Journal of Pharmaceutical Research International

33(60B): 2556-2561, 2021; Article no.JPRI.77592
ISSN: 2456-9119
(Past name: British Journal of Pharmaceutical Research, Past ISSN: 2231-2919, NLM ID: 101631759)

In-vitro Conservation of Phytochemically Enriched Orchids of Indian Western Himalayas

Saranjeet Kaur [a*]

[a] *Faculty University Institute of Biotechnology, Division Biosciences, Chandigarh University, District Mohali, Punjab, India.*

Author's contribution

The sole author designed, analyzed, interpreted and prepared the manuscript.

Article Information

DOI: 10.9734/JPRI/2021/v33i60B34914

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/77592

Review Article

Received 20 October 2021
Accepted 24 December 2021
Published 26 December 2021

ABSTRACT

Orchids are identified for their beautiful ornamental flowers. These flowers exceptionally possess long extended vase life. Besides being floriculturally significant, they also find their description in the ancient Ayurvedic system of medicine for their therapeutic uses. These monocot herbaceous plants possess diverse bioactive chemical compounds such as terpenes, alkaloids, etc. that are responsible for their therapeutic value properties. The orchids are collected stealthily from their natural habitats indiscriminately and have become rare in the wild and their populations can be saved through *in vitro* conservation techniques. The present communication conveys conservation techniques used for saving orchid species from getting extinct.

Keywords: In-vitro; monocot; orchids; therapeutic; alkaloids.

1. INTRODUCTION

The vast geographic expense of India, harbours a broad range of plant species of diverse habits and habitats. Taxonomically, the orchidaceae, is highly evolved family of monocotyledons, encasing 25,000-35,000 species in nearly 800 genera [1]. The orchid blooms are extremely beautiful and continue fascinating scientists and a layman globally. These natural marvels exhibit an array of mesmerizing shapes, sizes, and colours. In Indian system of medicine, the orchids also find mention for their curative properties [2].

In Indian system of medicine, an Ayurvedic formulation, 'Ashtavarga', which is known to be a revitalizing herbal medicine consists of 8 herbs

**Corresponding author: E-mail: sksekhkone7632@cumail.in;*

and out of these, four herbs to family orchidaceae namely *Habenaria intermedia* (Riddhi), *Habenaria edgeworthii* (Kakoli), *Dendrobium macrae*, and *Malaxis wallichii* (jivak) [1,3]. A sizeable number of phytochemicals and drugs are found in orchids. A variety of orchid species are known to possess glucoside and alkaloid compounds (Table 1).

2. ORCHIDS OF WESTERN HIMALAYA (SHIMLA HILLS)

The Indian Western Himalaya is expanded through tropical plains from alpine to arctic climates within an altitudinal range of 300-8611m. It receives an annual rainfall of almost 600-1800 mm. The region of Indian western Himalaya is one of the major hotspot of biodiversity [5]. In Shimla hill slopes of Tara Devi, Fagu, Mashobra, Charabra inhabits a variety of terrestrial orchid species for instance, *Satyrium nepalense*, *Epipactis helleborine, Calanthe tricarinata*, *Malaxis acuminata*, *Malaxis muscifera*, *Habeneria intermedia*, *H. pectinata*, *Habeneria edgeworthii*, *Liparis rostrata*, *Liparis ovata*, *Goodyera repens*, *Goodyera procera* of therapeutic value. Table 2 summarises a few terrestrial species with their phytochemical constituents of therapeutic value that inhabits Shimla hills (Table 2).

3. CONSERVATION STATUS

Therapeutic orchids are enriched with a large number of secondary metabolites such as glycosides, alkaloids, and flavonoids. These orchid herbs are used widely in the Indian ayurvedic medicinal system. Orchid species were collected from their foster homes unabatedly. This over-exploitation exceeds their natural regeneration. As a result, the entire orchidaceae family is placed under rare, threatened, and endangered category. It is tabulated in the appendix I & II of checklist prepared by IUCN [22].

