Chapter 2

REVIEW OF LITERATURE

Studies on family Valerianaceae exhibit considerable diversity in flower and fruit morphology particularly variation in flower morphology, which shows the number of stamens ranged from four to one (Eriksen, 1989). Pollen morphology of the family has been examined by various workers (Ting, 1949; Erdtmann, 1952; Wang, 1960; Taranvschi et al., 1966; Kuprianova and Alyoshina, 1978; Nowicke and Skvarla, 1979; Patel and Skvarla, 1979; Blackmore and Cannon, 1983, Backlund and Nilsson, 1997) in relationship to taxonomy (Clarke 1978), and diverse localites from North Western Europe (Clarke and Jones (1981) and Pakistan (Perveen and Qaiser, 2007). Studies on the population status assessment of the species of family Valerianaceae are available. For example assessment of availability and habitat preference of *Nardostachys jatamansi* – a critically endangered medicinal plant of west Himalaya was performed (Airi et al., 2000). Studies on the effects of different harvesting patterns on the population ecology of *Nardostachys grandiflora* (Valerianaceae) in Shey- Phoksundo National Park and in its buffer zone in northwestern Nepal have been investigated (Ghimire et al., 2005). The effect of harvesting pressure for trade on the population ecology of *N. grandiflora* was also analyzed and the species is reported to be vulnerable to harvesting. Similarly, impact of human pressure on the population structure of *Valeriana jatamansi* growing naturally in Pakistan has been reported; the results revealed significant decrease in overall population density after the collection period (Alyemeni and Sher, 2010); two to four-fold increases in different parameters (e.g., abundance, distribution, population density, herbage cover and population types, etc.) were observed in protected sites as compared to the unprotected areas (Alyemeni and Sher, 2010).

On account of its wide ranging value, *V. jatamansi* finds important place in existing literature and has been described as an important species of Asian origin and categorized as a psychopharmacological agent (Vaidya et al., 1997). The species is also known as a natural source of sedative and tranquilizing valepotriates (Violon et al., 1983; Mishra,
2004). Presence of Valerenic acid and valerinone in the plant is a source of drug valerian, that ranks at 8\textsuperscript{th} place among the top selling herbal supplements (Blumenthal, 2001). Of the 16 species of genus \textit{Valerina} recorded from India (Table 1.1), \textit{V. jatamansi} is mostly used in traditional medicine and has long history of uses, which finds mention as a medicine in the Rigveda, Charaka Samhita and modern medicine system. A number of species of \textit{Valeriana} have been well investigated worldwide for its phytochemical constituents. For example, using high performance liquid chromatographic method, combined with diode array detection, various active constituents like valtrate, isovaltrate, acevaltrate, didrovaltrate, isovaleroydroxy didrovaltrate, valerenic acid, hydroxyvalerenic acid and acetoxyvalerenic acid, baldrinal and homobaldrinal, in roots of \textit{V. officinalis} were identified (Bos et al., 1996). Similarly, study on volatile constituents of the essential oil of \textit{V. alliariifolia} has revealed the existence of 26 compounds, of which 20 (96.3\%) were identified as oil (Taherpour et al., 2010). Valerenic acid in \textit{V. jatamansi} as well as in \textit{V. officinalis} has been designated as a key marker compound (Singh et al., 2006). Study on interspecific and intraspecific comparisons of valepotriates content in three species of \textit{Valeriana}, namely, \textit{V. jatamansi}, \textit{V. officinalis} and \textit{V. officinalis} var. \textit{latifolia} showed difference in valepotriates content; \textit{V. jatamansi} showed the highest valepotriates content (Chen et al., 2002). Chemical characterization of different species of \textit{Valeriana} (i.e., \textit{V. jatamansi}, \textit{V. himalayana}, \textit{V. pyrolaefolia}, and \textit{V. hardwickii}) was carried out (Mathela et al., 2005). Likewise, the total phenolic content and antioxidant potential of certain species of family Valerianaceae (e.g., \textit{V. officinalis}, \textit{V. hardwickii}, \textit{V. jatamansi}, etc.) have also been reported (Cai et al., 2004; Surveswaran et al., 2007; Wojdylo et al., 2007; Kalim et al., 2010; Das. et. al., 2011; Bhatt et al., 2012). Total antioxidant potential of \textit{V. officinalis} has been tested by various \textit{in vitro} assays which clearly indicated the antioxidant potential of the species. Some other species of the same family, i.e., \textit{Patrinia scabiosaefolia} and \textit{Nardostachys jatamansi} also exhibited antioxidant potential in different \textit{in vitro} antioxidant assays (Cai et al., 2004; Ahmed et al., 2009). A new flavone glycoside linarin (LN) has been reported in \textit{V. officinalis} and possess sedative and sleep-enhancing properties (Fernandez et al., 2004).
Molecular markers have been used for characterization and genetic diversity analysis of various species of Valerianaceae. For instance, phylogenetic relationships in genus *Valeriana* were studied using RAPD markers (Takeuchi et al., 2001). Preliminary analysis of the sequence alignment of the chloroplast intergene atp beta-rbcL in tribe Valerianaceae exhibited insertion-deletion evolutionary events, which were combined with nucleotide substitutions in large zones in some of the selected taxa (Raymundez et al., 2002). Population structure and genetic variation analysis in *V. wallrothii* with relation to different ecological locations was performed using AFLP and chloroplast SSR markers (Grassi et al., 2004). Microsatellite markers were isolated and characterized in another species of Valerianaceae, *Plectritis congeta* for studying the evolution and concluded that the species is an ideal candidate for studying the ecological and evolutionary consequences of gene (McEwen et al., 2011). Plant regeneration from callus and suspension cultures of *V. edulis* ssp. *procera* via simultaneous organogenesis and somatic embryogenesis was investigated (Castillob et al., 2000). Analysis of various investigations performed in different aspects of target species is presented herewith.

