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MEDICINAL PLANTS DENSITY ALONG AN ALTITUDINAL GRADIENT IN AND AROUND AYUBIA NATIONAL PARK

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Abstract

Medicinal plants are an essential source of livelihood for many rural populations and are currently facing several threats of extinction in temperate Himalaya, such as excessive grazing and collection along altitudinal gradients. The present study was designed to investigate the species density of medicinal plants at different mid-altitude levels (2200, 2300, 2400, and 2500 m above the sea level (a.s.l.) between two forest-use types and to examine the possible association between medicinal plant densities and forest-stand structural variables along the altitudinal gradient. Factorial design analysis of variance showed that the densities of all medicinal plants differed significantly between the forest-use types ($p<0.00$) and elevation ($p<0.00$). Moreover, a significant interaction ($p<0.04$) was also observed between the forest-use types and elevation. In the old-growth forest, density of medicinal plants was 290/40 m^2 at the higher altitude (2500 m a.s.l.), approximately 1.5-fold less than the 475/40 m^2 density observed at lower altitude (2200 m a.s.l.). However, in derived woodland, density of medicinal plants at higher altitude was approximately 4-fold less than that at the lower altitude. At these altitudinal levels, medicinal plants densities, such as *Valeriana jatamansi*, were significantly higher under old-growth forest compared to derived woodland, where they were almost nonexistent. A rapid vulnerability assessment has also shown that *Valeriana jatamansi* and *Viola canescens* were highly vulnerable species. Litter cover was the influential variable that was most likely related to medicinal plant density. In conclusion, abundance of medicinal plants decreased along mid-altitude levels in both of the forest-use types. However, this decrease was extremely marked in the derived woodland, and this decline may be due to human activity. Hence, these factors must be considered in future studies to suggest protective measures that can be applied along altitudinal gradients in disturbed landscapes.

Key words: Ethnobotany, Himalaya, Forest-use types, Grazing, Livelihood.

Introduction

Pakistan has a rich diversity of medicinal plant species that are distributed over a wide altitudinal range (sea level to 8611 m) (Habib *et al*., 2011; Haq *et al*., 2010); their wide distribution thus represents different climatic zones and unique biodiversity. More than 600 medicinal plant species have been reported from the country (Shinwari, 2010). The Himalayas are best known for high-altitude temperate forests carrying diversity of plant species that are closely associated with the livelihood of local inhabitants. Ecologists have attempted to understand the variation in species diversity along altitudinal gradients in mountainous ecosystems (Grytnes & Veetas, 2002), and altitude has been identified as a limiting factor in plant species diversity (Chandra *et al*., 2010). Hence, the Himalayas can serve as an excellent system for the evaluation of medicinal plant abundance along an altitudinal gradient.

Despite the significance of understory medicinal plant species for rural livelihood, the diversity and abundance of these plants are threatened by various natural and anthropogenic factors (Gilliam, 2007; Wyatt & Silman, 2010). Altitude and disturbance from human settlements have a linear relationship with vegetation attributes such as species richness, diversity (Schuster & Diekmann, 2005) and species cover (Adnan & Hölscher, 2010). Shaheen *et al*. (2011) have observed that overgrazing can reduce vegetation cover, diversity and productivity. In mountainous areas, low-altitude regions are affected by settlements resulted in the exploitation of forest resources. However, high-altitude regions are subjected to grazing that is intended to maintain grasslands, with most forests remaining at mid-altitudes (Austrheim & Eriksson, 2001). A hump-shaped pattern of species richness has been observed in many studies. This pattern is characterized by higher diversity at middle altitudes (Sharma *et al*., 2009; Grytnes & Veetas, 2002). According to Bravo (2009), most of the studies along altitudinal gradients have been documented in disturbed forests rather than undisturbed forests. These studies have mainly focused on tree species richness along altitudinal gradients.

The Himalayan region in northwest Pakistan is currently under severe pressure from the local population due to the extraction of forest resources such as medicinal plants. Two types of forest-uses have been differentiated by previous research (Putz & Redford, 2010). Old-growth forest denotes forest areas that experience slight or no interference from humans and have the utmost protection level by the forest department from resource exploitation. This forest type is found primarily in the National Park. This forest contains many trees having large diameter and usually closed forest canopy. Derived woodland denotes the forest that is under high grazing pressure and in which local inhabitants frequently collect medicinal plants and other non-timber forest products. This type of forest is usually adjacent to surrounding villages (outside the National Park) and contributes substantially to livestock in terms of fodder. In addition, legal and illegal loggings have both occurred in the recent past. Studies have found the highest species richness at mid-altitudes (2100-2500 m) in the region (Sharma *et al*., 2009; Grytnes & Veetas, 2002). However, limited studies are available on variation in medicinal plants densities among different forest-use types specifically at middle altitudinal ranges. Therefore, the present study was designed with the following objectives: (i) to investigate the density of medicinal plants at different mid-altitude levels (2200, 2300, 2400, and 2500 m a.s.l.) in old-growth forests and derived woodlands and (ii) to examine the possible relationships between forest-stand structural variables and medicinal plant densities along the altitudinal gradient.

Materials and Methods

The current study was conducted in collaboration with World Wide Fund for Nature, Pakistan (WWF-P). Representative of the organization granted permission for data collection in old-growth forest. WWF-P is already working with government forest department for the cause of conservation of study area forests. Data collection in derived woodland forest was carried out with the permission of local community representatives.

Study area: This study was conducted in Ayubia National Park (ANP) (3312 ha in area) and in its surrounding forests located in northwest Pakistan (Fig. 1). The study area is located between 33º–01' and 34º–38' N latitude and 73º–20' and 73º–30' E longitude. Altitude varies between 1220 m and 2865 m above sea level (a.s.l.). The ANP and forests surrounding it are influenced by monsoons and have a mean annual precipitation of 1500 mm. Mean annual temperature is 12°C, and snow cover predominates from November to March (Anon., 2004). The soil is often shallow, with a texture mostly loamy, and the natural vegetation in the area corresponds to Himalayan moist temperate forest. The dominant tree species in the study area are *Pinus wallichiana* A.B.Jacks., *Abies pindrow* (Royle ex D.Don) Royle, *Taxus wallichiana* Zucc., *Populus ciliata* Wall. ex Royle and *Aesculus indica* (Wall. ex. Cambess.) Hook. (Aumeeruddy *et al*., 2004). There are 12 villages in the periphery of ANP with an approximate human population of 58,000 living in 8000 households (Anon., 2004). Local community adjacent to park mostly depends on the park's resources for fodder, fuel wood, livestock grazing, timber and other non-timber forest products (NTFPs) (Aumeeruddy *et al*., 2004).

