people which generate responsibility towards them to protect medicinal plant species (Huang *et al.*, 2002). People who live near to forests area can help in ensuring the survival of endangered medicinal plant species only if they are made aware of conservation methods.

Sacred groves are said to be small or substantial areas of vegetation that are protected on the basis of tradition and religious beliefs. In India there are 13,720 small scale sacred groves through which medicinal plant species is protected (Murtem and Chaudhry, 2014). Manipur is the state reported over 365 sacred groves. These groves persist significant ecologically value because they link ritual behavior to ecology, especially in order to protect threatened plant species. Traditional agricultural methods are eco-friendly and long-lasting approach which supports to conserve the diversity of medicinal plant species (Ahmad *et al.*, 2021). The native farmers who live near the forest region have gathered more knowledge about medicinal plant (Benniamin, 2011) and their expertise should be organized through appropriate documentation. We can disseminate the garden conservation for the protection of medicinal plants in small apartments, flats, house and bungalows. For the conservation of medicinal plant species proper monitoring is needed in above mentioned protected areas. People may be driven to support conservation efforts via financial incentives.

Ex-situ strategy of conservation focuses on the “off-site conservation” of the wild genetic diversity in natural environment. It is a pivotal procedure for the protection of medicinal plant species that are high risk of extinction (Werden *et al.*, 2020). *Ex situ* conservation objective to reintroduce the endangered species into their native habitat in order to secure their survival and periodically to produce substantial amount of planting resources used in the formation of therapeutic drugs. In recent years, the government promotes long-term plant species conservation through parks and botanical gardens. When the medicinal plant species were grown away from their natural habitat, not only they retain their high efficacy but their reproductive parts are extracted and maintained in the repository in seed banks for future regeneration (Kumar and Jnanesha, 2016). There are several *ex-situ* conservation

methods for medicinal plants such as seed bank, botanical garden etc.

In India, one of the Institute named Tropical Botanical Garden and Research Institute (TBGRI) located in Kerala has taken step towards the conservation and sustainable use of medicinal plant wealth of peninsular India. The TBGRI performs major activities such as seed gene bank, *in-vitro* gene bank and field gene bank to protect medicinal plant species (Bhattacharyya *et al.*, 2006). Botanical garden also plays a vital role in sustaining the ecosystem to escalate the survival of rare and endangered plant species. They include a large diverse variety of plant species that share a common condition. Botanical garden acts as center for release, revive and rehabilitation of rare, endangered and extinct prone species of medicinal plants and other significant plant genetic resources. Apart from this, it plays significant function in education and training center sectors such as horticulture, gardening, landscaping, *ex-situ* conservation and environmental awareness. A case study was done to elucidate the medicinal plant conservation through botanical garden wherein Malabar Botanic Garden of India is used to train local farmers for the cultivation and repository methods of medicinal plants more over giving market guidance. In 2 years, they have trained more than 200 farmers, who later established a farmers' association to exchange knowledge (Hawkins, 2007). Botanical gardens have been built in several regional communities. Example: Government Botanical Gardens in Tamil Nadu, Garden of Medicinal Plants in West Bengal and many more across the country (Kadam and Pawar, 2020).

CONCLUSION

The rhizomes, tubers and other parts of *R. purpurea* are well described in various Ayurvedic texts and literatures as an analgesic, widely used for treating various health ailments in Nepal and India. Despite the wealth of historical data supporting its traditional applications, there exists a shortage of comprehensive studies validating these assertions. While existing publications have demonstrated the plant's potential as a natural antioxidant and cytostatic agent, there remains a dearth of empirically grounded literature designating it as a promising candidate for future

drug discovery initiatives. The phytochemical profiling of the plant has revealed the presence of flavonoids and phenolic acids, known for their robust bioactive attributes. As such, it is advisable to investigate the individual compounds and their synergistic effects to determine optimal formulations and dosages. Notably, compounds like stigmasterol, due to their nanoscale dimensions, hold promise for advancement in nanobiotechnology research. To lay the groundwork for forthcoming research endeavors, an increased number of both *in-vivo* and *in-vitro* studies are imperative. Rigorous clinical trials are essential to substantiate the therapeutic efficacy of this traditional medicine within the conventional medical framework. Addressing these research gaps should be a focal point for future investigations, guided by the imperative to conserve these valuable botanical resources.

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