### 2.1. Phytochemical studies

Phytochemical investigation of the species have revealed the Linarin-isovalerianate (Thies, 1968), dihydrovaltrate (Bounthanh et al., 1981), valepotriates (Becker and Chavadeoi, 1985), sesquiterpenoids (Ron et al., 2000), 6-methylapigenin and hesperidin (Marder et al., 2003) as the major chemical ingredients in *V. jatamansi* (Table 2.1; Plate 2.1). Among these, valepotriates are the most active ingredients used in preparation of several medicines. Valepotriates are a group of monoterpenoids of iridoid type compound having epoxy group and beta-acetoxy isovaleric acids. The known group of iridoid compounds has been isolated from members of Valerianaceae, is called valeriana-epoxy triesters, and abbreviated as valepotriates (Thies and Funke, 1966). The valepotriates were first isolated in 1966 from *V. jatamansi* (Thies and Funke, 1966) and named valtrate, acevaltrate and didrovaltrate (Plate 2.1). Recently, new acylated iridoids, jatamanvaltrates A-M have been isolated from this species (Lin et al., 2009). The naturally occurring valepotriates/iridoid possess various activities like antibacterial, anticancer, anticoagulant, antifungal, anti-inflammatory, antioxidative, antiprotozoal,
hepatoprotective and neuroprotective (Dinda et al., 2009). Two bekkenollide type sesquiterpenoids have also been isolated from this species (Xu et al., 2011). The composition of valepotriates varies among the species and area (Chen et al., 2002).

Studies on rhizomes and root oil of *V. jatamansi* have shown the existence of a large number of chemical compounds like valerenic acid, isovalerenic acid, valerianine, valeranone, 1-pinene, 1-camphene, alpha-santalene, ar-curcumene, xanthorrizol, terpineol, alkaloids, bornyl isovalerinate, chatinine formate glucoside, etc. (Arora and Arora, 1963; Nadkarni, 1976; Bos et al., 1996). Citric acid, malic acid, maaliol, succinic acid and tartaric acid have also been isolated from the rhizomes of this species (Kapoor, 1990). The active terpenoids including valtrate, didrovaltrate, maaliol, patchouli alcohol, and 8-acetoxy patchouli alcohol were also reported (Keochanthala-Bountanth et al., 1993; Mathela et al., 2005). Besides, sesquiterpene hydrocarbons (ar-curcumene, beta-farnesene, alpha and beta patchoulenes and sesquifenchene), valerenone, cryptomeridiol, patchouli alcohol, etc. are reported (Houghton, 1999a; Chowdhury, 1999). Another, phytochemical investigation suggests that the root oil contain sesquiterpene hydrocarbon (ar-curcumene, β-farnesene, α- and β-patchoulenes and sesquifenchene), valeranone cryptomeridiol, maaliol, xanthorrhizzol and patchouli alcohol (Houghton, 1999b). Napthoic acid derivative, 4-methoxy-8-pentyl-1-naphthoic acid, acevaltrate, isovaleroxyhydroxyl didrovaltrate, didovaltrate and methyl eicosanoate were isolated from the rhizome of the species using HPLC (Pande and Shukla, 1994). Leaf and root oil of *V. jatamansi* analysed by GC and GC/MS revealed about 20-23 compounds (Mathela et al., 2005). Maaliol (39.2%) and 3-methylvaleric acid (26.5%) were the major constituents of the leaf oil, however, maaliol (64.3%) and β-gurjunene (7.2%) was found maximum in root oil. GC/MS and NMR studies of root oil of *V. jatamansi* showed two chemo types, Type-1 was characterized by the presence of maaliol (64.3%), viridiflorol (7.2%) and sesquiterpene hydrocarbon (19.2%) while Type-2 contained patchouli alcohol (40.2%), viridiflorol (5.2%), 8-acetoxy-patchoulialcohol (4.5%) and sesquiterpene hydrocarbon (34.5%). Viridiflorol and 8-acetoxy-patchouli alcohol were newly isolated compounds of *V. jatamansi* (Mathela et al., 2005).
More recently, assessment of essential oils, total phenolic, flavonoids, tannin content and antioxidant activity in aerial and root parts of wild and cultivated plants of *V. jatamansi* has exhibited significantly higher total phenols in cultivated individuals as compared to the wild ones (Bhatt et al., 2012). Considerably higher amount of patchouli alcohol was detected in the samples of planted source as compared to wild. Significantly higher total phenols, flavonoids and antioxidant activity have been detected in the aerial parts as compared to roots (Bhatt et al., 2012). Various secondary metabolites like total phenolics, flavonoids content and antioxidant activity were analyzed in *V. jatamansi* (Kalim et al., 2010). Studies on seasonal variation in plant growth, valepotriates content and essential oil in rhizomes of *V. jatamansi* collected from different conditions have revealed variation in content. For example, fresh weight, root length (July, August), valepotriates (January, October, November) and essential oil content (May) were significantly higher during these months (Singh et al., 2010).
<table>
<thead>
<tr>
<th>S. No</th>
<th>Group</th>
<th>Compounds</th>
<th>Major compound</th>
<th>Activity</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Sesquiterpenoids</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Fatty acids and their esters</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Alkaloids</td>
<td>Valerenone</td>
<td>Valeranone</td>
<td>Sedative</td>
<td>Arora and Arora, 1963, Nadkarni, 1976, Bos et al., 1996</td>
</tr>
<tr>
<td>3</td>
<td>Flavonoids</td>
<td>Linarin- isovalerianate, 6- methylapigenin, 2 S (-) hesiperdin</td>
<td>6- methylapigenin, 2 S (-) hesiperdin</td>
<td>Sedative and sleep enhancing</td>
<td>Thies, 1968, Marder et al., 2003</td>
</tr>
<tr>
<td>4</td>
<td>Bakkenolide-type sesquiterpenoid</td>
<td></td>
<td>Valerilactones A (1) &amp; Valerilactones A (1)</td>
<td>Neuroprotective effect</td>
<td>Xu et al., 2011</td>
</tr>
</tbody>
</table>
Plate 2.1

