# *In Vitro* Conservation of Phytochemically Enriched Orchids of Indian Western Himalayas

**Abstract**

Orchids are identified for theirelegant ornamentalflowers. These flowers exceptionally possess long extended vase life. Apartfrom being extremely floricultural, they also find their description in ancient Indian ayurvedic system of medicine for their therapeutic uses. These monocot herbaceous plantsembrace diverse bioactive chemical compounds such as terpenes, alkaloids etc. that are responsible for their therapeutic properties.Orchids are collected stealthily from their natural habitats discriminately, and have become rare in the wild and their populations can be saved through *in vitro* conservation techniques.The present communicationreports about theconservation techniques used for savingorchid species from getting extinct.

**Keywords:***in vitro*, monocot, orchids, therapeutic, alkaloids.

**Introduction:**

India,in its vast geographic expanse, harbours a broad range of plant species of diverse habits and habitats.The orchidaceae,taxonomically, is highly evolved monocot family encasing 25,000-35,000 species in nearly 800 genera [1]. The orchid blooms are extremely beautiful and continues fascinating scientists and a layman globally. These natural marvels exhibit an array of mesmerizing shapes, sizes andcolours. 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 and out of these, four herbs to family orchidaceae namely*Habenaria intermedia* (Ridhi), *Habenariaedgeworthii* (Kakoli), *Dendrobium macraei*, and*Malaxiswallichii* (jivak) [1], [3].A sizeable number of phytochemicals and drugs are found in orchids. A variety of orchid species are known to possessglucoside and alkaloid compounds (Table 1).

Table 1 Orchid species and phytochemical compounds [4]

| Sr. No. | Orchid species name | Phytochemical compound | Phytochemical compound class |
|---|---|---|---|
| 1. | *Aeridescrispum* | Acridin | Phenanthopyran |
| 2. | *Agrostophyllumcallosum* | | Stilbenoids triterpenoid, |
| 3. | *Agrostophyllumbreviceps* | Agrostophyllinol | Triterpenoid, |
| 4. | *Anoectochilusformosanus* | Kinsinoside | Glycoside |
| 5. | *Arundinagraminifolia* | Arundinan | Stilbenoids |
| 6. | *Bulbophyllum* species | Gymopsin | Phenenthrene |
| 7. | *Coelogynecriststa* | Coeloginanthridin, Coeloginanthrin | Phenenthrenes |
| 8. | *Coelogyneflaccida* | Flacccidin, Oxaloflaccidin | Phenenthrenes |
| 9. | *Cypripedium calceolus* | Cypripedin | 1-4 Phenenthrenequinone |
| 10. | *Dendrobium moschatum* | Rotundatin, moschatin | Phenenthrene |
| 11. | *Dendrobium nobile* | Gigantol,Dendrobine, Nobilonine | Bibenzyl |
| 12. | *Epipactishelleborine* | Oxycodone, benzyloxypropylindol, didehydroepoxymorphinan | Bibenzyl derivatives |
| 13. | *Eulophianuda* | Nudol | Phenenthrenes |
| 14. | *Eulophiaochreata* | Dimethoxyphenenthrene, dihydromethoxyphenenthrene | Phenenthrene |
| 15. | *Orchislatifolia* | Loroglossin | Glucoside |
| 16. | *Vanda criststa* | Melianin | Glycoside |
| 17. | *Vanilla planifolia* | Vanillin | Alkaloids, flavonoids, glycosides |
| 18. | *Vanda roxburghii* | Kinonside | Glucoside |

**Orchids of western Himalaya (Shimla hills)**

The Indian Western Himalaya isexpanded through tropical plains to 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 Shimlahill slopes of Tara Devi, Fagu, Mashobra, Charabra inhabits a variety of terrestrial orchid speciesfor instance *Satyrium nepalense*, *Epipactis helleborine*, *Calanthetricarinata*, *Malaxisacuminata*, *Malaxismuscifera*,*Habeneria intermedia*, *H. pectinata*, *Habeneriaedgeworthii*, *Liparisrostrata*, *Liparis ovata*, *Goodyerarepens*, *Goodyeraprocera* of therapeutic value. Table 2 summarises a few terrestrial species with their phytochemical constituents of therapeuticvalue that inhabitsShimla hills (Table 2).

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

| Sr. No. | Orchid species | Trade Name | Plant part used | Treats disorder | Bioactive compounds |
|---|---|---|---|---|---|
| 1. | *Malaxis acuminata* D. Don | Jivak | Pseudobulb | External haemorragesrheumatism; 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. | *Habeneria intermedia* D. Don | Vrddhi | Tuber | Health tonic, AphrodisiacAnthelmintic, 9] | Alkaloids, steroids, carbohydrates, flavonoids, terpenoids, phenolics,tannins [10] |
| 3. | *Habeneriapectinata*D. Don | Safed musli | Leaf, Tuber | Rheumatism; Leaf (grinded) treats snake bites [11] | - |
| 4. | *Habenariaedgeworthii*Hook.f. Collett. ‘ | Riddhi | Leaf and Tuber | Treats blood disorder, aphrodisiac, [11], [12] | Coumarin, Alkaloids, Phenolic compounds and glycosides [13] |
| 5. | *Goodyera repens* (L.) R. Br. | | 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 | 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 | Jibri or Shakalkal | Roots | Healthtonic [18] | - |

| | | | |
|---|---|---|---|
| 8. | *Epipactis helleborine* (L.) Crantz. | Rhizome | Narcotic value, - antidote to HIV [19], [20] |

**Conservation Status**

Therapeutic orchids are enriched with large number of secondary metabolites such as glycosides, alkaloids, and flavonoids. These orchid herbs are used widely in Indian ayurvedic medicinesystem. Orchid species are collected from their foster homes unabatedly. This over-exploitation exceeds their natural regeneration. As a result, the entireorchidaceaefamily is placed under rare, threatened and endangered category. It is tabulated in the appendix I & II of checklist prepared by IUCN (1991) [21].

The orchids requireamiable atmosphere to flourish in their territorial habitats. Their extinction could also pose a deep influence on 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 practises. *In vitro* techniques have emerged as a viable system to save and multiply their germplasm from getting extinct in nature.

**Strategyfor Conservation of Biodiversity:**

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

1. *In-situ* conservation

2. *Ex-situ* conservation

***In-situ* conservation:**

*In situ* conservation is dealsspecifically with saving plant speciesalready growing 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 long-term basis at genus, species and ecosystem level.

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

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 |

***Ex-Situ* Conservation:**

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

*In vitro*asymbiotic seed germination-The method of germinating orchid seeds *in vitro* in nutrient-enriched medium assists with conserving and propagating orchid species [22]. The technique developed by Knudson establishedasymbiotic seed germination protocols. This protocol helped in evading the requirement of mycorrhizain*in vitro* germination of orchid seeds. This technique also assists in achieving optimum percentage,

besides reducing the time lapse occurring in between pollination process and seed sowing [23]. The asymbiotic seed germination helps in achieving 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 [24].The asymbiotic seed germination technique has been successfully used in a large variety of orchid species of diverse habit and habitats[25],[26],[27],[28],[29],[30],[31],[32],[33],[34]

**Conclusion:**

The orchid species are valuable herbaceous monocot plant species which synthesize a variety of biochemical compounds. These hers find their mention in 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.

NOTE:

The study highlights the efficacy of " ayurvedic " which is an ancient tradition, used in some parts of India. This ancient concept should be carefully evaluated in the light of modern medical science and can be utilized partially if found suitable.

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