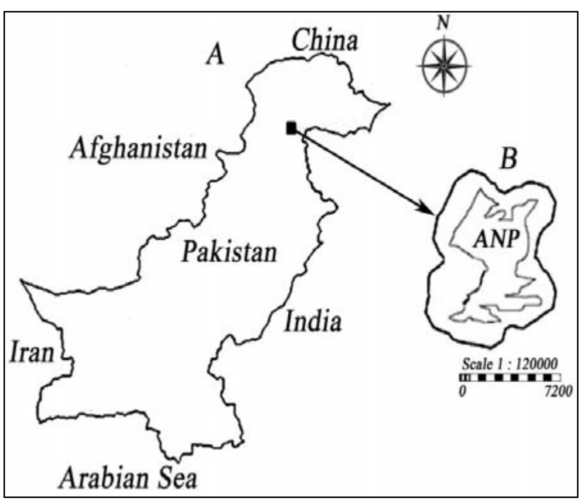

Fig. 1. Map of the study area. (A) Pakistan and study area location. (B) The study are with Ayubia National Park (ANP) border (inner boundary) and its associated forests (outer boundary).

Ethnobotanical data collection: Data on ethnobotanical uses of medicinal plants was collected in September 2012 for herb/fern species that were encountered in the ecological survey described below. A total of 60 informants (20 female and 40 male) having knowledge of medicinal plants were selected on information basis provided by community representatives. The selected informants generally ranged from 30 to 70 years of age. A structured questionnaire was designed and informants were asked about the traditional names of plants. Informants were asked about the local names of plants, plant parts used, type of disease treated, and other non-medicinal uses of plants. Scientific names and citation authors were verified using Flora of Pakistan (Ali & Qaiser, 2013) and Tropicos (2009). Medicinal plants prices were obtained from local market in the city of Abbottabad. Rapid vulnerability assessment (RVA) data for medicinal plants was collected from WWF-Pakistan reports and online literature (Sher & Hussain, 2007; Anon., 2004). RVA presents the status of threat/vulnerability (low to high vulnerability) to a particular plant species in the light of ten parameters including the habitat, part used, life form, use value, distribution, season of collection, regeneration, population size, quantity of collection, and national or international conservation status (Sher & Hussain, 2007).

During data collection, each respondent was visited or contacted at least three times to ensure the validity of the information provided by them. In case of any inconsistencies relative to the original information provided, the information was rejected and considered irrelevant. Only relevant information was subjected to further analysis. Data quality was further ensured through proper training of data collectors, identification of instances of missing information and duplication of material.

Sampling design and plot selection: Ecological data was collected from July to September 2012. Five line-transects were placed from low to high altitudes in each forest-use type (Malik, 2003). The line-transects were initiated from random sample points (Adnan & Holscher, 2011) that had previously been identified at 2200 m a.s.l. on the northern aspect. A Garmin eTrex HC series GPS was used to locate sample points in the field. Each line-transect consisted of four circular plots at four different altitudes (2200, 2300, 2400 and 2500 m a.s.l.) with a gap of 100 m elevation among them (Fig. 2A). Plots were laid down on the contour line where slope correction was applied to correct the plot area along the altitude. Each circular plot had a diameter of 35.7 m, and each plot was further divided into two parts. The plot with a radius of 17.85 m (1000 m^2 area) was used for a tree inventory, and the inner, smaller part of the same plot, with a radius of 3.2 m (40 m^2 area), was used for herb surveys (Johnsson *et al*., 1992) (Fig. 2B). In total, we sampled five line-transects in each forest-use type. Each line-transect included four plots at four fixed elevations indicating five plots per elevation.

Data collection on forest-stand structural variables and medicinal plant variables: We assessed the forest-stand structural variables including basal area, stem density, species richness, litter cover, litter thickness, soil nitrogen, potassium and phosphorus. Litter Thickness in a plot was measured in centimeters from the litter layer's top to the start of mineral soil using a vernier caliper. Percentage litter cover was assessed in a plot using visual estimation method (Leos, 2003). This procedure was performed with the help of five observers who were professional botanists and graduate students. Soil samples were collected from the center of each plot by inserting a 10-cm-longauger steel pipe in the topsoil. Soil samples were analyzed for the concentrations of nitrogen, phosphorus and potassium following standard protocols (Bremner & Mulvaney, 1982; Olsen & Sommers, 1982).

The density of medicinal plants (herb/fern species) was calculated by counting all individuals of plants species in each circular plot. The estimates of density were obtained as per the standard protocols (Curtis & McIntosh, 1951). Each variable was assessed based on the average of five plots at the same altitude in five transects of a forest-use type. Shannon–Wiener diversity index H' and Shannon evenness of medicinal plants were calculated for each plot (Magurran, 2004). Additionally, Sorensen's index was carried out to assess similarities in medicinal plant richness at a particular altitude between the two forest use-types.

Statistical analysis: Factorial design analysis of variance was applied for testing the effect of independent variables (forest-use and elevation) on dependent variables related to forest stand structure, soil and medicinal plants. Data on dependent variables was logarithmically transformed as factorial design analysis based on the assumption of normally distributed data. A Spearman correlation was calculated between the density of individual medicinal plant species and altitude for a given forest-use type. A Mann-Whitney test was also applied at the species level to medicinal plant density at any two similar altitudes between the two forest-use types. Across the two forest-use types at all altitudes, DCA (detrended correspondence analysis) was applied to identify forest tree stand structural variables, which most likely related to the medicinal plants densities. The DCA was performed using PC-ORD 5.06 (McCune & Mefford, 1999). The data compilation and analyses were performed using Microsoft Excel and SPSS version 16.0, respectively (Anon., 2007).