Major active constituents in *V. jatamansi* (Source: http://www.ncbi.nlm.nih.gov/structure)

**Valeranone**
Also known as: 2-Pentenoic acid, 3-(2,4,5,6,7,7a-hexahydro-3,7-dimethyl-1H-inden-4-yl)-2-methyl-, (4S-(4α(E),7β,7aα(alpha)))+(E)-, 3569-10-6.
Ambotz609870
Molecular formula: C_{15}H_{22}O_{2}
Molecular weight: 234.33398

**Alpha-Kessyl alcohol**
Also known as: 2-Cetyl-4,8,12-trimethyl-2,6,10,14-tetraoxa-1,15-pentadecane, 2-Cetyl-4,8,12-trimethyl-2,6,10,14-tetraoxa-1,15-pentadecane, 15290-63-1
Molecular formula: C_{15}H_{26}O_{2}
Molecular weight: 238.36574

**Patchouli alcohol**
Also known as: Patchoulool, patchoulanol, patchouli camphor, Patchoulic alcohol, (E)-patchoulol, (E)-patchool alcohol, CHEBI:7940
Molecular formula: C_{15}H_{26}O
Molecular weight: 222.36634

**Valeranone**
Also known as: Yatamanon, (4α,7β,8αalpha)-Octahydro-4α,8α-dimethyl-7-(1-methylethyl)-1H-naphthalen-9(1H)-one, 55528-90-0, ACI577JT, LS-95165, C168313
Molecular Formula: C_{15}H_{26}O
Molecular Weight: 222.36634
Valtrate
Also known as: Valpotriate, Bakdrisdon, Halazulione B, Valtrate [German], Valtratum [INN-Latin], Valtrato [INN-Spanish], CCRIS 5795, EINECS 242-174-2
Molecular Formula: C_{22}H_{36}O_{6}
Molecular Weight: 422.4688

Acetovlate
Also known as: Acetoxyvaltrate, Acetovaltrate, Acetvaltratum, Acetvaltrat, Acetylxyvalpotriate, Acetvaltratum [INN-Latin]
Molecular Formula: C_{22}H_{36}O_{10}
Molecular Weight: 489.5048

Didrovaltrate
Also known as: Didrovaltratum, Dihydroisovaltrate, Isovaltrate, dihydro-, Dihydrovaltrate, Isovalpotriate, dihydro-, Dihydroisovaltratum
Molecular Formula: C_{22}H_{36}O_{6}
Molecular Weight: 424.4668

Isovaltrate
Molecular Formula: C_{22}H_{36}O_{6}
Molecular Weight: 422.4688
1-Homoeacaualtrate

Also known as: AC1LCUAQ
Molecular Formula: C_{25}H_{34}O_{10}
Molecular Weight: 494.5314

Baldral

Also known as: CCRIS 2663, (7-Formyl)cyclopenta[c]pyran-4-yl)methyl acetate, BRN 5942017, 4-(Hydroxymethyl)cyclopenta[c]pyran-7-carboxaldehyde acetate
Molecular Formula: C_{12}H_{16}O_{4}
Molecular Weight: 218.2054

Valerosidate

Also known as: Valerosidatum, MEGexp0_000855, AConl_000663, NCGC00169481-01, C17068, BRD-K09737252-001-006
Molecular Formula: C_{27}H_{34}O_{11}
Molecular Weight: 462.4880
**6-methylapigenin**

Also known as: 5,7,4'-Trihydroxy-6-methylflavone, LMPK12110418
Molecular Formula: \( \text{C}_{16}\text{H}_{12}\text{O}_{7} \)
Molecular Weight: 284.2634

**Hesperidin**

Also known as: Citrinin, Hesperidine, Hesperidoside, Hesperitabs, Hesperitin-rutinoside, Hesperidin, (2S)-Hesperetin 7-rutinoside, USAF CF-3
Molecular Formula: \( \text{C}_{27}\text{H}_{34}\text{O}_{15} \)
Molecular Weight: 610.5605

