DIVERSITY AND DISTRIBUTION OF BUMBLEBEES IN GORKHA REGION OF CHITWAN ANNAPURNA LANDSCAPE (CHAL), NEPAL

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T.U. Registration No: 5-2-37-1716-2012

T.U Examination Roll No: 403/2073

Batch: 2073/2075

A thesis submitted in partial fulfillment of the requirements for the award of the degree of the Masters of Science in Zoology with special paper Entomology

Submitted to

Central Department of Zoology

Institute of Science and Technology

Tribhuvan University

Kirtipur, Kathmandu

Nepal

February, 2020

DECLARATION

I hereby declare that the work presented in this thesis has been done by myself, and has not been submitted elsewhere for the award of any degree. All sources of information have been specifically acknowledged by reference to the authors.

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RECOMMENDATION

This is to recommend that the thesis entitled **"DIVERSITY AND DISTRIBUTION OF BUMBLEBEES IN GORKHA REGION OF CHITWAN ANNAPURNA LANDSCAPE (CHAL)"** has been carried out by Ms. Anjeela Pandey for partial fulfillment of the requirement for Master's Degree in Zoology with the special paper of Entomology. This is her original work and has been carried out under my supervision. To the best of my knowledge, this work has not been submitted for any other degree in any institution.

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LETTER OF APPROVAL

On the recommendation of supervisor Associate Prof. Dr. Daya Ram Bhusal, Central Department of Zoology, Tribhuvan University, the thesis submitted by Ms. Anjeela Pandey entitled "**DIVERSITY AND DISTRIBUTION OF BUMBLEBEES IN GORKHA REGION OF CHITWAN ANNAPURNA LANDSCAPE (CHAL)** " is approved for the examination in partial fulfillment of the requirement for the Master's Degree of Science in Zoology with special paper Entomology.

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CERTIFICATE OF ACCEPTANCE

This thesis work submitted by Ms. Anjeela Pandey entitled **"DIVERSITY AND DISTRIBUTION OF BUMBLEBEES IN GORKHA REGION OF CHITWAN ANNAPURNA LANDSCAPE (CHAL)"** has been accepted as partial fulfillment for the requirement of Master's Degree of Science in Zoology with special paper Entomology.

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ACKNOWLEDGMENTS

Foremost, I would like to express my heartfelt gratitude and indebtedness to my honorable supervisor Associate Prof. Daya Ram Bhusal, Ph.D., Central Department of Zoology (Entomology) for his guidance, immense help, valuable suggestions, information and knowledge regarding the subject matters. This thesis would not have taken its present shape and quality without his untiring efforts.

My sincere thanks and gratitude are due to Prof. Tej Bahadur Thapa, Ph.D., Head of the Central Department of Zoology for providing administrative support and facilities. I am highly obliged and extend my thanks to all of my respective teachers and staff of the Central Department of Zoology, TU, Kirtipur.

I am thankful to the National Botanical Garden, Lalitpur for helping in the identification of host plants.

I am thankful to Kishore Chandra Ghimire, Prabha Ale Magar, Subash Pandey, and Shruti Shakya for their immense help and assistance during field data collection.

I too extend my heartfelt gratitude to University grant commission(UGC) for awarding faculty Research Grant as financial support to conduct this research. I am very much grateful to all people who encouraged my study and all my friends for their direct and indirect help in completing this work.

Anjeela Pandey

Examination Roll No.: 403/073

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LIST OF ABBREVIATIONS

Abbreviated form	**Details of abbreviations**
ACA	Annapurna Conservation Area
AICc	Akaike Information Criterion (corrected)
ANOVA	Analysis of Variance
Asl	Above sea level
DF	Degree of Freedom
GLM	Generalized Linear Model
GPS	Global Positioning System
m	Meter
WWF	World Wildlife Fund for Nature

ABSTRACT

This study was conducted in the Gorkha region of Chitwan Annapurna Landscape (CHAL), Nepal to explore the diversity and distribution of bumblebees. The fieldwork was carried from May to September 2019 across the altitudinal gradient by following an accessible walking trail. Bumblebee specimens were collected by using sweeping nets. Meanwhile, the physical variables such as altitude, temperature, and humidity were also noted. Host plants of collected bumblebees were also recorded. This study documented 12 *Bombus* species belonging to 8 subgenera. *B. haemorrhoidalis* was the most abundant species representing 30% of total species while *B. pressus* was the lowest abundant species representing 1% of the total species. The highest diversity of bumblebees was found in grassland (H=1.9585) while the lowest in the home garden (H=1.06086). Species richness of bumblebees was found to be positively correlated with altitude and humidity while negatively correlated with temperature. Similarly, bumblebees show significant effect (p=0.00000225) in preferences with the host plant's family and color of the host plant with significant effect (p=0.00124). This study has indicated that the diversity and distribution of bumblebees are mainly affected by physical parameters, habitat type and families, and color of the host plant.

1.INTRODUCTION

1.1 Background

Bumblebees (Hymenoptera: Apidae) are a genus of pollinating insects that play an important role in crop production and natural ecosystem services (Fontaine et al. 2006, Garibaldi et al. 2013). They are distributed widely across the globe, from Greenland to the Amazon Basin and from sea level to altitudes of 5,800 m in the Himalayas (Williams 1985). They occupy a remarkably wide diversity of habitats, from alpine meadows to lowland tropical forest (Sakagami 1976). Adults feed mainly on nectar, whereas larvae are fed on the mixture of crushed pollen grains and honey, which fulfills their requirements for growth. The establishment and development of colonies take place each summer and may take less than two months (Richards 1973), culminating with a switch to the production of males and young queens near the end of the season. After mating, young queens normally diapause through the winter, away from the nest, and they attempt to establish new colonies on their own in the following year. Workers are having a peculiar structure on meta tibia called pollen basket which is absent in males. Males are having 13 antennal segments and 7 visible tergites while their number is 12 and 6 in females respectively. Queen and worker bumblebees can sting, but unlike honey bees, a bumblebee's stinger lacks barbs so they can sting more than once. A strange dimension of bumblebees is the parasitic bumblebees belonging to a separate subgenus *Psithyrus.* Because of their habits and morphological feature, earlier these bees were grouped under a separate and independent group having the status of a genus. However, now this taxon has been lowered to a subgenus. The unique feature of these bees is that their queens and workers do not have pollen collecting 3 baskets on the tibia of their meta legs and thus they are dependent on the host *Bombus* species for food. The *Psithyrus* queen invades the nest of the real bumblebees, and if not killed or repelled by the workers of host bumblebees, it kills their queen and captures the nest. Later it lays its eggs in the nest and the actual bumblebee workers feed them.

There are 250 known species of bumblebees present on a global basis (Williams et al. 2008) whereas 34 species with 10 subgenera have been reported from Nepal (Williams et al. 2010). These bees nest either in the ground, often in abandoned rodent burrows, or above ground, in tall grasses (Kearns and Thomson 2001). Recent studies have shown that bumblebees routinely forage at considerable distances from their nests (Dramstad 1996, Osborne et al. 1999).

Bumblebees are important generalist pollinators of native plants and crops. The list of flowers visited by bumblebees is vast (Kearns & Thomson 2001, Goulson 2010). They are associated with high lands and play a key role in the functioning of the agricultural ecosystem as pollinators of crops which include tomatoes, peppers, raspberries, blueberries, chives, cucumbers, apples, strawberries, alfalfa, blackberries, beans, cherries, apricots, plums, peaches, eggplants and cranberries, orchards and wildflowers (Colla et al. 2011) and are both economically important pollinators serving as a principal model system for a variety of studies in social behavior and ecology (Goulson 2010). Their critical role in ecosystem conservation makes them important living organisms like other pollinators (Corbet et al. 1991). Bumblebees can pollinate at high speed by opening the pollen sacs with vibrating as buzzing behavior. Being capable of buzz pollination, they are excellent pollinator of Solanaceae such as tomatoes (Van den et al. 1991). The anthers of these flowers only release pollen when vibrated, which bumblebee achieve by placing their thorax close to the anthers and contracting their flight muscles at a frequency of about 400Hz (King 1993).

The abundance of bumblebee has shown serious declines in many parts of worlds (Buchmann and Nabhan 1996, Cane 2001, Kevan and Phillips 2001) especially in the last 50 years (Goulson et al. 2005). Bumblebee species are declining in Europe, North America, and Asia (Williams and Osborne 2009). A decline in pollinators like bumblebees presents a serious threat to agricultural production, conservation, and maintenance of biodiversity (Biesmeijer et al. 2006). The reason for declining in a natural population of several bumblebee species includes several factors such as human activities (Williams 2002) change in land use by changing agriculture policy (Yang 1999, Xie et al. 2008), the drainage system and grasslands improvement with fertilizers (Goulson et al. 2005), loss of rotations of legume crops by replacement with continuous cereals with chemical fertilizers (Kleijn and Raemakers 2008), application of pesticides (Williams 1986, Kevan et al. 1997) and intensive grazing of grassland (Xie et al. 2008). Moreover, climate change poses a threat to many high altitudes bumblebee species worldwide due to the ongoing decline in suitable habitats (Hoiss et al. 2012). Studying and understanding the association between species richness and elevation gradients is essential, as it provides insights into the observed patterns and processes responsible for the relation, which in turn supports conservation efforts (Stevens 1992, Raman et al. 2005, Acharya et al. 2011).

1.2 Objectives of the Study

General Objective

- To find the diversity and distribution of bumblebees in the Gorkha region of Chitwan Annapurna Landscape (CHAL), Nepal.

Specific Objectives

- To find the diversity and distribution of bumblebee species in the study area.
- To find the relation of bumblebees with physical parameters (altitude, temperature, and humidity).
- To explore the relation of bumblebees with their host plants.

1.3 Rationale of the study

Bumblebees are the most important insect pollinators of wild and cultivated flowering plants, especially in high altitudinal gradients. Very few studies on bumblebees have been carried out sporadically in Nepal. Gorkha is one of the most important parts of CHAL. However, there is no comprehensive information about ecologically and economically important bumblebees in this region. This research work will help by providing the baseline information on the species diversity, distribution from this less explored area that could be crucial for the conservation and management of bumblebees and its habitat in wild.

2. LITERATURE REVIEW

2.1 Species diversity and distribution

The first systematic study of bumblebee distribution in the eastern region was carried by Streinzer et al. 2018 in Arunachal Pradesh, India. They have recorded 21 species belonging to 6 subgenera of bumblebees from 700 specimens with the altitudinal range from 233m to 4,260 m while the total Indian fauna reported is 48 species (Saini et al. 2015). Williams et al. (2017) reported 130 species of bumblebee from China which is exactly half of the current estimated number of bumblebee species worldwide. They concluded China to be the greatest hotspot for bumblebee diversity worldwide. Williams et al. (2015) surveyed bumblebees, climate, and the glaciers across the Tibetan plateau, and they recorded 44 species of bumblebees. An et al. (2014) made a field survey from 2005-2012 in North China and they assigned to 76 species. One older specimen reported in London added 1 more species to this list-making 77 species represented 10 subgenera of the genus *Bombus*. Seven species were recorded from North China for the first time showing the greatest hotspot of the bumblebee diversity worldwide.

Saini et al. (2012) made remarkable research to investigate species diversity of bumblebee in three provinces Jammu, Kashmir, and Ladakh. They reported 27 species of bumblebee of which eleven species were common in the two regions, i.e. Kashmir and Ladakh. They also concluded that the Kashmir region had higher species diversity and richness than the other regions and most species of bumblebees were collected in altitudinal ranges of 3000-4000m. Four species *viz. Bombus cornutus*, *B. parthenius*, *B. miniatus*, and *B. morawitizianus* were recorded for the first time from that region, while *B. morawitizianus* turned out to be the first record to the national list. Colla et al. (2011) made a study on bumblebee of western United States, they found that, in states where boundaries are fully east of the 100th meridian, there are a total of 21 species. Similarly, Sabir et al. (2008) made a research on bumblebees from Northern Pakistan and they reported 13 indigenous sps. ranging from 2291-5344m above sea level. Cannani et al. (2006) studied flower choice and learning in bumblebees where they found that there may have been some interaction with their existed diversity at altitude stratum and possible floral searching behavior to a certain limit. William (1998) studied the diversity of bumblebee in Kashmir and he reported 26 species from that site. William and Nat (1998) make a research on the Natural History

Museum of London and found out that there were many more common elements in the fauna of Nepal and Kashmir, and they found that Kashmir shares 26 species of bumblebee. Osborne et al. (2008) surveyed bumblebee nest densities in gardens and countryside. They found that nest density was high in gardens and linear countryside habitats and lower in non-linear countryside habitats (woodland and grassland: 11–15 nests ha–1). McFrederick and LeBuhn (2006) studied nesting sites of bumblebees in urban parks in San Francisco and he discovered that bumblebee abundance in urban areas was found to be positively correlated with the number of rodent holes, hence suggested that nest sites might be a limiting factor.