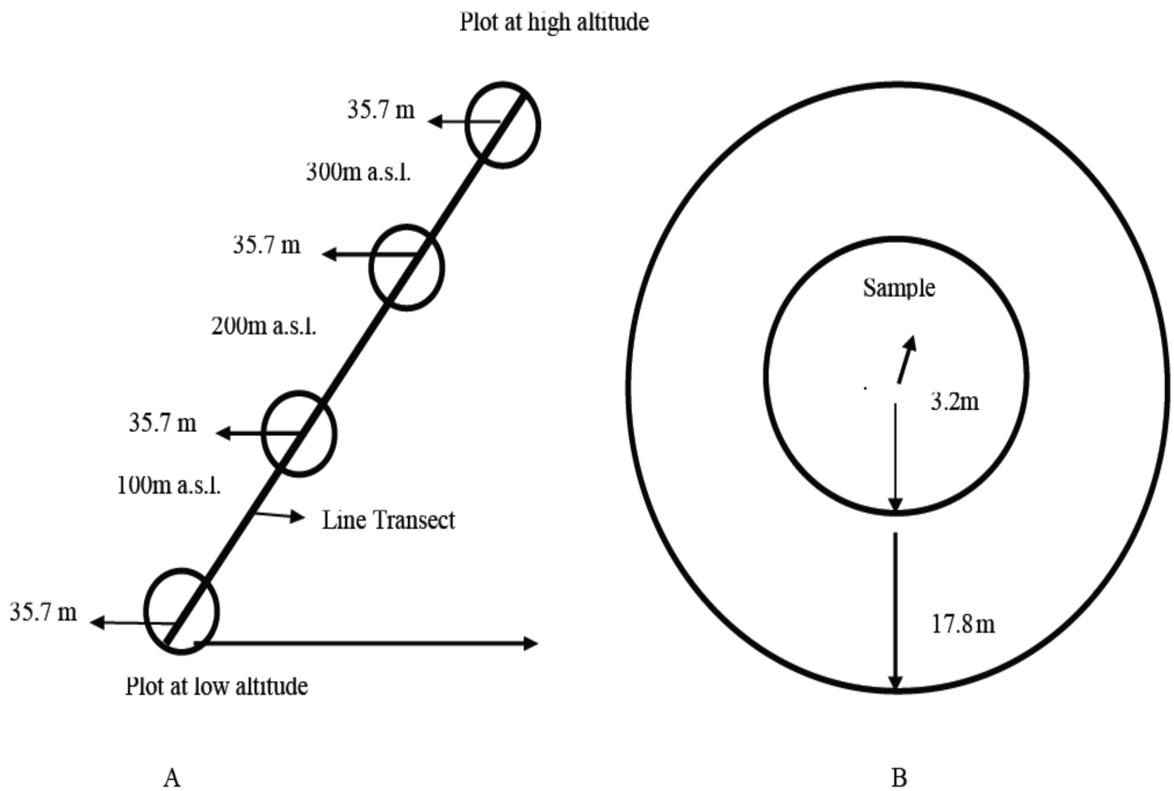

Fig. 2. Line-transect and plot design. "A" represents line-transect including the plots, from lower to higher altitudes. "B" represents a single plot with a 1000 m^2 area (17.8 m radius) for tree inventory and a 40 m^2 area (3.2 m radius) for the assessment of medicinal plants. Only one line-transect out of 10 in both forest-use types has been explained in this figure.

Table 1. Ethnomedicinal uses of plants and their conservation status. NTFPs: non-timber forest products; RVA: rapid vulnerability assessment.

Botanical names	Part used	NTFPs uses	Medicinal uses	Market price in US$/Kg	RVA (Sher& Hussain, 2007; WWF-P, 2004)
Adiantum incisum Forssk.	Whole plant	Medicinal and fodder	Respiratory infections, antipyretic and gynea	2.0	Moderately vulnerable
Agrimonia eupatoria L.	Leaves, flower	Medicinal and dye	Gastro-intestinal	0.2	Less vulnerable
Ajuga bracteosa Wall. ex Benth.	Whole plant	Medicinal	Antipyretic and gastro-intestinal	2.04	Highly vulnerable
Androsace rotundifolia Hardw.	Whole plant	Medicinal	Eczema	2.0	Moderately vulnerable
Aquilegia pubiflora Wal. ex Royle	Stem	Medicinal and fodder	Dermatitis, ulcer and gynea	0.5	Less vulnerable
Arisaema flavum (Forssk.) Schott	Roots and stem	Medicinal and insecticide	Eczema and snake bite	0.6	Moderately vulnerable
Bistorta amplexicaulis (D.Don) Greene	Leaves and roots	Medicinal and fodder	Respiratory infections, gastro-intestinal, antipyretic, gynea and antispasmodic	1.02	Moderately vulnerable
Chenopodium album L.	Leaves	Vegetable and medicinal	Gastro-intestinal and anthelmintic	0.2	Less vulnerable
Chrysanthemum leucanthemum L.	Whole plant	Fodder, medicinal and handicraft	Dermatitis	0.3	Less vulnerable
Clematis grata Wall.	Whole plant	Fodder and medicinal	Bile disorders	0.5	Less vulnerable
Dipsacus inermis Wall.	Leaves	Vegetable and medicinal	Pain and sore throat	2.0	Moderately vulnerable
Dryopteris ramose (C. Hope) C. Chr.	Leaves	Medicinal, fodder and vegetable	Tonic, gastro-intestinal, antimicrobial and anticancer	7.15	Highly vulnerable
Euphorbia wallichii Hook. f.	Whole plant	Medicinal and fodder	Eczema	3.0	Moderately vulnerable
Fragaria nubicola (Lindl. ex Hook. f.) Lacaita.	Leaves, seeds and fruit	Medicinal, fodder and vegetable	Gastro-intestinal, diuretic and anti-inflammatory	8.1	Less vulnerable
Galium aparine L.	Whole plant	Medicinal and fodder	Gastro-intestinal, antipyretic, anti-inflammatory, hepatic and diuretic	0.9	Moderately vulnerable
Hedera nepalensis K.Koch	Leaves, seeds and fruit	Medicinal and fodder	Gastro-intestinal, antipyretic and antidiabetic	5.11	Moderately vulnerable
Impatiens bicolor Royle	Seeds and fruit	Medicinal and fodder	Tonic, antipyretic and diuretic	0.3	Less vulnerable
Leucanthemum vulgare (Vaill.) Lam.	Whole plant	Fodder and medicinal	Antipyretic	0.3	Less vulnerable
Lygodium jopanicum (Thunb.) Sw.	Seeds	Medicinal	Antipyretic and ulcer	0.3	Less vulnerable
Nepeta laevigata (D.Don) Hand. –Mazz.	Leaves, seeds	Fodder and medicinal	Gastro-intestinal	1.0	Less vulnerable
Oxalis debilis Kunth	Roots	Medicinal	Edema, antipyretic, asthma, antidiabetic	0.8	Less vulnerable
Polygonatum verticilatum (L.) All.	Leaves	Medicinal and fodder	Rheumatism, gynea and aphrodisiac	8.1	Moderately vulnerable
Prunella vulgaris L.	Leaves	Medicinal	Anti-inflammatory	1.6	Less vulnerable
Ranunculus muricatus L.	Leaves	Fodder and medicinal	Respiratory infection	1.0	Less vulnerable
Rabdosia longituba (Miq.) H. Hara	Roots	Fodder and medicinal	Gastro-intestinal	0.9	Less vulnerable
Senecio salignus DC.	Leaves, roots and fruit	Medicinal and fodder	Gastro-intestinal, anti-inflammatory and snake bite	0.7	Less vulnerable
Swertia chirata Buch. –Ham. ex Wall.	Whole plant	Medicinal	Respiratory infections, eczema, antipyretic and antidiabetic	4.08	Moderately vulnerable
Urtica dioica L.	Leaves and roots	Medicinal and ethno-veterinary	Astringent and anthelmintic	1.2	Less vulnerable
Valeriana jatamansi Jones	Leaves and roots	Medicinal and fodder	Antipyretic and eczema	2.0	Highly vulnerable
Veronica laxa Benth.	Leaves	Medicinal and fodder	Respiratory infections and tonic	4.08	Moderately vulnerable
Viola canescens Wall.	Whole plant	Medicinal, fodder and vegetable	Respiratory infections	3.06	Highly vulnerable