**Linarin**

Also known as: 480-36-4, Acaciin, Acacetin-7-O-rutinoside, ACINSV1Q, Bio-0591, CHEMBL509502, MolPort-000-775-867, EINECS 207-547-6, ZINC04349491
Molecular Formula: \( \text{C}_{28}\text{H}_{32}\text{O}_{14} \)
Molecular Weight: 592.5452
2.2. Ethnopharmacological uses

Since long, *V. jatamansi* is widely known as an aromatic, stimulant, carminative, and antispasmodic, used in Ayurvedic medicine especially in the preparation of Sudarshan churan, Darsan gaylep, Papalyasava, etc (Parkash, 1999). The species is also used in the treatment of epilepsy and hysteria. Powdered drug, mixed with sugar is used in urinary troubles (Singh and Ali, 1998; Sharma, 2003). The dry roots/rhizomes of the plant are used to remove foul odor of mouth caused by tooth trouble (http://www.sdpi.org.com). Crushed leaves of the plant are rubbed in the forehead during extreme headache (Chevallier, 1999; Bhattacharjee, 2008). The roots/rhizomes are employed for the treatment of epilistic fits, head troubles, eye troubles, blood related diseases, diseases of liver, spleen, kidney ulcers, wounds, cardiac debility, dry cough asthma, chronic and intermittent fever (Awan, 1990; Prakash, 1999). Dry rhizomes of *V. jatamansi* are used in the preparation of perfumes and as incense (Bhattcharjee, 2008). Valeriana is also considered as a sacred plant in some part of India (Uniyal and Issar, 1967). As an ingredient, the species has been reported to be used in 39 Ayurvedic formulations (Table 2.2). Furthermore, the species is known to cure obesity, skin diseases, insanity, epilepsy and snake poisoning. The whole plant is used for nervous debility, as a hypnotic and in the treatment of spastic disorders. Clinical and animal studies have proved the central nervous system depressant effect for *Valeriana* species (Houghton, 1999a, Herrera – Arelland et al., 2001). Essential oil and extract are used in flavor, pharmaceutical and fragrance industries especially for flavoring tobacco, honey and root beer, etc., (Sah et al., 2010). The plant is widely known for its use in anxiety, insomnia, epilepsy, failing reflexes, hysteria, neurosis, sciatica, tranquilizer, emmenagogue (Nadkarni, 1976; Baquar, 1989), diuretic (Said, 1970) and as hepatoprotective agent (Awan, 1990). Various herbal preparations of the plant, for oral administration, have been used traditionally in the treatment of diarrohea (Awan, 1990), gastropasms (Kapoor, 1990) and hypertension (Chevallier, 1996). The diverse ethanopharmacological uses of the species are presented in Figure 2.1.
**Figure 2.1** Diverse ethnopharmacological uses reported for *V. jatamansi*

**Table 2.2** A list of some of the Ayurvedic preparations consisting *V. jatamansi* (Source: Prakash, 1999; Rawat and Vashistha, 2011)

<table>
<thead>
<tr>
<th>S. No</th>
<th>Preparation</th>
<th>S. No</th>
<th>Preparation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ablan liquid</td>
<td>21</td>
<td>Hormogynon</td>
</tr>
<tr>
<td>2</td>
<td>Alterix cordial</td>
<td>22</td>
<td>Mendo-Ashoka</td>
</tr>
<tr>
<td>3</td>
<td>Argosedine</td>
<td>23</td>
<td>Mendo-Serpentina</td>
</tr>
<tr>
<td>4</td>
<td>Ashoka Alters</td>
<td>24</td>
<td>Mendo-Valerian</td>
</tr>
<tr>
<td>5</td>
<td>Ashoka Cordial Co.</td>
<td>25</td>
<td>Menofel</td>
</tr>
<tr>
<td>6</td>
<td>Ashokavin</td>
<td>26</td>
<td>Neo-cardial liquid</td>
</tr>
<tr>
<td>7</td>
<td>Ashoka ovarian</td>
<td>27</td>
<td>Neuro-cardine</td>
</tr>
<tr>
<td>8</td>
<td>Bassidyle-F</td>
<td>28</td>
<td>Nervoplex mondo-valerian</td>
</tr>
<tr>
<td>9</td>
<td>Bromolax</td>
<td>29</td>
<td>Nuroval</td>
</tr>
<tr>
<td>10</td>
<td>Bromosedeon</td>
<td>30</td>
<td>Pancordial</td>
</tr>
<tr>
<td>11</td>
<td>Bromoserpentine</td>
<td>31</td>
<td>Pertussis syrum</td>
</tr>
<tr>
<td>12</td>
<td>Bromo-Valerian elixir</td>
<td>32</td>
<td>Ralbrom</td>
</tr>
<tr>
<td>13</td>
<td>Cynotone</td>
<td>33</td>
<td>Sedivel</td>
</tr>
<tr>
<td>14</td>
<td>Chandana lodhrasava</td>
<td>34</td>
<td>Sumenta</td>
</tr>
<tr>
<td>15</td>
<td>Chandrasekar Ras</td>
<td>35</td>
<td>Sero Bromides</td>
</tr>
<tr>
<td>16</td>
<td>Elixir- Bromoval</td>
<td>36</td>
<td>Tagardi Kwath</td>
</tr>
<tr>
<td>17</td>
<td>Elixir- Valerian brom</td>
<td>37</td>
<td>Uteronol</td>
</tr>
<tr>
<td>18</td>
<td>Elixir- Valerobrom</td>
<td>38</td>
<td>Valerian Bromide</td>
</tr>
<tr>
<td>19</td>
<td>Eutrine</td>
<td>39</td>
<td>Velobrom</td>
</tr>
<tr>
<td>20</td>
<td>Gynedol</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
In Nepal, decoction of the drug has been reported to be given to mothers after parturition, probably as a sedative (Sah et al., 2010). The species is considerably well known for its traditional use in inflammatory conditions such as scorpion stings and jaundice (Nadkarni, 1976). Besides, many other products from different parts of the plant have already been commercialized (Table 2.3).