Except for little work (Williams et al. 2010), there is no systematic study of the bumblebees in Nepal. They have recorded 34 species. They have recorded the largest number of bumblebee individuals as well as bumblebee species from the elevation range of 3,000 – 3,999 m. They also predicted that three additional species (*B. keriensis, B. sibiricus* and *B. tanguticus*) are likely to occur in Nepal because they have been recorded inside Tibet. Also, there is a probability of the occurrence of another five species in Nepal (*B. bohemicus, B. difficillimus, B. genalis, B. oberti,* and *B. simillimus*) near the edge of high Tibetan plateau border zone. Likewise, a study on bumblebees from Kaligandaki and Marsyangdi river basin in Chitwan Annapurna Landscape (CHAL) was done by Roka (2019), where she reported 10 species of bumblebee. *Bombus festivus* was the most abundant species representing 39.30% of total species. The highest diversity of bumblebee was found in the forest while the lowest in home garden. Similarly, Bhusal et al. 2019 collected six species of the bumblebees from Kathmandu valley. They found that the highest individual abundance was *Bombus haemorrhoidalis* and the least abundance was *B. trifasciatus.*

2.2 Relation of bumblebees with physical parameters

Peat and Goulson (2005) found that foraging rates of *Bombus terrestris* were positively correlated with humidity while temperature did not significantly influence their foraging rate. A positive correlation between the foraging of bumblebee and humidity was due to the higher nectar secretion rates at higher humidity (Peat and Goulson 2005). They also suggested that bumblebee avoid collecting pollen when there is dew or rain-water droplets on the vegetation, which would make grooming pollen into the corbiculae difficult. Williams et al. (2010) found bumblebee’s frequency positively correlated with altitude. Hoiss et al. (2012) found that species richness and abundance of bees showed a linear decline with increasing altitude because the diversity of species decline linearly with decreasing temperatures. But bumblebees can forage at extremely low temperatures (AllenWardell et al. 1997, Saini et al. 2012). The landscape context and the habitat quality influence the species composition of bumblebee as stated by Carvell et al. (2011). Similarly, Roka (2019) found that the foraging of bumblebee was positively correlated with altitude and humidity while negatively correlated with temperature.

2.3 Relation of bumblebees with host plants

Elliott (2009) studied foraging distances of bumblebee with flower availability and he reported that bumblebees often utilize floral resources over a landscape ranging from <100m to a few kilometers in radius depending on the vegetation structure around nesting sites. Inouye (1980) studied the effect of corolla length and proboscis on the rate of flower visitation by bumblebees. He found that shorter-tongued bees foraged faster on short-corolla flowers than long-tongued bees, although he found it difficult to measure the foraging times of long-tongued bees on shorter corolla flowers because of their apparent reluctance to feed on short corolla flowers. Heinrich (1977) studied bumblebees foraging in an old field in the U.S.A. He confirmed the species preferences of earlier studies but also found that each individual preferred a small subset of the overall species group. He named the main foraging flower the "major" and the secondary flower(s) the "minor(s)".

Raine and Chittka (2005) made a research on color preferences of the bumblebee *Bombus terrestris* with the foraging performance and fitness. They developed a conclusion that *Bombus terrestris* colonies showed significant variation in their unlearned preference for violet (bee UV-blue) over blue (bee blue) flowers. Bumblebee colonies with a higher average innate preference for violet (over blue) in the laboratory harvested more nectar per unit time under field conditions. Ohashi and Yahara (1998) examined the functional relationships between the floral display and two types of bumblebee response, the visitation rate per plant and the number of flowers visited on a plant, in an artificially arranged field population of *Cirsium purpuratum* (Asteraceae). They found that the visitation rate of bumblebees per plant was a decelerating function of floral display and that the number of flowering heads visited on a plant increased linearly with display size. Similarly, Giurfa et al. (1995, 1996) researched the color preferences of flower native bees. They found that bumblebees as well as honeybees exhibited innate color preferences encompassing preferences for colors dominated by blue wavelengths and also showed preferences for colors that contrast with the background. Similarly, Lunau et al. (1996) studied color choices of native bumblebees and their implications for color perception. They concluded that bumblebee prefers colors of high color purity.

Somme et al. (2014) researched spatial structure impacts on the pollination services of *Comarum palustre* (Rosaceae). They found that bumblebee and solitary bees were the major pollinators among all populations. Also reported that, within populations, plots with high flower densities attracted high numbers of bumblebee and other insects as a bumblebee with 10 different species, summed up to 96.1% of the bee visitors. High bumblebee and solitary bee abundance were observed in populations presenting high proportions of woody edges and in populations within landscapes presenting high proportions of woody areas and seed set resulting from open pollination varied with bumblebee and solitary bee visitation rate, leading to increased pollen limitation when pollinators were scarce. Sinu and Shivanna (2007, 2011) reported bumblebees to be the most effective pollinator of large cardamom (*Amomum subulatum*). Similarly, Daka et al. (2011) reported that two bumblebees, *B. haemorrhoidalis* and *B. breviceps* were the predominant pollinator of large cardamom.

Goulson (2010) studied ecology and conservation in Netherland and he discovered that commercially bumblebees are very efficient as a pollinator of greenhouse tomatoes. He also noticed that only 3 years after the commercial strains of the buff-tailed bumblebees *B. terrestris* became available, 95% of the tomato growers in the country were using them instead of honeybees or hand pollination and they rapidly gained a standard status in pollinating greenhouse tomatoes in Europe as well. Abrol (1998, 1989) studied on importance of native pollinators for crop production in India. He recorded that *Bombus* species are potential pollinators visiting horticultural and crops during March- December. He also reported *B. asiaticus, B. albopleuralis,* and *B. simillimus* are important pollinators of solanaceous and leguminous crops in Srinagar, India during the blooming period of several fruit and field crops. Buchmann (1983) made a research in buzz pollination in angiosperms. He found that the buzz-pollinated flowers had evolved independently many times, occurring in species from 65 families, including some of the world's most important crops such as tomatoes and potatoes. He also noted that among insects, the ability to sonicate is found among many species of bees (Hymenoptera: Apoidea) from seven families.

Similarly, King (1993) made research on buzz foraging mechanisms in bumblebee. He developed a conclusion that buzz-pollination is probably an innate behavior in some species, as native *Bombus terrestris* bumblebee foraging on buzz-pollinated flowers will exhibit buzzing on their first foraging trips. Free and Williams (1979) studied on pollinators and pollination limiting factors of beans. They reported that the yields of many fields, fruit and seed crops are enhanced by bumblebee visitation, like field beans are largely pollinated by longer tongued species such as *B. pascuorum and B. hortorum*, without which yields are poor. Similarly, Faegri and Van der Piji (1979) made a research and reported that bumblebee are the chief pollinators of red clover, alfalfa and in some areas cotton, raspberries, apple, and plum blossom and Norwegian orchards they found that honeybee pollination is usually limited by low temperatures, so bumblebees are the chief pollinators there. McGreror (1976) studied pollinators of cultivated crops and he found that the clover needs the cross-pollination for seed production. As the flower tube is so long, bumblebee being the long-tongued insects are relatively more effective pollinators than honeybees, as they work on the flowers faster and trip them more rapidly, collecting nectar or pollen. He also noticed the greater enhancement of seed set with the bumblebee than with honey bees.

Cozmunta et al. (2012) made a remarkable study on blooming and pollen production of two *Lamium* L. species, a Lamiaceae two-lipped entomophilous flower. They got that these flowers are frequently visited by long-tongued bees as the great amount of nectar is stored in deep corolla, primarily bumblebee and wild bees attracted by nectar and pollen reward, contributing to the cross-pollination of these plants. Stout et al. (2000) made a remarkable research in southern England on *Linaria vulgaris* (common yellow toadflax). They found that that plant suffered from high rates of nectar robbery by a bumblebee. In a wild population of *L. vulgaris,* they found that 96 % of open flowers were robbed. Five species of bumblebee were observed foraging on those flowers, although short-tongued species (*Bombus lapidarius, B. lucorum,* and *B. terrestris*) robbed nectar whilst longer-tongued ones behaved as legitimate pollinators (*B. hortorum* and *B. pascuorum*). Also, they found that nectar rewards were highly variable; on average there was less nectar in robbed than in unrobbed flowers, but this difference was not statistically significant.

3. MATERIALS AND METHODS

3.1 Study area

The CHAL is located in Central Nepal, covering an area of 32,057 km square, with elevation ranging from 200 m to 8091 m. It possesses diverse micro-climatic variation that creates a suitable habitat for many species of pollinating insects including bumblebees. CHAL is one of two priority working areas for the WWF- Hariyo Ban program. This landscape has a high biodiversity value and contains seven major sub-river basins: Trishuli, Marsyangdi, Seti, Kaligandaki, Budigandaki, Rapti, and Narayani. This landscape covers all or part of 19 districts. This study was conducted in the Gorkha region. Gorkha region is one of the important sites for biodiversity and human settlement. Its upper region lies in the Manaslu Conservation area. This study was conducted from 500-2,800m altitudes. Gorkha region is believed to be one of the most potential bumblebees habitats (Figure 1).

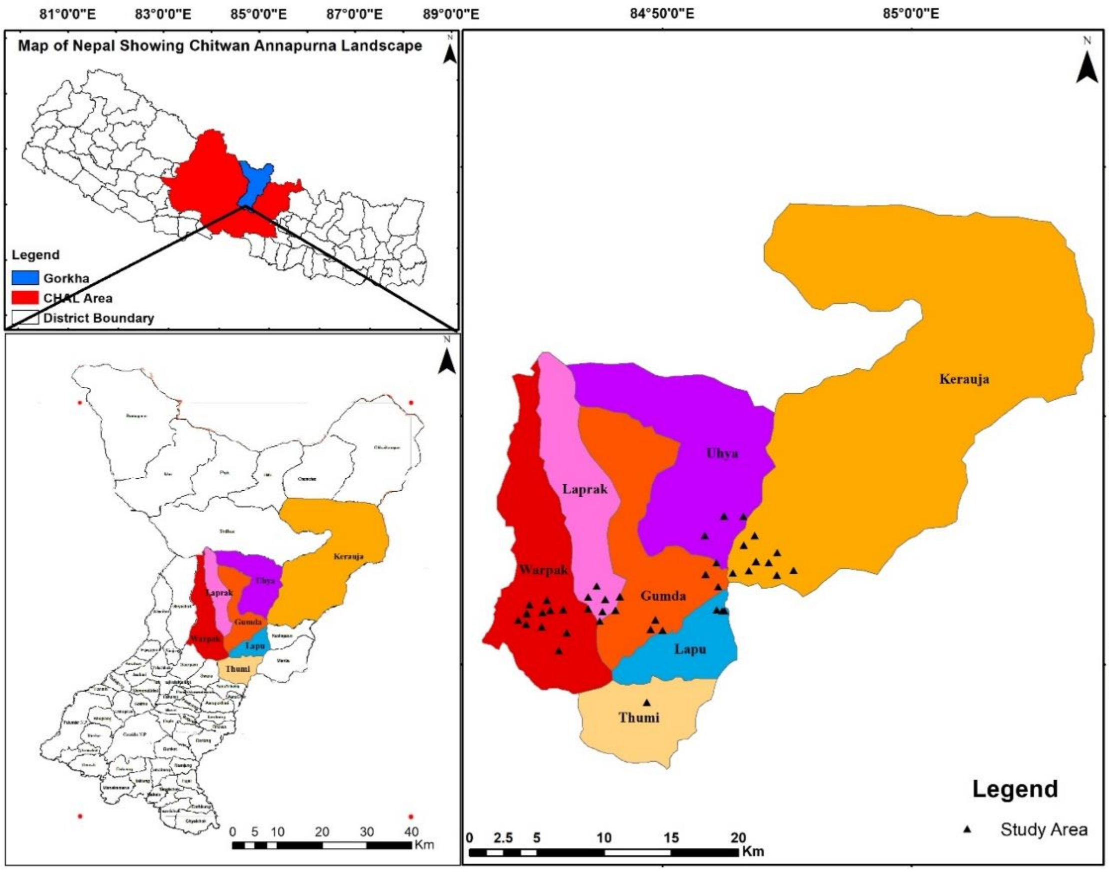

Figure 1: Map of the study area

3.2 Materials

- Sweeping net
- GPS (Garmin eTrex® 10)
- Collecting jar
- Camera (Canon EOS 1300D)
- Insect Box
- Refrigerator
- Hygrometer
- Entomological pin
- Silica gel
- Stereoscopic microscope

3.3 Sampling

3.3.1 Field design

The fieldwork was carried out from May to September 2019 in the Gorkha region of CHAL. Two accessible walking trail from 500-2,800m altitudinal range were followed in different habitats. Sampling was done during a period of about 45 minutes to 1 hour in all the habitats, or until it was satisfied that all the species on the site were recorded.