Table 2. Forest stand structural and medicinal plant variables along the altitudinal gradient in the two forest-use types. Factorial design analysis between independent variables (forest-use types and elevation) and dependent variables.

Variables	Old-growth forest mean ± SE				Derived woodland mean ± SE				Factorial design analysis of variance		
	2200 m a.s.l.	2300 m a.s.l.	2400 m a.s.l.	2500 m a.s.l.	2200 m a.s.l.	2300 m a.s.l.	2400 m a.s.l.	2500 m a.s.l.	Forest types	Elevation	Forest types *Elevation
Trees											
Total number of tree species (0.5 ha)	9	6	7	7	3	3	2	2			
	10				3						
Basal area (m^2/ha)	20 ± 5	22 ± 3	21 ± 2	17 ± 1	18 ± 3	16 ± 3	12 ± 3	12 ± 1	p<0.03	ns	ns
Stem density (≥10 cm dbh) (n/ha)	150 ± 8	160 ± 7	140 ± 7	140 ± 50	130 ± 4	130 ± 10	130 ± 2	130 ± 4	p<0.00	ns	ns
Species richness (n/1000 m^2)	3 ± 0.5	2 ± 0.5	3 ± 0.4	3 ± 0.5	2.2 ± 0.3	2.2 ± 0.3	2 ± 0.01	2 ± 0.3	ns	ns	ns
Litter cover (%)	80 ± 0.1	80 ± 0.1	80 ± 0.1	70 ± 0.1	80 ± 0.1	75 ± 0.1	60 ± 0.1	50 ± 0.1	p<0.00	p<0.03	ns
Litter thickness (cm)	4 ± 0.3	4 ± 0.3	4 ± 0.3	3 ± 0.3	3.7 ± 0.7	3.5 ± 0.6	2.8 ± 0.5	1.6 ± 0.1	p<0.03	ns	ns
Soil											
Nitrogen (ppm)	0.08 ± 0.01	0.06 ± 0.01	0.05 ± 0.01	0.03 ± 0.006	0.08 ± 0.01	0.1 ± 0.007	0.1 ± 0.009	0.1 ± 0.0008	ns	ns	ns
Potassium (ppm)	108 ± 12	160 ± 50	126 ± 17	91 ± 25	81 ± 10	75 ± 2.2	59 ± 4.5	54 ± 2.8	ns	ns	ns
Phosphorus (ppm)	5.9 ± 1.2	6.5 ± 1	5.1 ± 0.7	5.6 ± 1.6	6.5 ± 0.7	5.4 ± 0.5	3.9 ± 0.4	3 ± 0.4	ns	ns	ns
Medicinal plants											
Total density (n/40 m^2)	475 ± 43	436 ± 42	367 ± 50	290 ± 33	332 ± 60	224 ± 36	173 ± 23	86 ± 10	p<0.00	p<0.00	p<0.04
Shannon index *H'*	2.5 ± 0.01	2.5 ± 0.01	2.5 ± 0.01	2.4 ± 0.01	2.1 ± 0.01	2.1 ± 0.01	2.0 ± 0.01	1.9 ± 0.01	p<0.01	ns	ns
Shannon evenness E_H	0.92 ± 0.09	0.91 ± 0.09	0.95 ± 0.09	0.89 ± 0.08	0.8 ± 0.008	0.9 ± 0.09	0.8 ± 00.008	0.9 ± 0.09	p<0.008	ns	ns
Total number of medicinal herb species (200 m^2)	22	21	20	20	23	24	22	19			
Total number of medicinal herb species present at all altitudes (0.8 ha)	22				30						

Mean and standard error, n=5 plots per altitude of forest-use type. "ns" represents no significant difference. Elevation gradient is in m a.s.l. (meters above sea level)

Results

General attributes of medicinal plants: In total, 31 herbs belonging to 23 plant families were observed in both the old-growth forest and derived woodland. The leaves of majority of the herb species (14) were used in various NTFP applications. All herbs were used as medicinal plants, and 20 herbs were used for fodder purposes. Approximately 11 species of medicinal plants were used for gastro-intestinal ailments, 11 were used as antipyretics, and six were used to treat respiratory infections. The highly marketable medicinal plant species were the following: *Valeriana jatamansi* (3.0 US dollars/Kg), *Fragaria nubicola* (8.1 US dollars/Kg), *Dryopteris ramosa* (7.1 US dollars/Kg), *Veronica laxa* (4.0 US dollars/Kg) and *Viola canescens* (3.1 US dollars/Kg); these species were also the most valuable for local people (Table 1).

Among the 31 medicinal plants, *Valeriana jatamansi*, *Viola canescens* and *Ajuga bracteosa* were found to be highly vulnerable, whereas 12 species, including *Veronica laxa*, *Bistorta amplexicaulis* and *Adiantum incisum* were found to be moderately vulnerable (Table 1).