Table 1. Orchid species and phytochemical compounds [4]

Sr. No.	Orchid species name (Botanical name)	Phytochemical compound	Phytochemical compound class
1.	*Aerides crispum*	Acridine	Phenanthopyran
2.	*Agrostophyllum callosum*		Stilbenoids triterpenoid,
3.	*Agrostophyllum breviceps*	Agrostophyllinol	Triterpenoid,
4.	*Anoectochilus formosanus*	Kinsinoside	Glycoside
5.	*Arundina graminifolia*	Arundina	Stilbenoids
6.	*Bulbophyllum* species	Gymopsin	Phenanthrene
7.	*Coelogyne cristata*	Coeloginanthridin, Coeloginanthrin	Phenanthrenes
8.	*Coelogyne flaccida*	Flacccidin, Oxaloflaccidin	Phenanthrenes
9.	*Cypripedium calceolus*	Cypripedium	1-4 Phenanthrenequinone
10.	*Dendrobium moschatum*	Rotundatin, Moschatin	Phenanthrene
11.	*Dendrobium nobile*	Gigantol, Dendrobine, Nobilonine	Bibenzyl
12.	*Epipactis helleborine*	Oxycodone, benzyloxypropylindol, didehydroepoxymorphinan	Bibenzyl derivatives
13.	*Eulophia Nuda*	Nudol	Phenanthrenes
14.	*Eulophia ochreata*	Dimethoxy phenanthrene, dihydro methoxy phenanthrene	Phenanthrene
15.	*Orchis latifolia*	Loroglossin	Glucoside
16.	*Vanda cristata*	Melanin	Glycoside
17.	*Vanilla planifolia*	Vanillin	Alkaloids, flavonoids, glycosides

Table 2. Shows some therapeutic orchid species of western Himalaya (Shimla hills) with their chemical constituents

Sr. No.	Orchid species (Habit)	Trade Name	Plant part used	Treats disorder	Bioactive compounds
1.	*Malaxis acuminata* D. Don (terrestrial)	Jivak	Pseudobulb	External haemorrhage, rheumatism, dysentery, immunity promoter [6]	Glycosides, flavonoids, and piperitone, alkaloids, citronellal, beta-sitosterol, 1.8-cineolle, eugenol, Limonene, p-cymene, cetyl alcohol, O-Methylbatatasin [7,8]
2.	*Habenaria intermedia* D. Don (terrestrial)	Vrddhi	Tuber	Health tonic, Aphrodisiac, Anthelmintic, [9]	Alkaloids, steroids, carbohydrates, flavonoids, terpenoids, phenolics, tannins [10]
3.	*Habenaria pectinata* D. Don (terrestrial)	Safed musli	Leaf, Tuber	Rheumatism, Leaf (grinded) treats snake bites [11]	-
4.	*Habenaria edgeworthii* Hook.f. Collett (terrestrial)	Riddhi	Leaf and Tuber	Treats blood disorder, aphrodisiac, [11], [12]	Coumarin, Alkaloids, Phenolic compounds and glycosides [13]
5.	*Goodyera repens* (L.) R. Br. (terrestrial)		Tuber	Extract act as blood purifier [14] Cures appetite, cold, Stomach and kidney, disorder [1]	Alkaloids, Loroglossin (https://singapore-memories.com/pages/therapeutic-orchids)
6.	*Satyrium nepalense D.Don*. (terrestrial)	Salam misri/ Ban-alu	Root, Tuber	Antimicrobial [15] Roots used to treat malaria, dysentery [16],	Alkaloids, glycosides, flavonoids, unsaturated sterols/triterpenes [17]
7.	*Cypripedium cordigerum* D. Don (terrestrial)	Jibri [18]	Roots	Health tonic [19]	-
8.	*Epipactis helleborine* (L.) Crantz. (terrestrial)	-	Rhizome	Narcotic value, antidote to HIV [20], [21]	-

Normally, the orchids require an amiable atmosphere to flourish in their territorial habitats. Their extinction could also pose a deep influence on the ecological system. Consolidative scientific methodologies are required for their *ex situ* and *in situ* conservation. There is a need of continuous efforts to eco-restore these rare species through biotechnological practices. *In vitro,* techniques have emerged as a viable system to save and multiply their germplasm from getting extinct in nature.

4. STRATEGY FOR CONSERVATION OF BIODIVERSITY

Conservation term is a combination of 'preservation and utilization'. In broader sense, conservation refers in saving wild populations of plant species in their natural environment. Biodiversity of a species can be conserved by adopting scientific approaches as well as participation of the society. Principally, the conservation of plant genetic diversity is achieved by following measures:-

1. *In-situ* conservation
2. *Ex-situ* conservation

4.1 *In-situ* Conservation

In situ conservation deals especially with saving plant species in their natural environment. A particular species which is saved in its wild habitat where it thrives naturally refers to *in situ* conservation. It includes wild-life sanctuaries, sacred grooves, national parks, sacred sites, biosphere reserves, cultural landscapes, protected forest areas and gene banks. In natural environment, the diversity in plant species can be conserved on a long-term basis at genus, species, and ecosystem level.