3.3.2 Sample collection

Bumblebees were collected with the Sweeping net method. Then, the captured bumblebees were collected in empty plastic vials with breathing holes in the sides containing a tissue paper to absorb the moisture (An et al. 2014). Co-ordinates were recorded at each bumblebee sampling site with the help of GPS. Meanwhile, the physical environmental parameters (temperature and relative humidity) were noted with hygrometer, and altitude was noted with the help of GPS. Time of foraging was also noted in morning, afternoon, and evening time. Digital pictures of the foraged host plant by the bumblebee were taken and collected as herbarium for identification. The host plant color, family, and category (invasive/ noninvasive) were recorded. Finally, the collected bumblebees were preserved in a refrigerator (-4°) to prevent discoloration and brought to the lab for identification. Then, the specimens were dry-mounted on standard insect pins for identification.

3.3.3 Specimen identification

The collected bumblebees were observed in a Stereoscopic microscope and identified using the identification key (Williams et al. 2010, Saini et al. 2015). Herbarium of unidentified host plants was made and identified from the National Botanical Garden, Lalitpur. The collected bumblebee specimens were preserved in the Central Department of Zoology, Tribhuvan University as voucher specimens.

3.4 Data analysis

Data obtained from the field were managed in Microsoft Excel 2010 and analyzed using R version 3.5.0. The data were analyzed using the following diversity index and statistical tools.

3.4.1 Shannon-Wiener diversity index (H)

Shannon-Wiener diversity index was used to calculate the species diversity index in a different habitat.

$H' = -\Sigma \, Pi \, (\ln Pi)$

Where, Σ represents sum of $Pi(\ln Pi)$

H'= Index of species diversity

Pi = the proportion of individuals in the i^{th} species, $Pi = n_i/N$

ni = Importance value for each species (number of individuals)

N= Total importance value (total number of individuals)

3.4.2 Relative abundance (%)

Relative abundance was used to show the subgenus and species composition of a bumblebee. Relative abundance (%) = $(n/N) \times 100$

pi= n/N= Relative abundance of each species, calculated as the proportion of individuals of a given species to the total number of individuals in the community.

3.4.3 Pearson's Chi-square test

Pearson’s chi-square test was performed to know the distribution frequency of *Bombus* species in a different habitat.

$$\chi^2 = \sum_{i=1}^{n} \frac{(O_i - E_i)^2}{E_i}$$

χ^2 = Pearson's cumulative test statistic

Oi = the number of observations of type i

Ei = the expected (theoretical) count of type i,

3.4.4 Analysis of Variance

One-way ANOVA was performed to know the relation of bumblebee with the family, category, and color of the foraged flowering plants.

3.4.5 Generalized Linear Model (GLM)

Generalized Linear Model (GLM) was performed to determine the relation of bumblebee with the color of the flowers, a family of foraged flowering plants, category (invasive/non-invasive) and the habitats visited by the bumblebees.

3.4.6 Principal Component Analysis (PCA)

PCA was used to know the relation between physical parameters (altitude, humidity, and temperature) and the abundance of the bumblebee.

3.5 Species Distribution map preparation

Distribution estimation and mapping were done with ArcGIS 10.5 software and species location was overlapped with Google Earth and Landsat imagery to provide information on the spatial distribution. The species recorded locations of the bumblebees were overlaid as GIS layers to prepare a distribution map.

4. RESULTS

4.1 Diagnostic features of the collected *Bombus* species

S.N.	Species	Subgenus	Diagnostic features
1	*Bombus asiaticus* Morawitz, 1875.	*Sibiricobombus*	- Head, mesonotum and abdominal tergites 2-5 black - Pronotum, metanotum and abdominal tergum 1 are a dirty yellow - Clypeus without any pubescence - Visible parts of thorax and abdomen uniformly covered with thick pubescence **(Photo 8)**
2	*Bombus haemorrhoidalis* Smith, 1852.	*Orientalibombus*	- Head, thorax and abdominal tergum 3 entirely black - Abdominal tergites 1 and 2 yellow - Abdominal tergites 4 and 5 brick red - Pubescence short - Head covered with thick pubescence except for malar space **(Photo 1 and 2)**
3	*Bombus rotundiceps* Friese, 1916.	*Pyrobombus*	- First, 3 abdominal tergites are dirty yellow - Last 3 abdominal tergites are brick red - Mid tibia and hind tibia with long black hairs - T1-2 black - Head covered with thick pubescence except for malar space **(Photo 3 and 4)**
4	*Bombus turneri* Richards, 1929.	*Psithyrus*	- The outer surface of the hind tibia convex with moderate to long hairs without a comb of stout spines along the distal margin - Labral lamella narrow and strongly produced with the apex at least triangular - Pubescence of thoracic dorsum black with yellow intermixed anteriorly and posteriorly **(Photo 5)**
5	*Bombus festivus* Smith, 1861.	*Festivobombus*	- Thorax and last two abdominal tergites white - Head covered with thick pubescence expect malar space**(Photo 6)**

6	*Bombus miniatus* Bingham, 1897.	*Melanobombus*	- Pubescence of the head with the short branched hairs yellow, pale thoracic bands and T1 yellow **(Photo 10)** - Pubescence of the thoracic dorsum with a black band between the wing bases, anterior and posterior pale bands either dull yellow or grey-white - Hind tibia with the outer surface without short hairs.
7	*Bombus parthenius* Richards ,1934.	*Pyrobombus*	-Pubescence entirely yellow except malar space, which is black **(Photo 12)** **-**The distoposterior corner of mesobasitarsus bluntly pointed.
8	*Bombus pressus* Frison, 1935.	*Pressibombus*	-Pubescence of the thoracic dorsum with a black band between the wing bases**(Photo 13)** **-**Hind tibia distal posterior corner never produced as a spine, always shorter than its basal breadth
9	*Bombus eximius* Smith, 1852.	*Melanobombus*	-Hair of the thorax black -Hair of the mid and hind tibiae and the basitarsi orange - Wings light orange-brown**(Photo 14)**
10	*Bombus genalis* Friese, 1981.	*Alpigenobombus*	-Head, thorax, and abdomen covered with black pubescence -Clypeus white**(Photo 7)**
11	*Bombus grahami* Frison, 1933.	*Alpigenobombus*	- Pubescence of the thoracic dorsum grey-white with an intermixture of black - Oculo-malar distance shorter than half of the basal breadth of the mandible -Mandible apex with six large teeth **(Photo 11)**
12	*Bombus breviceps* Smith, 1852.	*Alpigenobombus*	-Short malar space -Stout bumblebee -Pleura of thorax white -Tergum of abdomen region makes arc shape **-**The color pattern of T3 spread to the center oT4 (yellow) while T4 margin and T5 is black and T6 is orange **(Photo 9)**

4.2 Abundance of bumblebees

A total of 209 individuals of 12 bumblebee species from eight subgenera were recorded during the study period. Subgenus *Alpigenobombus* were represented by a maximum number of three species, followed by *Melanobombus* and *Pyrobombus* with two species while remaining subgenera such as *Festivobombus, Orientalibombus, Psithyrus, Pressibombus* and *Sibiricobombus* were represented with one species each (Table 1).

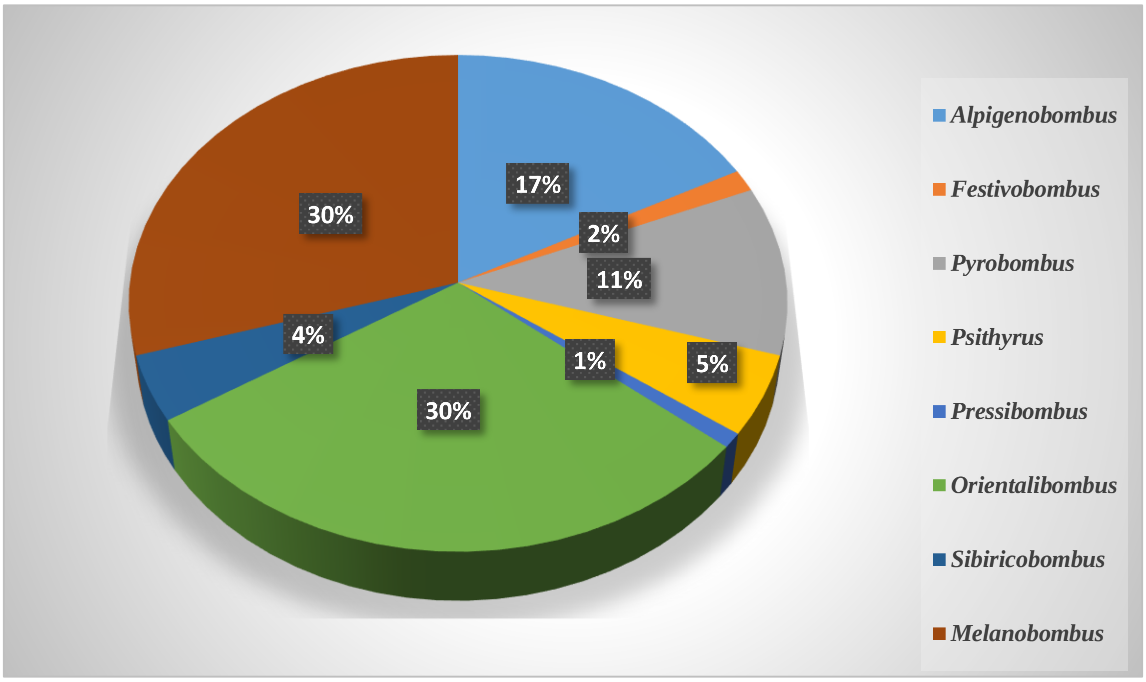

Figure 2: Subgenus composition of *Bombus spp*. in the study area.

Among eight subgenera of bumblebees, *Melanobombus* and *Orientalibombus* were most abundant representing 30% of total subgenus followed by *Alpigenobombus* (17%), *Pyrobombus* (11%), *Psithyrus* (5%), *Sibiricobombus* (4%), *Festivobombus* (2%) while *Pressibombus* (1%) was least abundant.