Medicinal plant abundance along the altitudinal gradient in the two forest-use types: Of all the studied medicinal plants, 30 species were found in the derived woodland, whereas 22 species were found in the old-growth forest (Table 2). The densities of all the medicinal plants differed significantly between both forest-use types ($p<0.00$) and along the elevation ($p<0.00$). A significant interaction ($p<0.04$) was also observed between the forest-use types and elevation with respect to medicinal plant densities. In the old-growth forest, highest mean density (475/40 m^2) of medicinal plants was observed at 2200 m a.s.l., whereas the lowest density (290/40 m^2) was observed at 2500 m a.s.l.. Similarly, in the derived woodland, the highest mean density (332/40 m^2) of medicinal plants was observed at 2200 m a.s.l., whereas the lowest density (86/40 m^2) was observed at 2500 m a.s.l. (Table 2).

Densities of each of the five medicinal plant species *Valeriana jatamansi* ($r = -0.99$, $p \leq 0.01$), *Urtica dioica* ($r = -0.985$, $p \leq 0.01$), *Viola canescens* ($r = -0.972$, $p \leq 0.01$), *Dryopteris ramosa* ($r = -0.998$, $p \leq 0.01$) and *Veronica laxa* ($r = -0.740$, $p \leq 0.01$) were negatively correlated with altitude in the old-growth forest. In contrast to old-growth forest, densities of four medicinal plant species *Viola canescens*($r = -0.869$, $p \leq 0.01$), *Dryopteris ramosa* ($r = -0.968$, $p \leq 0.01$), *Veronica laxa* ($r = -0.950$, $p \leq 0.01$) and *Fragaria nubicola* ($r = -0.988$, $p \leq 0.01$) were negatively correlated with altitude in the derived woodland (Fig. 3).

Comparison of medicinal plant abundance and diversity between the two forest-use types: Sorensen diversity index showed highest similarity (80 %) of medicinal plants between the two forest-use types at 2200 m a.s.l., intermediate similarity (78 %) at 2300 m a.s.l., and lowest similarity (70 %) at 2500 m a.s.l.

Densities of most of the species showed a significant difference between the two forest-use types at higher altitudes. For example, species such as *Veronica laxa* ($p \leq 0.01$) and *Bistorta amplexicaulis* ($p \leq 0.01$) showed significantly higher densities in the old-growth forest in comparison with derived woodland at 2400 m a.s.l. and 2500 m a.s.l. However, densities of several species such as *Valeriana jatamansi* ($p \leq 0.01$) showed a significantly higher difference between both forest-use types at all the altitudinal levels (Table 3).

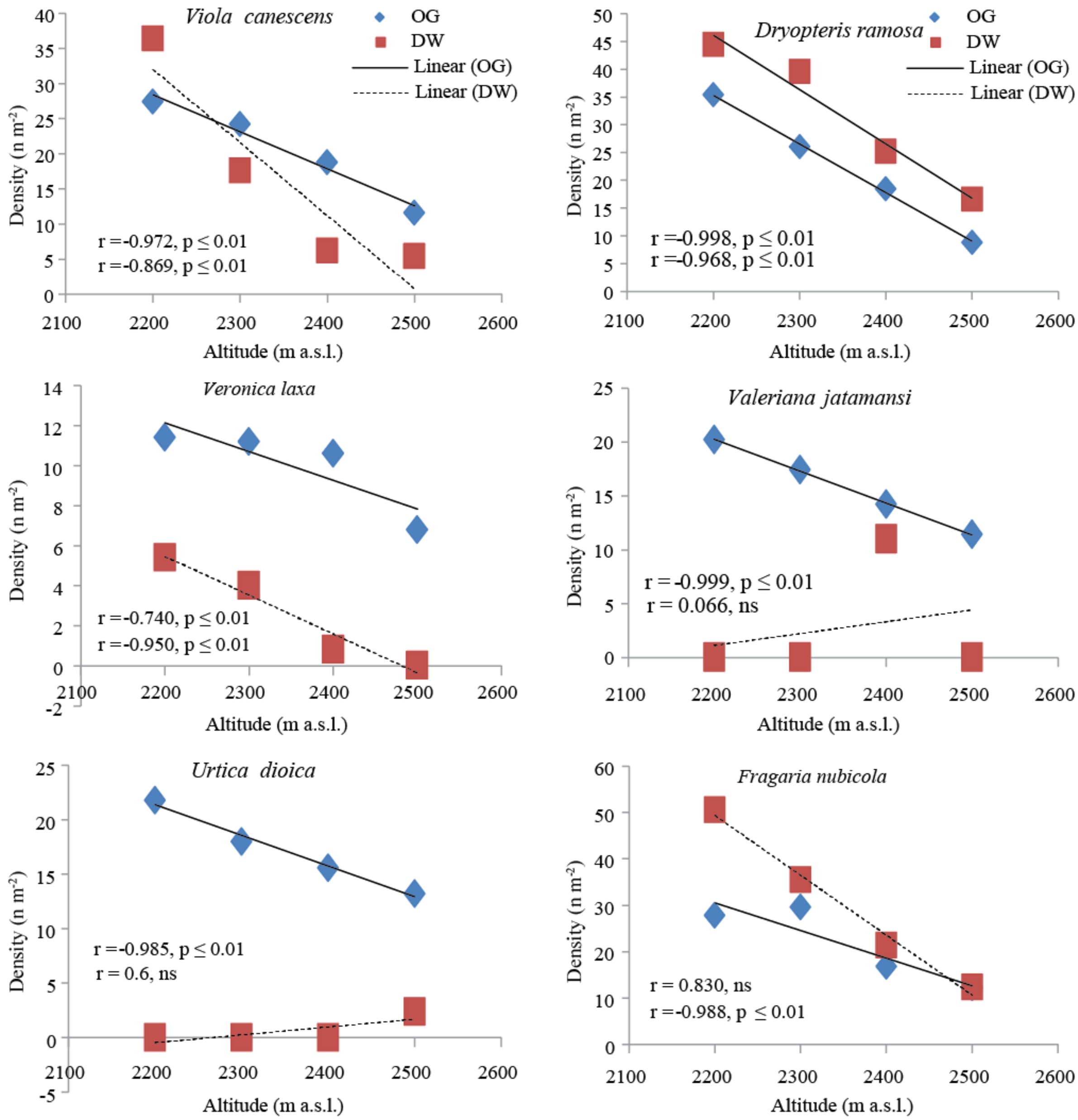

Fig. 3. Spearman correlation between the density of medicinal plants and altitude in two forest-use types. OG and DW represent old growth forest and derived woodland, respectively. ns indicates no significant correlation.