Many conservation approaches are adopted to save naturally growing diverse orchids by establishing National Orchid and Biodiversity Parks, biosphere reserves, orchid sanctuaries, etc.

4.2 *Ex-Situ* Conservation

Ex-situ conservation is a measure that is external to the natural habitat. Mainly, it is established in the botanical gardens many institutes exclusively engaged in botany such as Botanical Survey of India, several universities, R&D research centres, national parks, and farmer's field, and also done through *in vitro* seed banks, gene banks, and pollen banks, DNA libraries, and through advanced techniques involving cryopreservation and various plant tissue culture techniques.

Table 3. Sites and their states of *in situ* orchid conservation

S. No.	Sites	States
1	Sessa orchid sanctuary	West Kameng District, Arunachal Pradesh
2	Deorali orchid sanctuary	Gangtok, Sikkim
3	Kaziranga National Orchid and Biodiversity Park	Golaghat & Nagaon district, Durgapur, Assam
4	Pachmarhi Biosphere Reserve	Madhya Pradesh
5	Nilgiri Biosphere Reserve	Western Ghats & Nilgiri Hills, South India

In vitro asymbiotic seed germination-The method of germinating orchid seeds *in vitro* in a nutrient-enriched medium assists with conserving and propagating orchid species [23]. The technique developed by Knudson established asymbiotic seed germination protocols. This protocol helped in evading the requirement of mycorrhiza in *in vitro* germination of orchid seeds. This technique also assists in achieving an optimum percentage, besides reducing the time lapse occurring in between pollination process and seed sowing [24]. The asymbiotic seed germination helps in achieving a better percentage of germination from immature seeds, than from mature seeds, as the immature seeds are always in their physiologically active state and are devoid of any kind of dormancy or inhibitory factors [25]. The asymbiotic seed germination technique has been successfully used in a large variety of orchid species of diverse habit and habitats [26-35].

5. CONCLUSION

The orchid species are valuable herbaceous monocot plant species, which synthesise a variety of biochemical compounds. These herbs find their mention in the ancient ayurvedic system for their curative properties. This indigenous knowledge, if blended together with modern research activities has the capacity to make new drug formulations for the benefit of mankind in today's times to cure chronic diseases.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Author has declared that no competing interests exist.

REFERENCES

1. Hossain MM. Therapeutic orchids: traditional uses and recent advances- An overview. Fitoterapia. 2011;82:102-140.
2. Kaushik P. Glimpses of Medical Botany in Atharvaveda (*Kand* 1-3). Proceedings of National Workshop on Vedic Education, Gurukul Kangri University, Hardwar. 1983;196-203.
3. Handa SS. Orchids for drugs and chemicals. In: Vij SP, editor. Biology conservation and culture of orchids, New Delhi: Affiliated East West Press; 1986.
4. De LC, Medhi RP. Orchid-A diversified component of farming systems for profitability and livelihood security of small and marginal farmers. J Global Biosci. 2015;4:1393-1406.
5. Jalal JS, Jayanthi J. An annotated checklist of the orchids of western Himalaya, India. Lankesteriana. 2015;15:7-50.
6. Tamta BP, Sharma AK, Lokho P, Singh, A. Propagation and conservation of endangered orchid *Microstyliswallichii*Syn. *Malaxis acuminata* (Jeevak) in its natural habitats of Uttarakhand Himalayas. Int J Sci Environ Technol. 2015;4:424 – 432.
7. Rastogi RP. Compendium of Indian medicinal plants. New Delhi: CDRI Lucknow and NSIC.2011;2(437):1970-1979.
8. Bhatnagar SK. Handbook of medicinal plants. 3rd ed. Jaipur: Pointer Publisher (India); 2001.
9. Habbu PV, Smita DM, Mahadevan KM, Shastry RA. Protective effect of *Habenaria intermedia* tubers against acute and chronic physical and psychological stress paradigs in rats. Rev Brasileira de Farma. 2012; 22:568-579.
10. Virk JK, Bansal P, Kumar S, Singh R, Rawal RK, Gupta V, Mathiani M. Isolation of sinapic acid from *Habeneria intermedia* D. Don.: A new chemical marker for the identification of adultration and substitution. Curr Traditional Med. 2019; 4(1).
DOI: org/10.2174/2215083804666181030101709
11. Singh A, Duggal S. Medicinal orchids – An overview. Ethnobot Leaflets. 2009;13: 399-412.
12. Chauhan NS. Medicinal and aromatic plants of Himachal Pradesh. New Delhi: Indus Publishing Company; 1999.
13. Sedai CP. Phytochemical profiling of *Habenaria edgeworthii* hook. f. ex collett (vriddhi) of Deoban under Chakrata forest division. M. Sc. dissertation. Dehradun, Uttrakhand: FRI University, Indian Council of Forestry Research & Education; 2019.
14. Badola HK, Pal M. Endangered Medicinal plant in Himachal Pradesh. Curr Sci. 2002;83:797-798.
15. Singh DK. Antioxidant and antimicrobial activities of medicinally important salep orchid, *Satyrium nepalense* d. don (salammishri). Plant Cell Biotech Mol Bio. 2019;20:752-758.
16. Hossain MM. Traditional therapeutic uses of some indigenous orchids of Bangladesh. Med Aromatic Plant Sci Biotech. 2009;3:100-106.
17. Mishra AP, Saklani S. *Satyrium nepalense*: A rare medicinal orchid of western Himalaya (India): Phytochemical screening, antimicrobial evaluation and conservation studies. Indonesian J Pharm. 2012;23;162-170.
18. Teoh ES. Orchid species from Himalaya and south east Asia. Vol. 1(A-E). Switzerland: Springer Nature; 2021.
19. Pant B. Medicinal orchids and their uses: tissue culture a potential alternative for conservation. African J Plant Sci. 2013;7:448-467.
20. De Clercq E. Trends in drug development for the treatment of AIDS: Compounds interfering with the initial stages of HIV replicate cycle. Euro J Pharma Sci. 1994;2:4-6.
21. Vij SP, Kaur Saranjeet, D. Manjari. Orchids in medicine. Orchid News (TOSI). 1997;13:19.
22. IUCN. IUCN directory of protected areas in Oceania prepared by the World Conservation monitoring centre. UK. 1991;447.