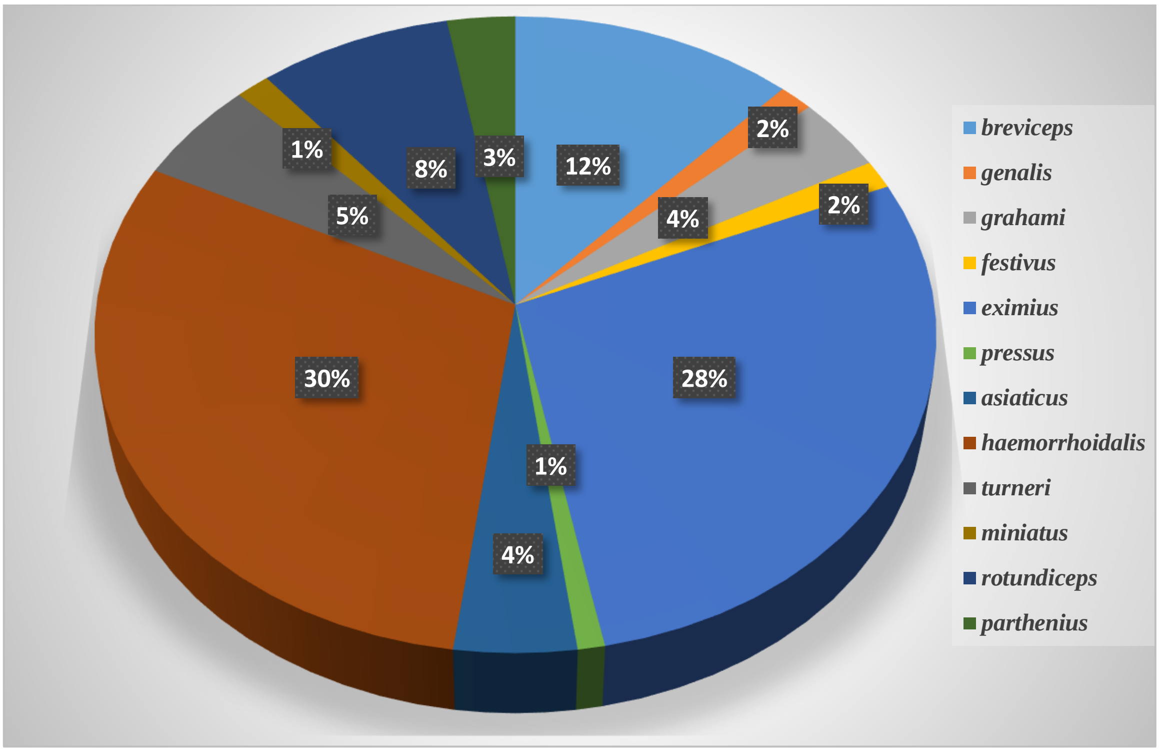

Figure 3: Species composition of *Bombus spp.* in the study area

The species abundance recorded from Gorkha region of CHAL is in the following order: *B. haemorrhoidalis* (30%); *B. eximius* 28%); *B. breviceps* (12%); *B. rotundiceps* 8%); *B. turneri* (5%); *B asiaticus* (4%); *B grahami* (4%)*;* *B. parthenius* (3%); *B. festivus* (2%) *B. miniatus* (2%); *B. genalis* (1%); and *B. pressus* (1%).

4.3 Altitudinal Variation

Based on elevation, all recorded bumblebees (12) species were recorded from 2,100-2,800m, six species from 1,400-2,099m, one species from 700-1,399m, and also one species from 0-699m (Annex I).

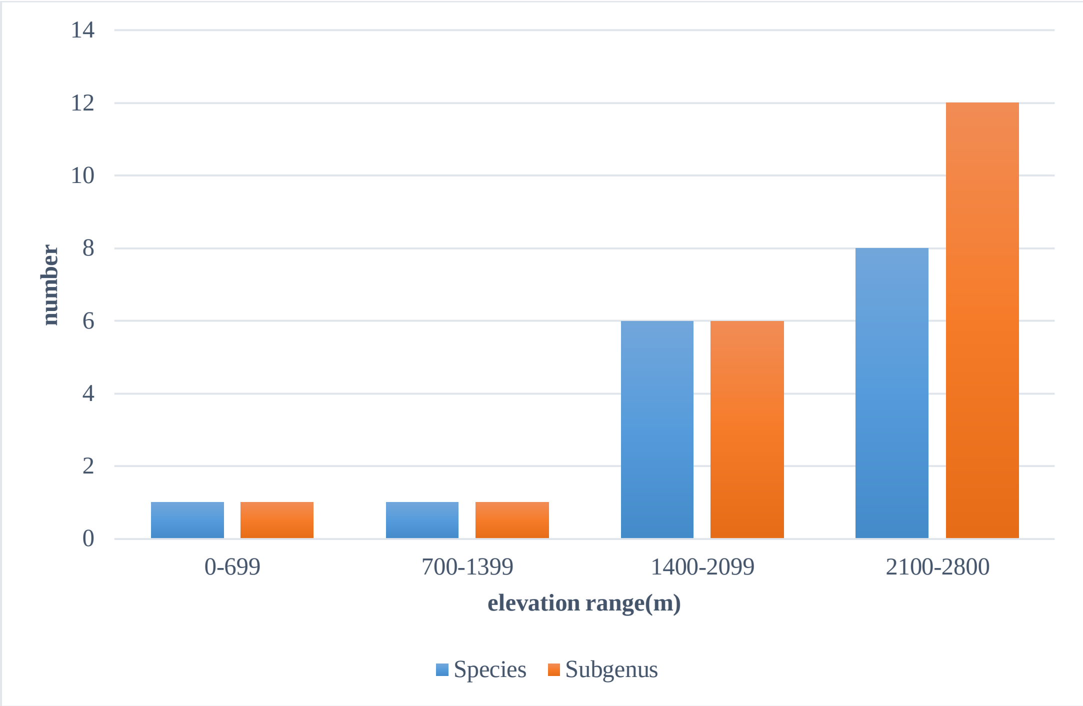

Figure 4. Species and Subgeneric diversity along the elevational gradient

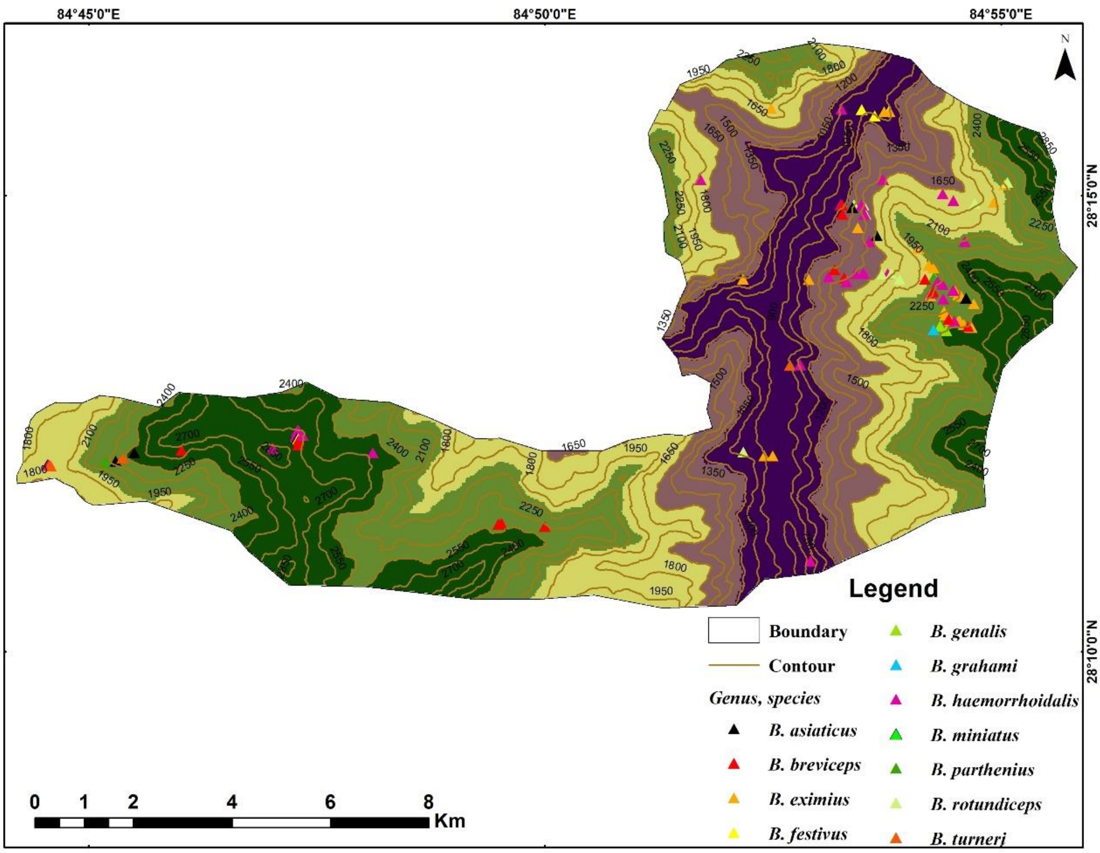

Figure 5: Bumblebees species recorded in different elevation categories

4.4 Habitat diversity

The distribution frequency of *Bombus* species had a significant relationship with the habitat used for forging. Hence, foraging of *Bombus* species is affected by habitat type. The dominant bumblebee species was *B. haemorrhoidalis* with the highest relative abundance in all the habitats. Likewise, *B. miniatus* and *B. pressus* were relatively least abundant and found foraging only in grassland. *B. genalis* and *B. parthenius* were only found foraging in grassland.

Table 1: Distribution frequency of *Bombus species* on different habitat using Pearson's chi-square test.

Species ⟹ Habitat ⇩	*asi*	*bre*	*exi*	*fes*	*gen*	*gra*	*hae*	*min*	*par*	*pre*	*rot*	*Tur*
agriculture	1	4	7	1	0	0	15	0	0	0	4	0
forest	3	0	17	0	2	3	9	0	2	0	3	6
grassland	2	5	4	0	0	2	6	1	0	1	1	1
human garden	1	0	1	2	0	0	4	0	0	0	0	0

Chi-squared = 60.52, df = 33, p-value = 0.002421

Abbreviations: *asi = asiaticus, bre =breviceps, exi = eximius, fes = festivus, gen= genalis, gra=grahami, hae = haemorrhoidalis, min = miniatus, par = parthenius, pre = pressus, rot = rotundiceps, tur = turneri*

Table 2: Diversity of bumblebee in different habitats

S.N	Habitats	Number species (S)	Shannon index (H)
1	Agriculture	69	1.32712
2	Forest	90	1.68186
3	Grassland	41	**1.9585**
4	Human garden	9	1.06086

The highest Shannon Wiener diversity index (H) was found in grassland while the lowest Shannon Wiener diversity index (H) was found in the human garden.

4.5 Relations of bumblebee with physical parameters

PCA was performed to know the relationship between the frequency of bumblebees with physical parameters (altitude, humidity, and temperature).