Relationship between forest-stand structural and medicinal plant variables: Litter cover (%) differed significantly between both forest-use types ($p<0.00$) and elevation ($p<0.03$). However, no significant interaction was observed between forest-use types and elevation with respect to litter cover. In the old-growth forest, highest significant value of mean litter cover (80%) was observed at 2200 m a.s.l., whereas lowest significant value of mean litter cover (70%) was observed at 2500 m a.s.l. However, in the derived woodland, highest significant value of mean litter cover (80%) was observed at 2200 m a.s.l., whereas the lowest significant mean litter cover (50%) was observed at 2500 m a.s.l. In addition, mean soil nitrogen, potassium and phosphorus were observed lower at higher altitudes in both forest-use types although not significant (Table 2).

As DCA indicate 40 plots of the two forest-use types, axis 1 (eigenvalue = 0.32) showed a significant correlation with litter cover ($r= -0.39$ at $p\leq0.01$). For the medicinal plants, axis 1 showed a significant correlation with the densities of *Valeriana jatamansi* ($r= -0.42$), *Veronica laxa* ($r= -0.57$) and *Urtica dioica* ($r = -0.78$) at $p\leq0.01$. This result indicates that the densities of such medicinal plants may increase markedly when litter cover increases (Fig. 4).

Table 3. Medicinal plant densities in old-growth forest (OG) and derived woodland (DW) at all altitudes. Mean and standard error, n=5 plots per altitude of forest-use type. na indicates data not available. Different small letters denotes significant differences ($p \leq 0.05$) (Mann–Whitney test). Elevation gradient is in m a.s.l. (meters above sea level).

Density (n/40 m^2)	2200m a.s.l. Mean ± SE		2300 m a.s.l. Mean ± SE		2400 m a.s.l. Mean ± SE		2500 m a.s.l. Mean ± SE	
	OG	DW	OG	DW	OG	DW	OG	DW
Adiantum incisum	$34^a \pm 11.3$	$34^a \pm 9.2$	$43^a \pm 16.8$	$29^a \pm 5.7$	$44^a \pm 10.4$	$31^a \pm 8.4$	$39^a \pm 9.2$	$10^b \pm 3.2$
Agrimonia eupatoria	$12^a \pm 7.4$	0^a	$8^a \pm 5.0$	$1.4^a \pm 1.4$	$9^a \pm 6.7$	$0.6^a \pm 0.6$	$6^a \pm 4.4$	$3.2^a \pm 3.2$
Ajuga bracteosa	na	2 ± 1.2	na	1.4 ± 0.8	na	0.6 ± 0.6	na	0
Androsace rotundifolia	na	0	na	0	na	1 ± 1	na	0
Aquilegia pubiflora	$5^a \pm 3.0$	$17^a \pm 6.4$	$5^a \pm 3.0$	$11^a \pm 3.9$	$3.4^a \pm 2.0$	$12^b \pm 2.8$	$0.8^a \pm 0.8$	$5^b \pm 1.9$
Arisaema flavum	$17^a \pm 6.3$	$11^a \pm 4.6$	$9^a \pm 3.5$	$10^a \pm 4.2$	$8^a \pm 1.7$	$5^a \pm 2.2$	$5^a \pm 2.6$	$2^a \pm 1.7$
Bistorta amplexicaulis	$44^a \pm 23.6$	$6^a \pm 5.6$	$52^a \pm 17.3$	$1.6^b \pm 1.6$	$44^a \pm 16.3$	$2.4^b \pm 2.4$	$35^a \pm 10.9$	$3^b \pm 3$
Chenopodium album	$1.4^a \pm 1.4$	$0.8^a \pm 0.8$	$1.2^a \pm 1.2$	$2^a \pm 2$	$0.8^a \pm 0.8$	0^a	$0.8^a \pm 0.8$	0^a
Chrysanthemum leucanthemum	$12^a \pm 5.2$	$10^a \pm 10$	$5^a \pm 3.1$	$1.8^a \pm 1.8$	$7^a \pm 4.9$	$1.4^a \pm 1.4$	$9^a \pm 4.4$	$0^b \pm 0$
Clematis grata	na	2.4 ± 2.4	na	1.8 ± 1.2	na	0	na	0.6 ± 0.6
Dipsacus inermis	$4.4^a \pm 4.4$	$0.6^a \pm 0.6$	$7^a \pm 4.5$	$0.4^a \pm 0.4$	0^a	$1.4^a \pm 1.4$	0^a	$0.6^a \pm 0.6$
Dryopteris ramosa	$35^a \pm 10.1$	$44^a \pm 14.2$	$26^a \pm 4.5$	$40^a \pm 8.8$	$18^a \pm 2.9$	$25^a \pm 9.2$	$9^a \pm 2.1$	$16^a \pm 5.5$
Euphorbia wallichii	na	3.8 ± 3.8	na	0	na	0	na	0
Fragaria nubicola	$28^a \pm 8.1$	$51^a \pm 19.1$	$27^a \pm 9.2$	$37^a \pm 10.3$	$17^a \pm 4.8$	$21^a \pm 9.3$	$12^a \pm 2.1$	$12^a \pm 6.2$
Galium aparine	na	0	na	3.6 ± 3.6	na	0	na	0
Hedera nepalensis	$13^a \pm 5.3$	0^b	$16^a \pm 4.2$	$3^b \pm 3$	$14^a \pm 3.9$	$1^b \pm 1$	$15^a \pm 5.1$	0^b
Impatiens bicolor	$35^a \pm 17.1$	$17^a \pm 7.2$	$37^a \pm 15.9$	$11^a \pm 6.3$	$33^a \pm 15.3$	$11^a \pm 4.2$	$28^a \pm 13.7$	$4^a \pm 1.4$
Leucanthemum vulgare	na	1.6 ± 1.6	na	0.8 ± 0.8	na	0	na	0
Lygodium jopanicum	$39^a \pm 8.1$	$30^a \pm 12$	$35^a \pm 7.4$	$24^a \pm 7.2$	$30^a \pm 5.3$	$16^a \pm 8.1$	$24^a \pm 3.8$	$7^b \pm 2.1$
Nepeta laevigata	na	0	na	0	na	0	na	4.6 ± 4.6
Oxalis corniculata	$53^a \pm 12.3$	$33^a \pm 22.1$	$43^a \pm 9.6$	$2.6^b \pm 2.6$	$31^a \pm 13.1$	$3.2^b \pm 1.9$	$32^a \pm 14.6$	$2^b \pm 1.1$
Polygonatum verticilatum	$0.6^a \pm 0.6$	$3^a \pm 2.3$	0^a	$9^a \pm 7.6$	0^a	$5^b \pm 3.4$	0^a	$2^a \pm 1.9$
Prunella vulgaris	na	1 ± 1	na	0	na	0	na	0
Ranunculus muricatus	$36^a \pm 4.1$	$9^b \pm 3.2$	$39^a \pm 2.5$	$2^b \pm 1.2$	$36^a \pm 6.7$	$7^b \pm 4.6$	$25^a \pm 4.2$	$2^b \pm 1.0$
Rubdosia longituba	$23^a \pm 7.1$	$9^a \pm 9$	$7^a \pm 4.4$	$10^a \pm 5.2$	$9^a \pm 3.7$	$7^a \pm 5.3$	$7^a \pm 2.8$	$3^a \pm 1.9$
Senecio salignus	1.2 ± 1.2	na	3 ± 1.7	na	2 ± 1.3	na	0.2 ± 0.2	na
Swertia chirata	na	4 ± 2.4	na	2.4 ± 1.7	na	2 ± 1.3	na	0.8 ± 0.4
Urtica dioica	$20^a \pm 8.3$	0^b	$17^a \pm 7.1$	0^b	$14^a \pm 5.9$	$11^a \pm 11$	$11^a \pm 5.0$	0^b
Valeriana jatamansi	$11^a \pm 4.7$	0^b	$11^a \pm 4.8$	0^b	$11^a \pm 4.1$	0^b	$7^a \pm 2.6$	$2.4^b \pm 2.4$
Veronica laxa	$11^a \pm 4.7$	$5^a \pm 3.4$	$11^a \pm 4.8$	$4^a \pm 2.5$	$11^a \pm 4.1$	$0.8^b \pm 0.8$	$7^a \pm 2.6$	0^b
Viola canescens	$27^a \pm 3.3$	$36.4^a \pm 26.3$	$24^a \pm 2.9$	$18^a \pm 8.5$	$19^a \pm 1.4$	$6^a \pm 4.7$	$12^a \pm 1.02$	$11^a \pm 4.9$
Total	**22 ± 3.2**	**11 ± 2.7**	**20 ± 3.4**	**7 ± 2**	**17 ± 3**	**6 ± 1.4**	**13 ± 2.5**	**3 ± 0.7**