Table 2.3 A list of available products in the market with *V. jatamansi* as an ingredient (- not known)

<table>
<thead>
<tr>
<th>S. No</th>
<th>Product</th>
<th>Amount of <em>V. jatamansi</em></th>
<th>Property</th>
<th>Company</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Anxocare</td>
<td>5 mg</td>
<td>Reduction Aggression</td>
<td>-</td>
<td>Umesh and Suryanarayana, 2000</td>
</tr>
<tr>
<td>2</td>
<td>Anti-Wrinkle cream</td>
<td>-</td>
<td>Anti-Wrinkling</td>
<td>-</td>
<td>Ravichandran et al., 2005</td>
</tr>
<tr>
<td>3</td>
<td>Lukol</td>
<td>6 mg</td>
<td>Leucorrhoea</td>
<td>Himalaya Drug Co. India</td>
<td>Gupta and Bhanot, 1973</td>
</tr>
<tr>
<td>4</td>
<td>Mentat tablet</td>
<td>50 mg</td>
<td>Attention deficit hyperactivity disorder</td>
<td>-</td>
<td>Kalra et al., 2002</td>
</tr>
<tr>
<td>5</td>
<td>Valmane</td>
<td>Whole</td>
<td>Insomnia</td>
<td>Whitehall Pharmaceutical</td>
<td>Dalva et al., 2002</td>
</tr>
<tr>
<td>6</td>
<td>Iymem capsule</td>
<td>20 mg</td>
<td>Mental irritability, Anxiety-Neurosis, Insomnia</td>
<td>Osho Pharma Pvt. Ltd.</td>
<td>Osho Pharma Pvt. Ltd. Prize list</td>
</tr>
<tr>
<td>7</td>
<td>Iymem Syrup</td>
<td>15 mg</td>
<td>Mental irritability, Anxiety-Neurosis, Insomnia</td>
<td>Osho Pharma Pvt. Ltd.</td>
<td>Osho Pharma Pvt. Ltd. Prize list</td>
</tr>
<tr>
<td>8</td>
<td>Tagar capsule</td>
<td>-</td>
<td>Lowering blood pressure</td>
<td>Asian Medicine and More</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td>Single herb capsules</td>
<td>-</td>
<td>Reduces Anxiety Stress and Weakness</td>
<td>Medicca (India) Private Limited</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>Fretnil tablet</td>
<td>-</td>
<td>Anxiety, Mental irritability</td>
<td>Charak Pharmaceuticals</td>
<td><a href="http://www.biogetica.com/clinical_research/sumenta_tablet.pdf">http://www.biogetica.com/clinical_research/sumenta_tablet.pdf</a></td>
</tr>
<tr>
<td>11</td>
<td>Sumenta tablet</td>
<td>20 mg</td>
<td>Anxiety, Mental irritability</td>
<td>-</td>
<td></td>
</tr>
</tbody>
</table>
2.3. Pharmacological uses

2.3.1. Stress and Nervous disorder

Plant extract of *V. jatamansi* has been reported to attenuate stress, anxiety and depression (Bhattacharya et al., 2007). Further, clinical trials have confirmed that the root extract decreases sleep latency, improves sleep quality and, therefore, considered useful in treating anxiety and insomnia (Leathwood and Chauffard, 1983). The species has also been found beneficial for cerebro-spinal system, hypochondriasis, insomnia, migraines, nervous unrest, nervous tension, neuralgia and neuroasthemia (Cionga 1961; Bhatt et al., 2013). It has been established that the rhizome oil can be used for spinal rubs in diseases where the spinal cords need lessened sensibility to pain and stimulation (Morazzoni and Bombardelli, 1995). Valerian is reported for depressant action on the Central Nervous System (CNS) and antispasmodic activity (Cionga, 1961). The extract showed the depressed CNS activity in mice by oral administration (Veith et al., 1986). The valiracyl exerted a pronounced neurotropic effect (Dunaev et al., 1987), and suppressed the orientation reflex of animals, decreased a spontaneous and caffeine-stimulated motor activity, potentiated and prolonged the action of barbiturates, significantly reduced aggressiveness of animals, decreased sensitivity to the convulsant effects of corasil and thiosemicarbazide, produced the antihypoxic and mild myorelaxant actions (Dunaev et al., 1987). It was reported that the neurotropic effects of valiracyl were related to the increased level of the GABA inhibition mediator and decreased intensity of bioenergetic processes in the brain (Dunaev et al., 1987). The effect of chlorophyll and aqueous extract on ischemia and reperfusion-induced cerebral injury was examined and the extract markedly attenuated in terms of decreased infarct size, increase in short term memory, motor co-ordination, and lateral push response (Rehni et al., 2007). The species has also been reported as a psychopharmacological agent and a natural source of valepotriates (Mishra, 2004). Use of *V. jatamansi* in several of pharmacological applications has been depicted (Figure 2.2).
2.3.2. Gastrointestinal and cardiovascular disorder