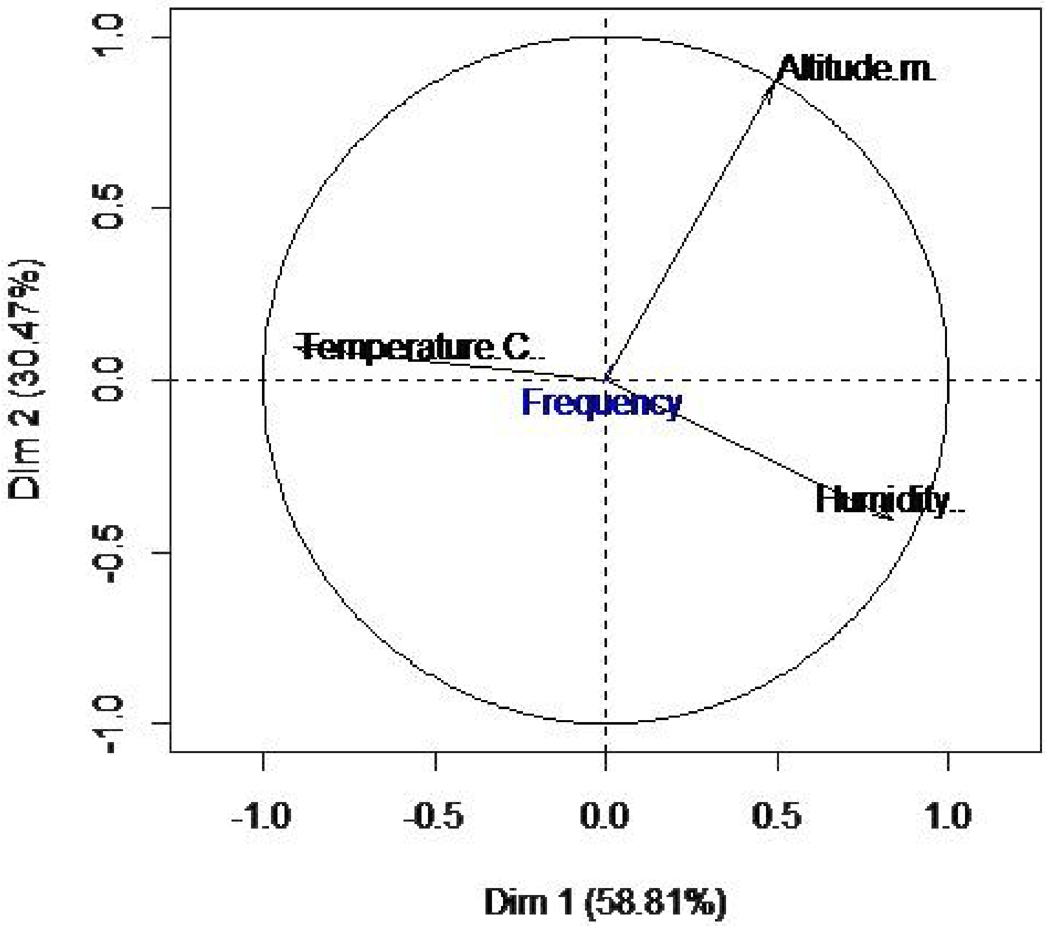

Figure 6: PCA showing a relationship between altitude, temperature, humidity, and frequency of bumblebee

The variable factor map explained by two canonical axes (Dim 1=58.81% and Dim 2= 30.47%) shows that the frequency of bumblebee is highly correlated with humidity and altitude. While negatively correlated with temperature.

4.6 Relation of bumblebee with host plant

During the study period, bumblebee individuals were found foraging upon different host plants belonging to 22 different families. Those host plants were found to be in agriculture, grassland, forest, and human settlement state in the study area. The color of the flowers of the host plants was found varied having eight different colors (Annex II).

Table: 3 One-way ANOVA performed to find out the relationship of bumblebee abundance with a family of host plants

Factor	Df	Sum Sq	Mean Sq	F -value	p-value
Family	21	81.59	3.885	4.028	**0.00000225 *****

Bumblebee relationship with the host plants family was found to be highly significant i.e. P< 0.00000225

Table: 4 Generalized Linear Model (GLM) performed to find out the relationship of bumblebee abundance with a specific family of host plants

Call: glm(formula = abundance~color=gaussian(identity)				
Coefficients	Estimate	Std. Error	t -value	p-value
(Intercept)	2.667	5.670	4.703	0.0000097 ***
Asteraceae	-5.417	6.179	-0.877	0.38315
Balsaminaceae	-1.282	6.291	-2.038	**0.04462 ***
Brassicaceae	1.667	1.134	-1.470	0.14531
Cactaceae	2.833	8.966	3.160	**0.00218 ****
Caprifoliaceae	-1.667	7.501	-2.222	**0.02892 ***
Cucurbitaceae	-9.167	6.649	-1.379	0.17159
Ericaceae	-3.333	8.019	-0.416	0.67869
Fabaceae	1.208	8.019	0.000	1.00000
Gesneriaceae	-4.167	7.501	-0.555	0.58002
Hypericaceae	-1.667	1.134	-1.470	0.14531
Lamiaceae	-1.042	6.649	-1.567	0.12088

Laureceae	-1.667	1.134	-1.470	0.14531
Magnoliaceae	3.333	1.134	0.294	0.76952
Malvaceae	-1.667	8.019	-2.078	**0.04066 ***
Melastomataceae	-1.667	8.966	-1.859	0.06646
Oleaceae	-5.758	6.397	-0.900	0.37062
Polygonaceae	4.266	6.945	0.000	1.00000
Ranunculaceae	-1.667	8.019	-2.078	**0.04066 ***
Rosaceae	-1.000	6.945	-1.440	0.15352
Solanaceae	-1.381	6.777	-2.038	**0.04467 ***
Verbenaceae	2.833	8.966	3.160	**0.00218 ****
AIC:	324			

It was found that bumblebee abundance has a highly significant relationship ($P<0.00000225$) with the family of the host plants. To know the relationship of the bumblebee with the host plants specific family, GLM analysis was performed. Host plants belonging to the family Cactaceae ($p<0.00218$) and Verbenaceae ($p<0.00218$) found to be significant for bumblebee abundance as foraging host plants.

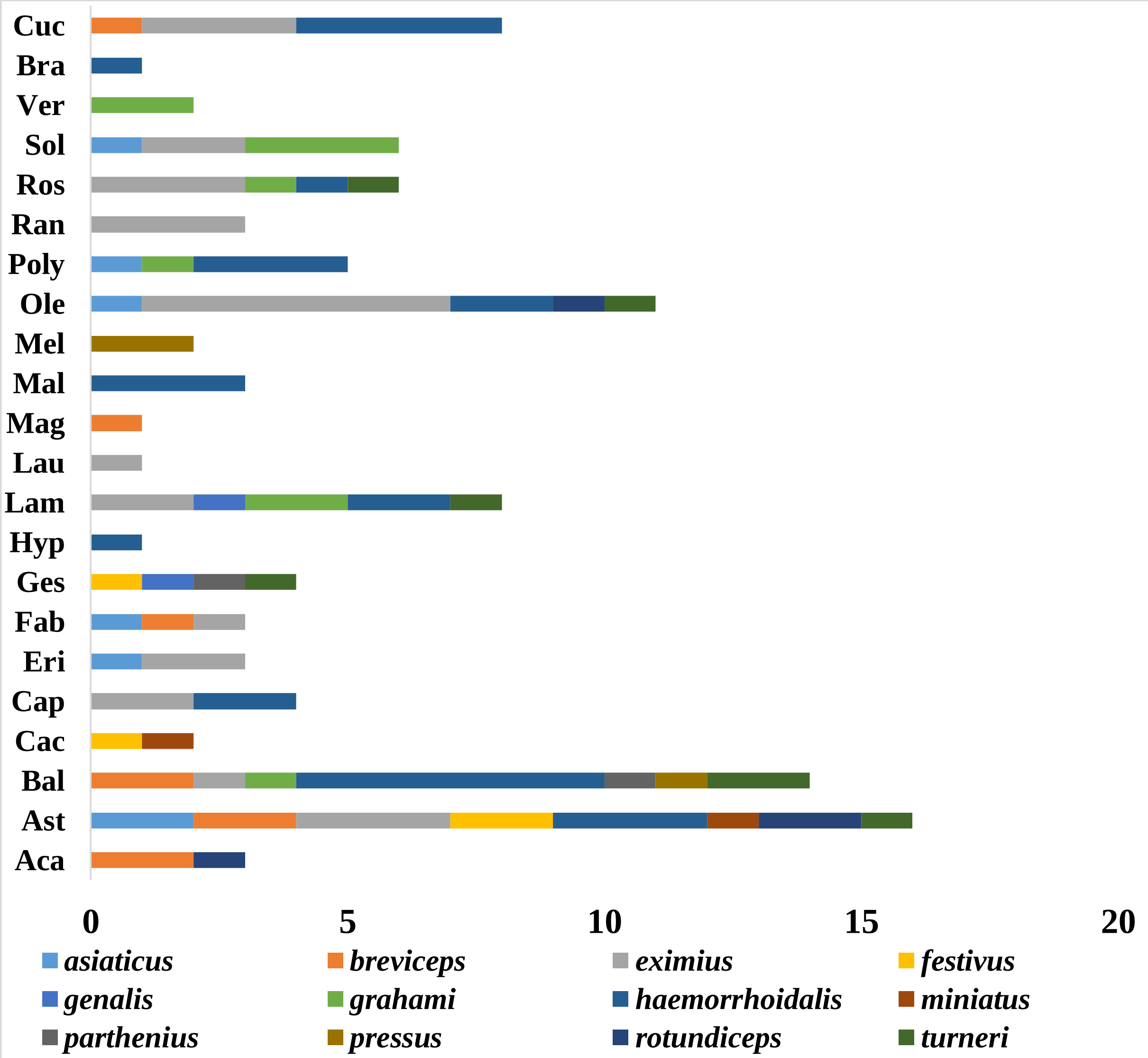

Figure 7: Bumblebee abundance with the family of host plants.

The highest abundance of bumblebee has been found foraging the host plants of family Asteraceae whereas the least abundance of bumblebee had been found foraging on host plants of Laureceae, Hypericaceae.

Table: 5 One-Way ANOVA performed to find out the relationship of bumblebee abundance with flower color of host plants

Factor	Df	Sum Sq	Mean Sq	F value	P-value
Host plant color	7	36.51	4.564	3.529	**0.00124 ****

Bumblebee abundance has a significant relationship ($p<0.00124$) with the color of the flower of host plants.

To find out the relationship of bumblebee abundance with the specific color of the flower of host plants, GLM analysis was performed. The flower with color reddish-yellow was found to be significant ($p<0.00154$) for the bumblebee abundance.

Table: 6 Generalized Linear Model (GLM) performed to find out the relationship of bumblebee abundance with specific flower color of host plants

Call: glm(formula = abundance~color=gaussian(identity))				
Coefficients:	Estimate	Std. Error	T-value	p-value
Intercept	2.4000	0.5086	4.719	0.0000078 ***
pink	-0.5429	0.6659	-0.815	0.41689
purple	-0.6500	0.6483	-1.003	0.31850
purple brown	-1.4000	1.2458	-1.124	0.26381
red	-1.0429	0.5925	-1.760	0.08147
reddish-yellow	3.1000	0.9515	3.258	**0.00154 ** **
White	-0.2621	0.5507	-0.476	0.63519
whitish-purple	-1.1143	0.6659	-1.673	0.09741
yellow	-0.4571	0.5437	-0.841	0.40248

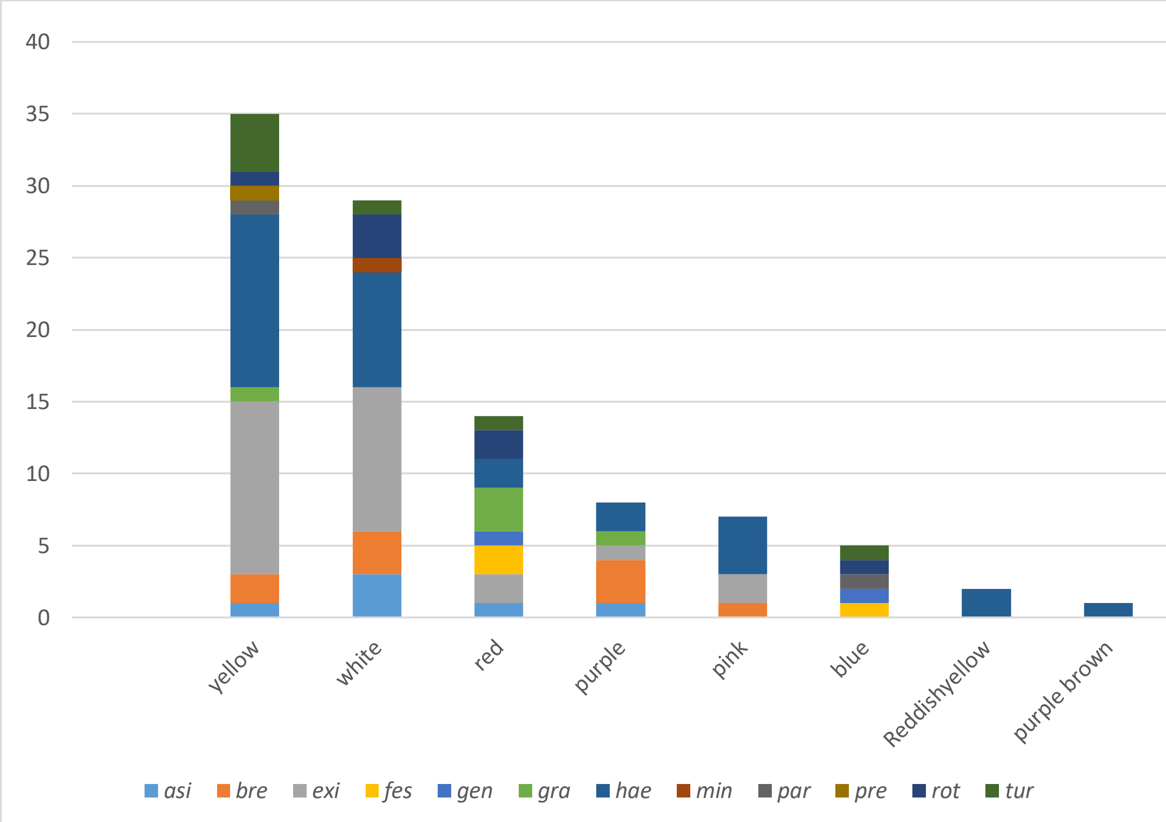

Figure 8: Bumblebee preferences with flower color of host plants

Abbreviations: *asi* = *asiaticus*, *bre* =*breviceps*, *exi* = *eximius*, *fes* = *festivus*, *gen*= *genalis*, *gra*=*grahami*, *hae* = *haemorrhoidalis*, *min* = *miniatus*, *par* = *parthenius*, *pre* = *pressus*, *rot* = *rotundiceps*, *tur* = *turneri*

Bumblebees were found visiting on eight different flower colors. Among which bumblebee species were found frequently foraging on yellow flowers followed by white flowers. While the purple-brown flower was least visited by the bumblebees.