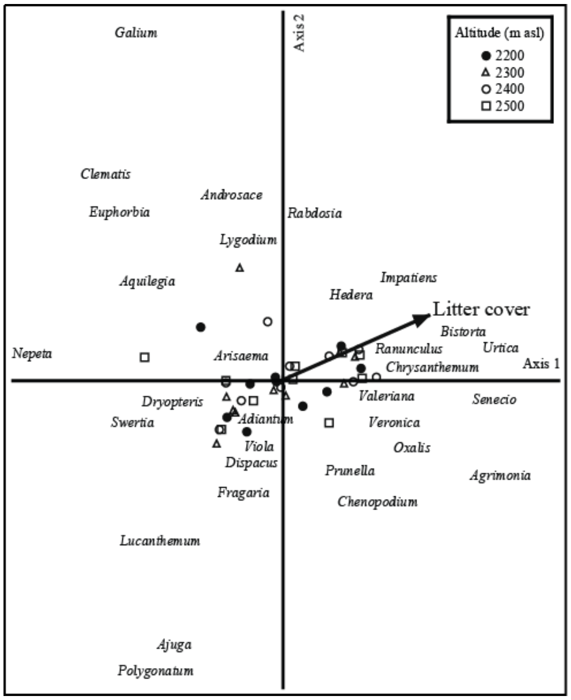

Fig. 4. DCA (Detrended correspondence analysis) plotted for the medicinal plants densities to litter cover at all altitudes for the two forest-use types. The matrix contain 40 plots, 31 medicinal plants and forest-stand structural variables (tree basal area, species richness, Shannon index, Simpson index, Shannon evenness, litter cover, litter thickness, stem density, and soil nitrogen, phosphorus and potassium). Axis 1: eigenvalue = 0.32, explained variance = 48%; Correlation threshold $r^2 > 0.23$; Vector scaling 216. Significant correlation (Spearman) at $p<0.01$ was observed for litter cover ($r = 0.39$) with axis1. For medicinal plants, a higher significant correlation (Spearman) of axis 1 at $p<0.01$ was observed for *Valeriana jatamansi* ($r= 0.42$), *Veronica laxa* ($r= 0.57$) and *Urtica dioica* ($r = 0.78$). Axis 2 provided little information and was therefore not included in the dataset.

Discussion

Medicinal plants and livelihood: NTFPs, particularly medicinal plants, are essential income source for the local people of Himalayan region of Pakistan. Results of this study indicate that the local community of Nathiagali in the immediate vicinity of ANP possesses valuable traditional knowledge on the uses of medicinal plants. The study shows that most plant species reported from the region are used for gastro-intestinal, antipyretic and expectorant treatments. These conditions may be caused by the poor hygienic conditions and smoke from fuel wood (Adnan & Holscher, 2012). In the study area, leaves are the plant part that is most frequently harvested for remedy preparation. This type of harvesting does not influence the life cycle of mother plant as severely as does the collection of roots, bark and stems (Ticktin, 2004). These results are similar to those other ethnobotanical studies conducted in Pakistan (Abbasi *et al*., 2010; Khan *et al*., 2011). Majority of the medicinal plants carry high prices due to increased market demand. Medicinal plants play an essential part in the socio-economic welfare of local people who depend on them, contributing to their total annual income (Michael &Perez, 2001; Adnan & Holscher, 2012). Species such as *Dryopteris ramosa, Fragaria nubicola, Veronica laxa, Valeriana jatamansi* and *Viola canescens* are among the actively traded medicinal plant species in the region. The market value of *Valeriana jatamansi*, *Viola canescens* and *Veronica laxa* has increased two-fold, whereas that of *Dryopteris ramosa* and *Fragaria nubicola* has increased six-fold, in the past five years (Khan, 2003). Increase in the prices of medicinal plant species may be due to gradual reduction in the density and cover abundance of such species over a period of time. Various forest-stand structural variables and anthropogenic factors have been identified as major reasons behind the decline in abundance of economically important flora (Khan *et al*., 2011; Adnan & Holscher, 2011).