Studies exhibits various uses of *V. jatamansi* for treating different gastrointestinal disorder such as diarrhea, abdominal spasm, diverticulitis, irritable bowel, nervous dyspepsia, stomach cramp and stimulates digestion (Houghton, 1999b). The valepotriates hydrolyze rapidly and metabolize in gastrointestinal tract to yield the breakdown products baldrinal, homobaldrinal, decylbaldrinal and valtroxal consisting of an unsaturated version of ring skeleton (Thies, 1968; Schneider and Willems, 1982). Reports have indicated that these compounds are partially responsible for sedative activity as they get well absorbed and have shown to significantly decrease motality in mice (Schneider and Willems, 1982). *V. jatamansi* extract is also used for lowering blood pressure and strengthening and palpitations of the heart (Morazzoni and Bombardelli, 1995).

2.3.3. Sleeping and tranquillizing effect

Valerian is reported to be effective in treating various sleep disorders in human. The components of valerian include valerenic acids such as monoterpenes, sesquiterpenes and
iridoid glycosides, which are responsible for sedative and antispasmodic activity. As such, in the volatile oil component, valerian sesquiterpenes are responsible for biological activity (Houghton, 1999a). Pharmacological screening of valerenal and some other components showed that the sedative action can be attributed to the essential oil and valepotriates fractions (Wagner et al., 1980; Hendricks et al., 1981). It is known that valerenic acid inhibits the enzyme system responsible for central catabolism of GABA (Riedel et al., 1982), and the valerian extract releases [3H] GABA by reversal of the GABA carrier, which is Na (+) dependent and Ca (2+)-independent (Santos et al., 1994). This increase in [3H] GABA release appears to be independent from Na (+)-K (+)-ATPase activity and the membrane potential. The evaluation of commercially available root extract of the species has revealed pronounced sedative properties in mice with respect to a reduction in motility and an increase in the thiopental sleeping-time (Leuschner et al., 1993). A direct comparison with diazepam and chlorpromazine has revealed a moderate sedative activity for the tested extract (Leuschner et al., 1993). 6-Methylapigenin (4’, 5, 7-dihydroxy-6-methylflavone or MA) isolated from V. jatamansi as a new flavonoid compound is a benzodiazepine binding site (BDZ-bs) ligand and found to possess anxiolytic property while another flavanone glycoside 2S (−) hesperidin isolated from this species was able to potentiate the sleep enhancing properties (Marder et al., 2003). MA functioned as a competitive ligand for the brain GABA_A receptors (Wasowski et al., 2002). Several flavonoid glycosides including goodyerin (Du et al., 2002), linarin and hesperidin (Fernandez et al., 2004) have been reported sedative and anticonvulsant agents likely to interact with GABA_A receptors. Studies on herbal formulations named Mamsyadi vati, which contains V. jatamansi, Nardostachys jatamansi, Withania somnifera, and Convolvulus pluricaulis along with Panchakarma (Shirodhara) have shown a significant improvement in the duration of sleep, quality of sleep, and mood upon awakening (Narayan et al., 2000). Another study on V. jatamansi has established that flavonoids might be responsible for sleep-enhancing properties (Sharma et al., 2007). V. jatamansi root extract showed sleep quality improvement and modulates brain monoamine level in rats (Sahu et al., 2012); it revealed that the water extract has a sleep quality improving effect which may be dependent upon levels of monoamines in cortex and brainstem.
2.3.4. **Antidepressant Activity**

Depressive disorder is a common affliction; however, therapeutic agents are currently available for treating depression. The rate of success in depressed patients is about 65-70% but serious side effect may limit treatment strategies (Keith and Matthew, 1993). In this context, medicinal plants have been found beneficial. For example, clinical study on *V. jatamansi* extract has reported to attenuate stress, anxiety and improvement in the symptoms of depression (Bhattacharya et al., 2007). *V. jatamansi* extract significantly reduced locomotor activity at 200 mg/kg in the tail suspension test and has a negative functional interaction with antidepressant like effects. The methanolic and aqueous ethanolic extracts of the *V. jatamansi* have indicated that antidepressant like action of this plant was not contingent upon its terpenoids (Subhan et al., 2010). However, a considerable degree of antilocomotor activity was reported at the higher doses of terpenoids in tail swim test or forced swim test (Subhan et al., 2010).

2.3.5. **Spasmolytic activity**

The valepotriates (valtrate and didrovaltrate) of the species have long been reported to exert a spasmolytic effect (Wagner et al., 1980). The commercial mixture of valepotriates was found effective compared to the same dose of papaverine (Gilani et al., 2005). Antispasmodic and blood pressure lowering activities of the root part indicated that these activities are possibly mediated through activation of K+ (ATP) channel thereby justified its use in gastrointestinal and cardiovascular disorders (Gilani et al., 2005). The species is reported to be beneficial in relaxing female menstrual cramps, aid liver functions, head congestion, muscle spasm, relieve pain, etc. (Morazzoni and Bombardelli, 1995).