Table 7. Bumblebee abundance relationship with the category (invasive/ non-invasive species) of plants by using one- way ANOVA

Factor	Df	Sum Sq	Mean Sq	F- value	P-value
invasive/non-invasive	1	4.69	4.694	3.113	**0.0806**

The abundance of the bumblebee had a no-significant relationship (P<0.0806) with the category (invasive/ non-invasive species) of the foraged flowering plants.

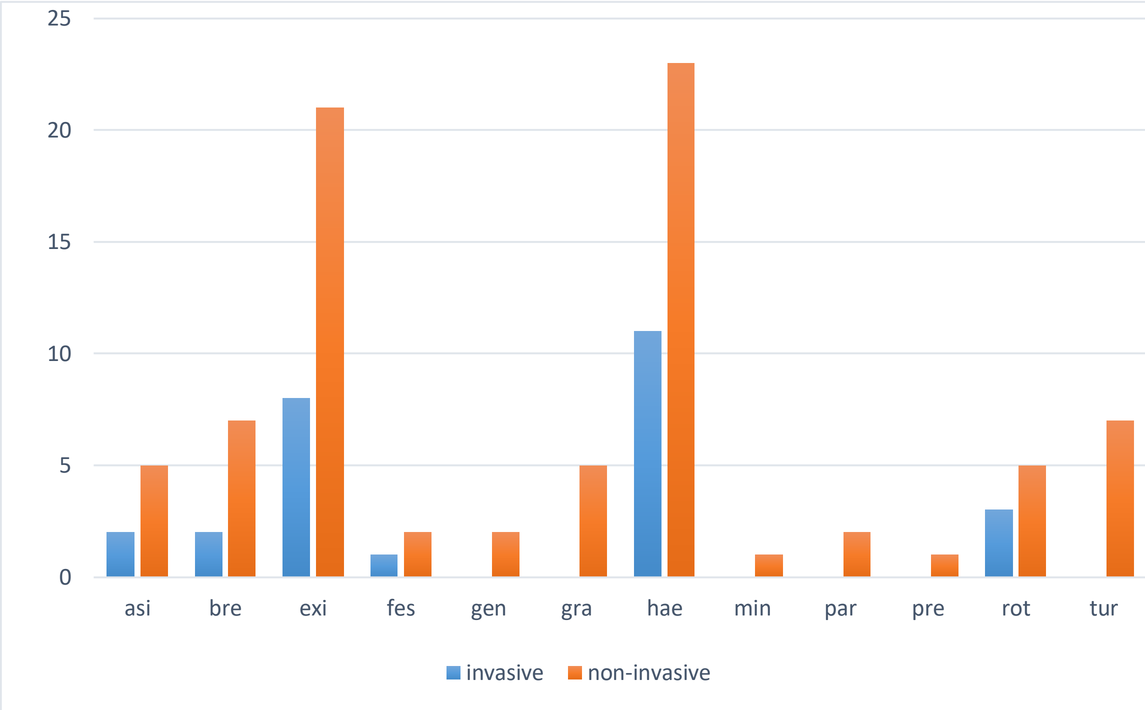

Figure 9: Abundance of bumblebee with categories (invasive/ non-invasive species) of host plants

Abbreviations: *asi* = *asiaticus*, *bre* =*breviceps*, *exi* = *eximius*, *fes* = *festivus*, *gen*= *genalis*, *gra*=*grahami*, *hae* = *haemorrhoidalis*, *min* = *miniatus*, *par* = *parthenius*, *pre* = *pressus*, *rot* = *rotundiceps*, *tur* = *turneri*

The highest abundance of the bumblebee was found foraging on non-invasive species of foraged flowering plants while the lowest abundance of bumblebee was observed on the invasive species plant.

4.7 Foraging time of bumblebees

The time of foraging bumblebees was divided into morning, afternoon, and evening to detect the highest foraging time of bumblebees.

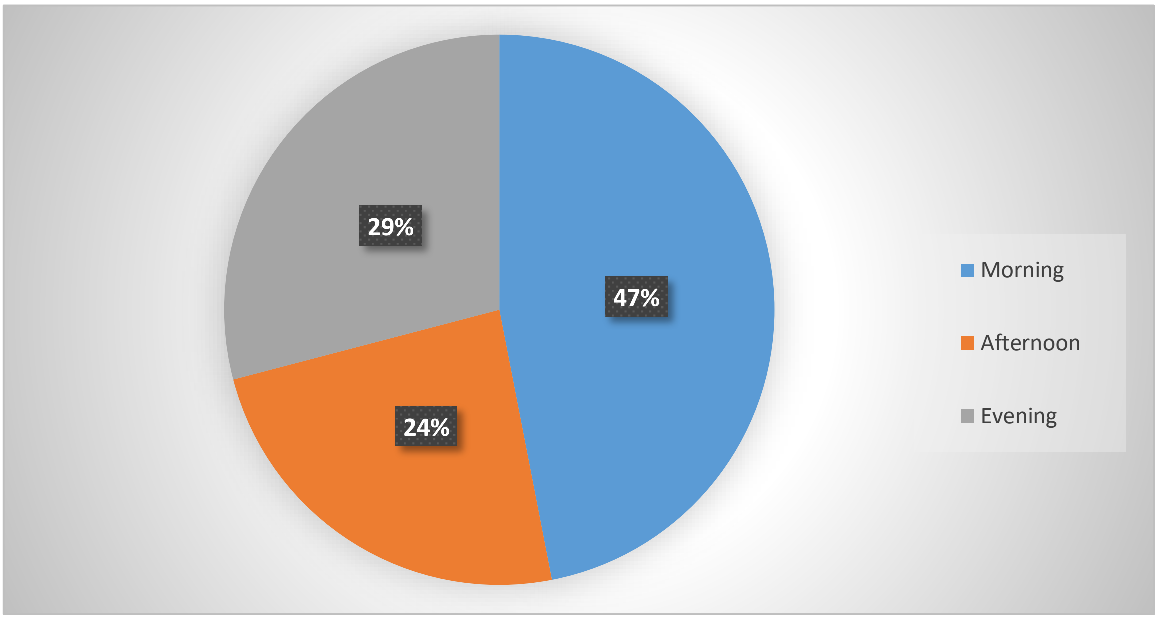

Figure 9: Foraging activity of bumblebees

Bumblebees were found foraging maximum at morning time as compared to afternoon and evening.

5. DISCUSSION

5.1 Diversity and Distribution

This survey represents the first systematic study of bumblebee diversity in the Gorkha region of Chitwan Annapurna Landscape(CHAL). Among 209 individuals, 12 species of eight genera were identified. It represents 37.5% of the total 34 species of bumblebee from Nepal. Out of 8 subgenera of *Bombus*, *Melanobombus and Orientalibombus* were most abundant while *Pressibombus* subgenus was least abundant. *B. haemorrhoidalis* was reported to be the most abundant species. A similar result was obtained in the eastern as well as western Himalaya (Sinu et al. 2011). Likewise, Bhusal et al. (2019) also obtained *B. haemorrhoidalis* as the most abundant species in the Kathmandu valley. *B. genalis, B. miniatus,* and *B. pressus* were low in abundance in the study area.

The diversity of bumblebee was found greater at grassland (1.9585) and forest (1.68186) habitats while low in the home garden (1.06086). *B. asiaticus* was found as foraging in all from the study site which is similar to Sabir et al. (2011). Species richness, species diversity, and abundance of the bumblebee have a positive correlation to flower frequency (Gunnarsson and Federsel 2014). Despite low food abundance relatively higher species richness is found in forest and grassland habitats because of the diversity of resources that provide food over a long period (Walther-Hellwig and Frankl 2000, Quaranta et al. 2004, Pywell et al. 2005). These habitats are critical for species richness and abundance of bumblebee in agro-ecosystem (Mand et al. 2002) as it supplies pollen and nectar resources continuously throughout the spring and summer months only (Steffan- Dewenter and Tscharntke 1999). These habitats also provide more undisturbed and protected nesting habitats for reproduction and overwintering than agricultural habitat (Backman and Tiainen 2002, Quaranta et al. 2004, Pywell et al. 2005). Hence, also known as refuge habitats that show a positive correlation with the diversity of wild bee fauna (Banaszak 1996). On the other hand, agricultural habitat has a negative effect in bumblebees due to fragmentation and habitat destruction which results in loss of essential continuous floral resources (nectar and pollen) and suitable nesting habitats. There is also a greater possibility of toxicological effects of insecticides in agriculturally dominated landscapes (Thompson and Hunt 1999). The home garden has the lowest bumblebee visitors which correlates with (Reininghaus 2017) as ornamental plants used for home gardens are with low nectar and pollen resources.

Covering a 500-2,800m range of altitudes and habitat types, there was found clear patterns of species-specific altitude ranges. *B. pressus* species were only found at the high elevations, and they occupy similar altitude niches as in other regions of the world (Williams et al. 2009, 2010). Highest species diversity at altitudes between 2,500-2,800 m, in contrast to William et al. (2010). They found the highest species diversity at altitudes between 3,000-4,000m. In general, species diversity was found to decline towards lower elevations and in the lowland (<1,000 m), only one species (*B. haemorrhoidalis* – 598m) was recorded, which represents the lowest elevations at which these species, and bumblebees in general, has ever been recorded in the Himalayan range (Williams 1991, Williams et al. 2010). Bumblebees often occur in a wide altitudinal range, but only a few species reach the tropical lowland, where conditions are usually unfavorable for these cold-adapted bees (Williams 1991, Gonzalez et al. 2004, Williams et al. 2009). *B. haemorrhoidalis,* and *B. breviceps* cover a wide range of altitudes and usually were most abundant at medium elevations. A similar result was obtained in Arunachal Pradesh (Streinzer et al. 2018).

5.2 Relation of bumblebee with physical parameters

Bumblebee frequency was found positively correlated with altitude and humidity while negatively correlated with temperature. The study correlates with (Peat and Goulson 2005) who stated that foraging rates of *Bombus terrestris* were positively correlated with humidity while temperature did not significantly influence the foraging rate. A positive correlation between foraging of bumblebee and humidity is because nectar secretion rates were higher at high humidity (Peat and Goulson 2005). They also suggested that bumblebee avoid collecting pollen when there is dew or rain-water droplets on the vegetation which would make grooming pollen into the corbiculae difficult. Also, bumblebee's frequency was found positively correlated with altitude which also correlates with (Williams et al. 2010, Streinzer et al. 2018) who has concluded species diversity of bees increases with altitude. In contrast with (Hoiss et al. 2012) who concluded that species richness and abundance of bees showed a linear decline with increasing altitude because the diversity of species declines linearly with decreasing temperatures. But bumblebees can forage at extremely low temperatures (AllenWardell et al. 1997, Saini et al. 2012) so their frequency was found negatively correlated with temperature and positively correlated with altitude in the study area.