Medicinal plants along the altitudinal gradient in old-growth forest: Overall, density of medicinal plants decreased as the altitude increased in old-growth forest. In the old-growth forest, overall decrease in species density might be due to changes in the forest-stand structural variables. In this study, we found that litter cover decreased with increasing altitude. This decrease might be associated with low productivity of litter at higher altitudes. Such a decrease might also be due to the effect of mean temperature on decomposition rate of litter as well as to changes in the microclimate (Shanks & Olson, 1961; Sher *et al*., 2005). At higher elevations, low temperatures can decrease the activity of microorganisms involved in decomposition of litter (Wang *et al*., 2007), which can, in turn, decrease the abundance of medicinal plants. Lower decomposition rates of litter and lower concentrations of nutrients due to increased lignin content in coniferous forests have also been reported (Ferrari, 1999). Non-decomposed litter at higher altitudes accumulates, leaving patches of incorporated litter and causing the soil to be susceptible to erosion. Moreover, the results of our study show a decrease in the nitrogen concentration at higher altitudes. This decrease might be a result of the decrease in litter cover.

Medicinal plants along the altitudinal gradient in derived woodland: Overall, density of medicinal plants decreased along the altitudinal gradient in derived woodland. Individual species such as *Viola canescens*, *Dryopteris ramosa*, *Veronica laxa* and *Fragaria nubicola* decreased as the altitude increased in derived woodland. This decrease may be a result of the reduction and improper decomposition of litter cover due to lesser microbial activities along the altitudinal gradient. Low litter cover may also be due to the lower tree basal area at higher altitudes in derived woodland. Low decomposition of litter cover can reduce soil fertility (Barbier *et al*., 2008), which may reduce the supply of vital nutrients to plants, thus reducing their abundance. According to Toutain (1987), soil nutrient availability is positively correlated with the rate of foliar litter decomposition. Our study also highlights that the concentrations of phosphorus and potassium decreased at higher altitudes. In addition, other anthropogenic factors such as over-collection of litter for agricultural purposes (bedding for animals and provision of nutrients in agricultural fields) may also be the reason for substantial reduction in medicinal plant densities in the derived forest compared to that in old-growth forest. Intense grazing pressure may reduce litter cover and thickness, as overgrazing makes the soil compact and produces mechanical injuries to the seedlings and soil organisms involved in litter decomposition.

The comparison of medicinal plants between old-growth forest and derived woodland: The DCA results showed that densities of certain medicinal plants, such as *Valeriana jatamansi* and *Urtica dioica,* decreased as the litter cover decreased with increasing altitude. Litter cover has a direct relationship with soil nutrients as mentioned earlier (Barbier *et al*., 2008; Toutain, 1987). Hence, decrease in medicinal plant densities might also be due to the lower concentration of nutrients in soil with increasing elevation. For example, *Urtica dioica* has been identified as a nitrophilous species (Olsen, 1921).

In the old-growth forest, medicinal plant richness was lower than that in derived woodland. The higher number of medicinal plants in derived woodland might be due to the diverse activities of humans and animals. Whittaker (1975) and Connell (1978) have also noted that little disturbance offers a big opportunity for species turnover, colonization and persistence of high species richness. In the old-growth forest, due to higher litter cover, nutrient availability is higher than that in derived woodland. High availability of nutrients allows only less number of few species to become so robust that the less vigorous species are excluded. In contrast, growth of all species is restricted at the sites with low availabilities of nutrient, and there is space for the survival of less vigorous species. As a result, competitive exclusion does not occur (Wang *et al*., 2007). This might be another reason for high species richness in derived woodland. However, the abundance of individual medicinal plants was more in old-growth forest than in the derived woodland at all altitudes (Adnan & Holscher, 2011). Overall, the densities of *Valeriana jatamansi*, *Bistorta amplexicaulis*, *Veronica laxa* and *Fragaria nubicola* were higher at 2300 m a.s.l. and 2400 m a.s.l. in old-growth forest than in derived woodland. This may be due to the high level of protection given to old-growth forest.

Our study also confirmed that the species similarity between the two forest-use types was also lower at higher altitudes. This finding may be due to the high level of anthropogenic activity in derived woodland. Grazing pressures are always directed toward higher altitudes to avoid any damage to agricultural crops (Bravo *et al*., 2009). Adnan & Hölscher (2012) reported that *Valeriana jatamansi* and *Veronica laxa* were more abundant in old-growth forest than in other forest-use types. However, due to their multipurpose uses, these species are facing great pressure in derived woodlands from grazing and over-collection. According to the Anon.,(2004), both these species are vulnerable due to their high market value, and this high value may also be a possible reason for overexploitation. Anthropogenic pressure may also be the differentiating factor between the forest-use types in terms of decreasing species abundance and diversity with altitude. This factor can not only disturb the vegetation directly but may also alter the environmental variables important for species abundance and diversity. For example, the indirect effects of overgrazing include soil erosion, which can significantly reduce the abundance of ground vegetation (Sher & Hussain, 2007). Vesk and Westoby & Vesk (2000) and Sher *et al*. (2005) have found that excessive grazing dovetails with the degradation of vegetation and decreases species spread due to direct consumption and habitat disturbance. Similarly, studies from various areas of the world have suggested that the composition, cover and diversity of the herbaceous layer may improve if the area is protected from a particular set of disturbances (Parrotta, 1995; Roberts & Zhu, 2002). Hence, there is great opportunity in the region to protect derived woodlands from anthropogenic disturbances to improve the abundance status of the vegetation.

Conclusions

The abundance of medicinal plants decreased across a gradient of mid-altitude levels in both of the forest-use types. Low litter cover might be associated to the decreased abundance of medicinal plants at higher altitude. Old-growth forest had a higher abundance of medicinal plants at middle altitudinal levels due to the high level of protection relative to the derived woodland. Several species such as *Valeriana jatamansi* are extinct at most altitudinal levels in the derived woodland. However, anthropogenic variables were not considered in this study. As a result, it is difficult to understand the relationship between environmental variables and medicinal plant density across the mid-altitude levels in derived woodland. Therefore, the inclusion of more forest-use types and anthropogenic variables may allow more specific conclusions about the topic in future research. Such research may suggest protection measures in the disturbed landscapes across the altitudinal gradient that could promote the reappearance of various economically valuable species, potentially contributing to both the livelihood of the local people and ecological restoration in the region.

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