23. Knudson L. Nonsymbiotic germination of orchid seeds. Bot Gaz. 1922;73:1-25.
24. Sagawa Y. Green pod culture. The Florida Orchidist. 1963;6:296–297.
25. Yam TW, Weatherhead MA. Germination and seedling development of some Hong Kong orchids. Lindleyana. 1988;3:156-160.
26. Park SY, Murthy HN, Paek KY. *In vitro* seed germination of *Calanthe sieboldii*, an endangered orchid species. J Plant Biol. 2000;43:158–161.
27. Lo SH, Nalawade SM, Kuo CL, Chen CL, Tsay HS. Asymbiotic germination of immature seeds, plantlet development and ex vitro establishment of plants of *Dendrobium* tosaenso Makino – A medicinally important orchid. *In vitro* Cell Dev Bio. – Plant. 2001;10:528–535.
28. Roy J, Banerjee N. Cultural requirements for *in vitro* seed germination, protocorm growth and seedling development of *Geodorumdensiflorum* (Lam.) Schltr., Indian J Exp Biol. 2001;39:1041–1047.
29. Buyun L, Lavrentyeva A, Kovalska L, Ivannikov R. *In vitro* germination of seeds of some rare tropical orchids, Acta Uni Lat. 2004;676:159-162.
30. Shimura H, Koda Y. Micropropagation of *Cypripedium macranthos* var. *rebunense* through protocorm-like bodies derived from mature seeds. Plant Cell Tiss Organ Cult. 2004;78:273–276.
31. Deb CR, Temjensangba S. Effect of different factors on non-symbiotic seed germination, formation of protocorm-like bodies and plantlet morphology of *Cleisostoma racemiferum* (Lindl.) Garay. Indian J Biotechnol. 2004;5:223-228.
32. Pierce S, Ferrario A, Cerabolini B. Outbreeding and asymbiotic germination in the conservation of the endangered Italian endemic orchid Ophrys benacensis. Plant Biosys. 2010;144:121–127.
33. Pant B, Shrestha S, Pradhan S. In vitro seed germination and seedling development of Phaius tancarvilleae (L'Her.) Blume. Sci World. 2011;9:50-52.
34. Kaur Saranjeet, Bhutani K K. In vitro propagation of Dendrobium chrysotoxum (Lindl.). Flori Orna Biotechnol. 2011;5:50-56.
35. Kaur Saranjeet, Bhutani KK. Asymbiotic seed germination and multiplication of an endangered orchid – Paphiopedilum venustum (Wall. ex Sims.). Acta Soc Bot Pol. 2016;85:3494-3505.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/77592