5.3 Relation of bumblebee with their host plant

5.3.1 Foraging of bumblebee in different families and categories of plants

The variation in the distribution in different locations with different flora showed a diverse floral host range for the collection of nectar and pollen of this species (Nagano et al. 2014). The species richness and abundance of bumblebees are significantly affected by types of flower resources, indicating that there is a strong association of bumblebees with families of host plants across the study area. Many explanations have pointed out how flora characters such as morphology, color, and scent are associated with the attraction of the bee communities (Stone et al. 2003, Cnaani et al. 2006, Fornoff et al. 2017). Elliott (2009) showed the relation of foraging distances, bumblebee, and flower availability (Williams and Osborne 2009) indicating that species richness of bumblebee depends on increasing flower cover and the type of host plant and their richness compared to the other factors. The floral resources are the factors that alter the species richness, species diversity, and abundance of the bumblebees (Potts et al. 2010). The reduction in species richness, evenness, and abundance of the bumblebee is due to habitat fragmentation and reduction in the floral resources (Goulson et al. 2008, Xie et al. 2008), but Inouye (1980) studied the effect of morphological characters such as corolla length and proboscis on the rate of flower visitation by bumblebees where short-tongued bees foraged faster on short-corolla flowers than long-tongued bees. Differential significant effect of plant families and the nature of flowers on the abundance of bumblebees is probably associated with the specific floral characters of that particular family and nature of flowers was found during the study. No statistically significant effect of categories of host plants (invasive and noninvasive) was observed in the study, especially for the abundant species of bumblebee. The bumblebee species were found foraging abundantly in non-invasive categories of plants, while the invasive species, *Bidens pilosa* was found as a continuous floral resource during the study period. The families, Asteraceae, Oleaceae, and Cucurbitaceae were the most visited families by the bumblebee.

5.3.2 Flower color preferences by the bumblebees

This study has a significant effect on the color of flowers for the abundance of bumblebees, indicating the differential color preference by bumblebee species in the area. Colors appear to be particularly important to bumblebee for flower recognition (Menzel 1967, Menzel and Backhaus 1991, Menzel and Shmida 1993). Bumblebee spp. show significant ($p=0.00124$) relationship with flower color. They show a difference in preference towards flower color. Bumblebees were found foraging mostly on yellow color flowers followed by white color flowers among eight different types of host flower color. Similarly, this result somehow correlates with Keasar et al. (1997) who stated that the flower color preference of bumblebee is blue, green, and white respectively. This study shows similarity to the result of Odell et al. (1992) which stated that pollinator visits yellow color flower than white color flowers as yellow color flower emits methyl benzoate which is one of the major floral volatiles to attracts pollinators. Color of flower guides bees from distance. The reason for the preference of white and yellow color flower by bumblebee is because bumblebee prefers more saturated color over less saturated one (Rohde et al. 2013) and also colors of high purity (Lunau et al. 1996). Similarly, the result shows agreement towards bumblebee's innate color preferences that contrast with the background (Spaethe et al. 2001, Dyer and Chittka 2004). The possibility of preference of white and yellow color flowers maybe because white melittophilous flowers regularly absorb UV light (Kevan et al. 1996) and bees have a trichromatic color vision in the UV light (Spaethe et al. 2001, Dyer and Chittka 2004). On the other hand, the result of the study area shows the purple color as least visiting for foraging of a bumblebee which is in contrast to (Raine and Chitka 2007) who stated preference of violet/ purple color over the blue color flower as the violet color flowers produce 41% more nectar than the blue color flowers (the next rewarding flower color after violet). The reason for the purple-brown color flower as least visiting for foraging in the study site is because of the least abundance of purple color flower than white and yellow color flowers.

5.4 Foraging time of a bumblebee

The foraging time of bumblebee was higher in the morning time then decreasing during the afternoon and gradually increasing in the evening. The result seems correlated with Abak et al. (2000) who have recorded the foraging activity of bumblebee as pollinators for eggplants in increasing manner between 9.00 am – 11.00 am reaching the peak activity between 10.00 am and 11.00 am. Then, it gradually decreases and ceases between 1.00 pm – 2.00 pm and again started foraging in the evening between 3.00 pm-6.00 pm. The result also correlates with Willmer et al. (1993) who have stated bumblebee species prefer young flowers strongly who offered both pollen and nectar especially early in the morning when pollen was abundant. In contrast, Peat and Goulson (2005) stated that pollen was preferred to be collected when the day was warm and humidity was low during the middle of the day. The foraging activity of bumblebee was high during morning time after the sunrise because bumblebee avoids collecting pollen when there is dew or rain-water droplets on the vegetation which would make grooming pollen into the corbiculae difficult.

6. CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The present study has been carried out to explore the species diversity and distribution of bumblebees in the Gorkha region of CHAL. Among 209 individuals, 12 species of eight genera were identified from the study area. This study concluded that *B. haemorrhoidalis* was the most abundant species whereas, *B. pressus* was the least abundant species. Foraging of *Bombus* species is affected by habitat type. The dominant bumblebee species was *B. haemorrhoidalis* with the highest relative abundance in all the habitats. *B. miniatus* and *B. pressus* were relatively least abundant and found foraging only in grassland. *B. genalis* and *B. parthenius* were only found foraging in grassland. Foraging of bumblebee was found positively correlated with altitude and humidity while negatively correlated with temperature. Bumblebee was found to have a significant relationship with foraging host plant families. Host plants of family Cactaceae and Verbenaceae were found to be significant for bumblebee abundance. Bumblebee abundance was also found to have a significant relationship with the flower color of host plants. Reddish yellow flower color was found to be significant for the bumblebee abundance. The bumblebee species was found foraging abundantly in non-invasive categories of plants. Bumblebees were found foraging maximum at morning time as compared to afternoon and evening.

6.2 Recommendations

The following are some of the recommendations drawn from the study which will be helpful in the conservation of bumblebee species within the study area.

- A detailed study is needed to reach more conclusions for the species diversity, distribution, ecology, and threats of bumblebee in the other parts of the Gorkha region of CHAL.
- Morphologically, cryptic species pose significant problems for the field assessment of species richness and diversity. It is better to identify species by DNA barcoding, especially in a similar type of color pattern groups.

7.REFERENCES

Abak, K., Ozdogan, A.O., Dasgan, H.Y., Derin, K., and Kaftanoglu, O. 2000. Effectiveness of bumblebees as pollinators for eggplants grown in unheated greenhouses. ISHS Acta Horticulturae **514**: 197-204.

Abrol, D.P. 1998. Mites associated with bumblebee *B. haemorrhoidalis* (Hymenoptera, Apidae, Bombini). Indian Bee Journal **60**(3): 153-154.

Allen-Wardell, G., Bernhardt, P., Bitner, R., Burquez, A., Buchmann, S., Cane, J. et al. 1997.The potential consequences of pollinator declines on the conservation of biodiversity and stability of food crop yields. Conservation Biology **12**(1): 8-17.

An, J., Huang, J., Shao, Y., Zhang, S., Wang, B., Liu, X., Wu, J. Williams, P.H. 2014. The bumblebees of North China (Apidae, *Bombus Latreille).* Zootaxa **3830**(1): 001-089.

Backman, J.C., and. Tiainen, J. 2002. Habitat quality of field margins in a Finnish farmland area for bumblebees (Hymenoptera: Bombus & Psithyrus). Agriculture, Ecosystems, and Environment **89** (1-2): 53-68.

Banaszak, J. 1996. Ecological bases of conservation of wild bees: the conservation of bees 1st ed. Academic Press, London, UK 55-62p.

Bhusal, D.R., Ghimire, K.C., Dulal, S., Baniya, P., and Thakur, S. 2019. Spatial Relation of Bumblebees(Hymenoptera-Apidae) with Host-Plant and their Conservation Issues: An Outlook from Urban Ecosystem of Kathmandu Valley, Nepal. European Journal of Ecology **5**(2): 1-7

Biesmeijer, J.C., Robert, S.P.M., and Reemer, M. 2006. Parallel declines in pollination and insect-pollinated plants in Britain and the Netherlands. Science **313**(5787): 351- 354.

Borror, D.J., Delong, D. M. and Triplehorn, C.A. 1981. An introduction to the study of the insects 5th ed. The Dryden Press, Philadelphia 705p.

Buchmann, S., Nabhan, G., and Ingram, G. 1996. Our Forgotten Pollinators: Protecting the Birds and Bees. Apiservices **6**: 1-4.

Buchmann, S.L. 1983. Buzz pollination in angiosperms. In Handbook of Experimental Pollination Biology. Edited by Jones CE, Little RJ. van Nostrand Reinhold Company, Inc, **1983**: 73-113.

Cane, J. H. 2001. Habitat fragmentation and native bees: a premature verdict. Conservation Ecology **5**(1): 3.

Cannani, J., Thomson, J.D. and Papaj, D.R. 2006. Flower choice and learning in foraging bumblebees: Effect of variation in nectar volume and concentration. Ethology **112**: 278-285.

Carvell, C., Roy, d.B., Smart, S.M., Pywell, R.F., Preston, C.D., and Goulson, D. 2011. The decline in forage availability for bumblebees at a national scale. Biological Conservation, **132**: 481-489.8

Cnaani, J., Thomson J.D., Papaj, D.R. (2006) Flower choice and learning in foraging bumblebees: effects of variation in nectar volume and concentration. Ethology, **112**(3):278–285

Colla, S.R., Richardson, L., Williams, P.H. 2011. Bumblebees of the Eastern United States. United States Department of Agriculture 104p.

Corbet, S.A., Williams, I.H., Osborne, J.L. 1991. Bees and the pollination of crops and wildflowers in the European Community. Bee World **72**: 47-59.

Cozmunta A.M., Bretan L., Cozmuta L.M., Nicula C., Peter A., 2012. Lead traceability along soilmelliferous flora-bee family-apiary products chain. J. Environment. Monitoring, **14**(6): 1622- 1630.

Crosswhite, F.S. and Crosswhite, C.D. 1970. Pollination of *Castilleja sessiflora* in southern Wisconsin Bulletin of the Torrey Botanical Club, **97**:100-105.

Daka, T.N., Sudarshan, M.R., Saju, K.A. 2011. New record of bumblebees, B. breviceps Smith as a pollinator of large cardamom. Current Science **100**: 926-928.

Dramstad, W. E. 1996. Do bumblebees (Hymenoptera: Apidae) forage close to their nests? Journal of Insect Behaviour **2**: 163–182.

Dyer, A. G., and Chittka, L. 2004. The biological significance of distinguishing between similar colors in spectrally variable illumination: bumblebees (Bombus terrestris) as a case study. Journal of Comparative Physiology A **190**(2):105-114.

Elliot, S. 2009. Subalpine bumblebees foraging distances and densities with flower availability. Environmental Entomology **38**: 748-756.

Faegri, K., and Vander, P. L. 1979. The principles of pollination ecology (3rd ed.). pergamon Press Ltd.

Fontaine, C., Dajoz, I., Meriguet, J., and Loreau, M. 2006. The functional diversity of plant-pollinator interaction webs enhances the persistence of plant communities. PLoS Biol. **4**(1): e1.

Fornoff, F., Klein, A.M., Hartig, F., Benadi, G., Venjakob, C., Schaefer, H.M., Ebeling, A. 2017 Functional flower traits and their diversity drive pollinator visitation. Oikos, **126**(7):1020–1030

Free, J. B., Williams, I. H. 1976. Pollination as a factor limiting the yield of field beans (Vicia faba L.). Journal of Agricultural Science, 87: 395-399.

Free, J.B. 1993. Insect Pollination of Crops 2nd ed. Academic Press, London, 684p.

Free, J.B., Williams, I.H. 1976. Pollination is a factor limiting the yield of field beans (*Vicia faba* L.). J. Agricultural. Science **87**: 395-399.

Fussell, M., and Corbet, S. A. 1992. Flower usage by bumblebees a basis for forage plant management. Journal of Applied Ecology **29:** 451–465.