2.3.6. **Antimicrobial activity**

Studies on *V. jatamansi* essential oil have exhibited antibacterial activity against large number of pathogenic bacteria and antifungal activity against fungal pathogens (Suri and Thind, 1978; Thind and Suri, 1979; Girgune et al., 1980). Antimicrobial activity in the extract of *V. jatamansi* in different solvents system (methanol, chloroform, hexane and water) was found more effective than positive control -Ampicillin and Erythromycin (Sati et al., 2011). Antibacterial activity of the ethanolic extracts of *V. jatamansi* against five
bacterial species namely, *Escherichia coli, Salmonella typhi, Proteus vulgaris, Pseudomonas aeruginosa* and gram-positive *Staphylococcus aureus* was also evaluated and found to vary in activity against these species (Jan et al., 2012).

### 2.3.7. Other uses

Among others, *V. jatamansi* is used in the preparation of polyherbal combination of anti-wrinkle cream (Ravichandran et al., 2005) and forms ingredient of herbal antidepressant formulation ‘*Sumenta*’ (Prakash, 1999). Dried leaves of *V. jatamansi* have been reported for central analgesic activity (Shrivastava and Sisodia, 1970). Didrovaltrate present in *V. jatamansi* is reported to inhibit alternative synthesis in the complement system of the serum and its possible use in some autoimmune diseases (Houghton 1999b; Baibado and Cheung, 2011). As such, the anti-inflammatory activity in methanolic and ethanolic extract of the species has been reported (Subhan et al., 2007) and are known to inhibit inflammation mediators such as histamine, serotonin, prostaglandins and bradykinins, etc. (Vinegar et al., 1969). The presence of high amount of flavonoids (Falogun et al., 2003) and tannins (Starec et al., 1988) enhance anti-inflammatory activity. The root extracts of the species exhibit larvicidal and adulticidal activity against different mosquito species (Dua et al., 2008). *In vitro* anthelmentic activity of the rhizome of *V. jatamansi* against adult Indian earthworms (*Pheretima posthuma*) has been reported (Potdar et al., 2011). Further, root extract of the species was found beneficial against leishmania; the methanol and chloroform extracts showed activity against *Leishmania donovani* and *Leishmania major* (Ghosh et al., 2011).

### 2.4. Propagation

*V. jatamansi* generally propagated through seeds and vegetatively using root segments. The seeds can be sown at various times of year and by different methods. *In vitro* propagation of *V. jatamansi* has been reported using shoot tip and axillary bud explants (Mathur et al., 1988). The highest plant formation frequency (96-100%) of encapsulated buds of *V. jatamansi* was observed on 0.7% agar, both with or without MS nutrients, while non-encapsulated buds under the same conditions failed to grow and died following yellowing (Mathur et al., 1989). Murashige and Skoog (1962) medium supplemented with
Kinetin/BAP (5.0 mg/L) and IAA (1 mg/L) induced optimal growth of shoots within 6-8 days from both apical and axillary bud explants. Shoot generation via callus phase occurred in a medium containing 1.0 mg/L Kinetin and 0.25 mg/L NAA, and medium supplemented with 5.0 mg/L Kinetin and 1.0 mg/L IAA was considered best for complete plantlet formation. Nearly a thousand regenerated plants from callus were successfully transferred to the field for hardening (Mathur and Ahuja, 1991). Subsequent studies by other workers reported hundred percent survival during hardening and field establishment of tissue cultured plants of this species (Kaur et al., 1999). However, no reports are available on tissue culture of *V. himalayana*. Valepotriates were isolated from colchicin-treated tissue cultures of *V. jatamansi* and their structure elucidated by means of (13) C-NMR spectra (Becker et al., 1984). Hairy roots of *V. jatamansi* were obtained following co-cultivation of detached leaf explants with *Agrobacterium rhizogenes* strains A4 and LBA 9402; the A4 strain appeared to be better in terms of both relative rate of hairy root formation and growth of the respective hairy root line (Banerjee et al., 1998). In terms of the production of total as well as individual valepotriates, the LBA 9402 induced hairy root line appeared to be a better performer than the A4 induced one (Banerjee et al., 1998).

2.5. Cultivation practices

Wild plants vary in quality and consistency which seriously hamper economic returns; therefore, cultivation has advantages over collection from wild. Yield of dry roots, root/shoot ratio and production of roots in the whole plant biomass have been reported to increase with advancement in age after transplanting (Singh et al., 2000). Overhead shade, and use of nylon nets are reported to be good for better growth as compared to natural shade *vis-a-vis* crop raised in the open field as well valepotriates content (Singh et al., 2000). Studies have revealed that the time and form of direct seed sowing is determined by local climatic condition. In case of nursery, sowing should begin between June and early August and the seedling can be planted at the end of September. Valerian which has been planted in the autumn can be harvested in the next year in the October. A marked difference in terms of percentage of patchouli alcohol, ar-curcumene, betapatchoulene and gamma-patchoulene between the cultivated and wild plants were reported (Singh et al.,
Propagation by vegetative means has been applied only in case of varieties which were able to develop large amounts of stolons. Impact of transplantation time on the growth and yield of *V. jatamansi* has been examined (Singh et al., 2000); maximum plant yield in terms of height, aerial biomass, underground biomass, rhizome yield, and root yield was obtained after 9 months of transplantation.

### 2.6. Post harvesting handling

The powdered valerian roots have been reported to undergo rapid loss in essential oil and active content, therefore, optimization of suitable storage condition have attracted studies. The concentration of valerenic acids in roots was reported to enhance markedly from the dormant stage to a peak in the growing vegetative growth stage (spring) and then fall substantially in the senescence stage (Houghton, 1999a). It has been suggested that the crude drug should be stored in a well closed container, protected from light (European pharmacopeia 2nd edition 1993). Similarly, storage temperature has also been reported important and higher temperature (>40°C) causes decomposition of the valepotriates thereby yielding valeric and isovaleric acids. The characteristic odor of these acids signals to improperly dried or stored material. Additionally, hydroxyvalerenic acid may be considered to be a decomposition product of acetoxyvalerenic acid when drug is stored at a too high humidity (Freytag, 1983; Bos et al., 1996). Valerenic acid and its derivatives are relatively stable in both plant material and drug. These compounds are however, degraded after storage of several weeks (Houghton, 1999a).

### 2.7. Genetic and Phylogenetic studies

Vary few studies are available on the genetic characterization, genetic diversity and phylogenetic analysis of target species. Genetic characterization of 7 morphotypes of *V. jatamansi* was performed using RAPD markers (Singh, 2007). Similarly, analysis of six populations of *V. jatamansi* was conducted using AFLP markers (Rajkumar et al., 2011) and reported to have high within and low among population variations. Assessment of genetic diversity in *V. jatamansi* and classification of the germplasm using RAPD markers have also been reported (Kumar et al., 2012). Phylogeny of Valerinaceae (Dipsacales) was
investigated using nuclear and chloroplast DNA sequences data (Bell, 2004). Phylogeny of Valerianaceae including *V. jatamansi* and various other species of this family based on *matK* and ITS markers, with reference to *matK* individual polymorphism was also performed (Hidalgo et al., 2004). Further, the ploidy screening, detection of mixoploidy and aneuploidy, cell cycle analysis, assessment of the degree of polysomaty, determination of reproductive pathway, and estimation of DNA amount or genome size variation in *V. jatamansi* were analyzed; the study reported 32 Chromosomes (2n), 2C DNA Content (3.08 pg) and 2C DNA Content (3012.24 Mbp) (Subramani et al., 2011; Nag et al., 2011).

### 2.8. Health Concern

Reports suggest that prolonged use and higher dose of Valerian can cause mental depression, increase tolerance and have serious side effects. Excess amount could be stimulating rather than a relaxing effect, heaviness and pain in the head stupor (Morazzoni and Bombardelli, 1995). High short-term doses of Valerian have been reported to cause headaches, muscle spasms, dizziness, digestive upsets, insomnia, and confusion (Chan, 1998). Higher dosage of the species could experience longer sleep than usual (Chan et al., 1995). Use of Valerian together with alcoholic beverages, benzo- diazepines, barbiturates, or antihistamines has been reported for mild sedative effect. Some components of Valerian are metabolized in the liver, therefore, excess content could damage liver. It has potential to interact with liver metabolized prescription medicines (Willey 1995; Mullins, 1998). Similarly, cytotoxicity and mutagenicity of valepotriates have been described in *in vitro* cell system. The compounds have shown to inhibit DNA and protein synthesis in *in vitro* cultured mammalian cells (Bounthanht et al., 1981; Von der hude et al., 1985; Hansel, 1990; 1992; Keochanthala- Bounthanht et al., 1993). Therefore, it is necessary to check the extent of valepotriates as health hazard and to find out the particular valepotriates responsible for it. Valerian has also been reported as not safe for pregnant or nursing women and children (Houghton, 1999a).

### 2.9. Gap areas and direction for future research

Towards further strengthening the review of information study was conducted in four most authentic publicly available databases like Ingenta, Science direct, Agricola and Pubmed to
find out the most popular research area and year wise trends of publication in *V. jatamansi*. Survey revealed a total of 77 articles were published in *V. jatamansi* since 1971 to 2011, of which maximum articles (48) appeared during 2000-2011 (Figure 2.3A). This depicts growing research interest on the target species. While considering the subject area, maximum attention (44%) has gone to Pharmacology followed by Phytochemistry (Figure 2.3 B). This indicates that the species contain active phytochemical content which has effective pharmacological activity. Therefore, it can be used in the preparation of several medicines. Only 5% of the publications were published on the molecular biology aspect, thereby suggesting the need for more studies on this aspect so as to facilitate development of proper strategies for the conservation. Foregoing account clearly indicates that in spite of large number of studies conducted on target species, further investigation on different aspects of phytochemical and genetic diversity are essentially needed. Particularly, in *V. himalayana*, lack of information in different aspects warrants detailed investigation. In general, marker assisted identification of elite individuals will be beneficial for properly harnessing commercial potential.

The review clearly reveals that morphological, phytochemical and genetic diversity studies across multilocational populations have not been performed in *Valeriana* species. Likewise, DNA damage inhibition efficiency of plant extract has not been evaluated. Further the impact of habitat types and altitudinal range on species performance remains uninvestigated. Also, microsatellite marker development and comparative assessment of the two species, namely, *V. jatamansi* and *V. himalayana* have not been taken into the consideration. Realizing these gaps, the present study attempted to fill in these areas through systematic investigations.
Figure 2.3 Distribution of published articles on *V. jatamansi*: A - Publications trends over the years; B - Publication trends across subject area.

(A) Figure showing the distribution of published articles on *V. jatamansi* with labels for Phcytochemistry, Tissue culture, Pharmacology, Microbiology, and Other, each with a percentage.