Garibaldi LA, et al. 2013. Wild pollinators enhance fruit set of crops regardless of honey bee abundance. Science **339**(6127):1608–1611.

Gegear, R. J., and Laverty, T. M. 2005. Flower constancy in bumblebees: a test of the trait variability hypothesis. Animal Behaviour **69:** 939–949.

Giurfa, M., Núñez, J., Chittka, L., and Menzel, R. 1995. Color preferences of flower native honeybees. J. Comp. Physiol, A **177**:247-259.

Giurfa, M., Vorobyev, M., Kevan, P., and Menzel, R. 1996. Detection of colored stimuli by honeybees: minimum visual angles and receptor-specific contrasts. J. Comp. Physiology **178:** 699-709.

Gonzalez, V. H., A. Mejia, and C. Rasmussen. 2004. Ecology and nesting behavior of Bombus atratus Franklin in Andean highlands (Hymenoptera: Apidae). J. Hymenoptera. Res. **13**: 28–36.

Goulson, D. 2010. Bumblebees behavior, ecology, and conservation 2nd ed. Oxford University Press, New York, 330p.

Goulson, D., Henly, M.E., Darvill, B., Ellis, J.S., Knight M.E. 2005. Causes of rarity in bumblebee. Biological Conservation **122**: 1-8.

Goulson, D., Lye, G.C., and Darvill. 2008. Diet breadth, coexistence, and rarity in bumblebees. Biodiversity Conservation, **17**(13): 3269-3288.

Greigh- Smith, P. 1983. Quantitative plant ecology. University of California Press, Barkeley, California, U.S.A.

Gunnarsson, B. and Federsel, L.M. 2014. Bumblebees in the city abundance, species richness, and diversity in two urban habitats. Journal of Insect Conservation **18**(6): 1185-1191.

Heinrich, B. 1972a. The pattern of endothermy in bumblebee queen, drone, and worker. Journal of Comparative Physiology **77**: 65-79.

Heinrich, B. 1972b. Physiology of brood incubation in the bumblebee queen *Bombus vosnesenskii*. Nature **239**: 223-225.

Heinrich, B. 1976. The source specialization of individual bumblebees. Ecological Monographs **46**: 105-128.

Heinrich, B., Mudge, P.R. and Deringis, P.G. 1977. Laboratory analysis of flower constancy in foraging bumblebees: *Bombus ternarius* and *B. terricola*. Behavior. Ecology. Sociobiology **2**(3): 247– 265.

Hoiss, B., Krauss, J., Potts, S. G., Roberts, S. and Steffan-Dewenter, I. 2012. Altitude acts as an environmental filter on phylogenetic composition, traits, and diversity in bee communities. In: Hoiss, B., Krauss, J., Potts, S. G., Roberts, S. and Steffan Dewenter, I. eds. Proceedings of Royal Society B: Biological Science, 5th September 2012, Wurzburg, Germany. The Royal Society publishing: 4447–4456.

Inouye, D.W. 1980. The effect of proboscis and corolla tube length on patterns and rates of flower visitation by bumblebees. Oecologia, **45**: 197-201.

Inouye, D.W. 1983. The ecology of nectar robbing. The Biology of Nectaries2nd ed. Columbia University Press, New York, 152–173p.

Kearns, C. A., and J. D. Thomson 2001. The Natural History of Bumblebees: a sourcebook for investigation, first edition. Boulder: University Press of Colorado. Kevan, P. G. 1976. Fluorescent nectar (technical comment). Science **194**: 341–342.

Keasar, T., Bilu, Y., Motro, U., and Shmida, A. 1997. Foraging choices of bumblebees on equally rewarding artificial flowers of different colors. Israel Journal of Plant Sciences **45(**2-3): 223-233.

Kells, A.R., and Goulson, D. 2003. Preferred nesting site of bumblebee queen (Hymenoptera: Apidae) in agroecosystems. Biological Conservation **109**: 164-174.

Kevan, P. G., and T. P. Phillips. 2001. The economic impacts of pollinator decline: an approach to assessing the consequences. Conservation Ecology **5**(1): 8.

Kevan, P. G., Giurfa, M., and Chittka, L. 1996. Why are there so many and so few white flowers? Trends Plant Science, **1**:280-284.

Kevan, P.G., Grec, C.F., and Belaoussoff, S. 1997. Log-normality of biodiversity and abundance in diagnosis and measuring of ecosystemic health: pesticide stress on pollinators on blueberry heaths. Journal of Applied Ecology **34**(5): 1122–1136.

King, M.J. 1993. Buzz foraging mechanism of bumblebees. Journal of Apicultural Research **32**: 41–49.

Kleijn, D. and Raemakers, I. 2008. A retrospective analysis of pollen host plant use by stable and declining bumble bee species. Ecology **89**(7) 1:811–1823.

Kopij, G. 2002. The bird of Schlabathebe National Park, Lesotho. Koedoe **45**(1): 65-75.

Lunau, K., Wacht, S., and Chittka, L. 1996. Color choices of naive bumblebees and their implications for color perception. Journal of Comparative Physiology A **178**:477- 489.

Mand, M., Mand, R., and Williams, I. H. 2002. Bumblebees in the agricultural landscape of Estonia. Agriculture, Ecosystems, and Environment **89**(1-2): 69-76.

Mcfrederick, Q.S., Lebuhn, G. 2006. Are urban parks refuges for bumblebees? Biol. Conserv **129**: 372-382.

Mcgreror, S.E. 1976. Insect pollination of cultivated crop plants, Agricultural Handbook, U.S.Dept. Agriculture, No. **46.**

Menzel, R. 1967. Untresuchungen zum erlernen von spektralfarben durch die Honigbiene (Apis mellifera). Zeitschrift fuer Vergleichende physiologie **56**: 22-62.

Menzel, R., and Backhaus, W. 1991. Color vision in insects. In Vision and visual dysfunction, The perception of color, Gourras, P. (eds.). Macmillan, London, p. 262-288.

Menzel, R., and Shmida, A. 1993. The ecology of the flower color and the natural color vision of insect pollinators: The Israeli flora as a study case. Biological Revolution **68**: 81-120.

Mol1er, A. P. 1995. Bumblebee preference for symmetrical flowers. Proceedings of National Academy of Sciences USA **92**: 2288-2292.

Mol1er, A. P., and Sorci, G. 1998. Insect preference for symmetrical artificial flowers. Oecologia **114**: 37-42.

Nagano, Y., Abe, K., Kitazawa, T., Hattori, M., Hirao, A.S., Itino, T. 2014

Odell, E., Raguso, R. A., and Jones, K. N. 1992. Bumblebee foraging responses to variation in floral scent and color in Snapdragons (Antirrhinum: Scrophulariaceae). The American Midland Naturalist **142**(2):257-266.

Ohashi, K., and Yahara, T. 1998. Effects of variation in flower number on Pollinator visits in *Cirsium purpuratum* (Asteraceae). American Journal of Botany **85**(2): 219–224.

Osborne, J. L., Clark. S.J., Morris, R.J., Williams I.H., Riley, J.R., Smith, A.D., Reynolds, D. R. and Edwards, A. S. 1999. A landscape-scale study of bumblebees foraging range and constancy, using harmonic radar. Journal of Applied Ecology **36**: 519- 533.

Osborne, J.L., Martin, A.P., Shortall, C.R., Todd, A.D., Goulson, D., Knight, M.E., Hale, R.J. and Sanderson, R.A. 2008b. Quantifying and comparing bumblebee nest densities in gardens and countryside habitats. Journal of Applied Ecology **45**: 784–792.

Peat, J., and Goulson, D. 2005. Effects of experience and weather on foraging rate and pollen versus nectar collection in the bumblebee, Bombus terrestris. Behavioral Ecology and Sociobiology **58**(2): 152-156.

Pekkarinen, A., Varvio-Aho, S.L., and Pamilo, P. 1979. Evolutionary relationships in northern European *Bombus* and *Psithyrus* species (Hymenoptera, Apidae) studied based on allozymus. Annales Entomologicae Fennici **45**: 77-80.

Plowright, R. C., and Laverty, T. M. 1984: The Ecology and Sociobiology of bumblebees. Annual Review of Entomology **29**: 175-199.

Potts, S.G., Biesmeijer, J.C., Kremen, C., Neumann, P., Schweiger, O., Kunin, W.E. (2010) Global pollinator declines trends, impacts, and drivers. Trends in Ecology & Evolution **25**(6):345–353

Pywell, R.F., Warman, E.A., Carvell, C., Sparks, T.H., Dicks, L.V., Bennett, D., et al. 2005. Providing foraging resources for bumblebees in intensively farmed landscapes. Biological Conservation **121**(4): 479–494.

Quaranta, M., Ambrowselli, S., Barro, P., Bella, S., Carini, A., Celli, G. et al. 2004. Wild bees in agroecosystems and semi-natural landscapes: 1997-2000 collection period in Italy. Bulletin of Insectology **57** (1): 11-61.

Raine, N. E., and Chittka, L. 2007. The adaptive significance of sensory bias in foraging context: Floral color preferences in the bumblebee Bombus terrestis. Plos One **2**(6): 556.

Reininghaus, H. 2017. Local and Landscape Effects on Arthropod Communities along an Arable-Urban Gradient. Ph. D. Thesis. Agricultural Sciences in Gottingen (PAG) at the Faculty of Agricultural Sciences, Georg-August-University Gottingen, Germany.

Richards, K. W. 1973. Biology of Bombus polaris Curtis and B. hyperboreus Schonherr' at Lake Hazen, Northwest Territories (Hymenoptera: Bombini). Quaestiones entomological **9**: 115-157.

Richards, K.W. 1978. Nest site selection by the bumblebees (Hymenoptera: Apidae) in Southern Alberta, The Canadian Entomologist **110**: 301-318.

Rohde, K., Papiorek, S., and Lunau, K. 2013. Bumblebees (Bombus terrestris) and honeybees (Apis mellifera) prefer similar colors of higher spectral purity over trained colors. Journal of Comparative Physiology A **199**:197–210.

Roka, E. 2019. Distribution and diversity of bumblebees in Chitwan Annapurna Landscape (CHAL), Nepal. M. Sc. Thesis. Central Department of Zoology, Tribhuvan University, Katmandu, Nepal.

Sabir, A.M., Suhail, A., Ahmed, S. and Khalid, S. 2011. Diversity of Bumblebees (Bombani Apidae: Hymenoptera) in Northern Pakistan. International Journal of Agriculture and Biology **13**: 159-166.

Sabir, A.M., Suhail, A., Rafi, A., Ahmad, S., Saleem, M., Mahmood, K. 2008. Bumblebees belonging to Genus Bombus (Bombus: Apidae: Hymenoptera) of Northern Pakistan.28th Pakistan Congress of Zoology (International) Government Collage University, Faisalabad, Pak. **74.**

Saini, M. S., Raina, R. H., and Ghator, H. P. 2015. Indian bumblebee 1st ed. Shiva Offset Press, Dehra Dun, India, 247 p.

Saini, M.S., Raina, R.H., and Khan, Z. H. 2012. Species diversity of bumblebee (Hymenoptera: Apidae) from different mountain regions of Kashmir Himalayas. Journal of Scientific Research **4**(1): 263-272.

Sakagami SF. 1976. Specific differences in the bionomic characters of bumblebees: a comparative review. J Fac Sci Hokkaido Univ VI Zool. **20**(3):390–447

Schweitzer, D.F., Capuan, N.A., Young, B.E., and Colla S.R. 2012. *Conservation and management of North American bumblebees.* NatureServe, Arlington, Virginia, and USDA Forest Service, Washington, D.C.

Sinu, P.A., Kuriakose, G., Shivanna, K.R. (2011) Is the bumblebee (Bom-bus haemorrhoidalis) the only pollinator of large cardamom in the central Himalayas, India Apidologie, **42**:690–695

Somme, L., Mayer, C., Jacquemart, A. L. 2014. Multilevel spatial structure impacts on the pollination services of *Comarum palustre* (Rosaceae). Plosone **9**(6): e99295. Doi: 10.1371/journal.pone.0099295

Spaethe, J., Tautz, J., and Chittka, L.2001. Visual constraints in foraging bumblebees: Flower size and color affect search time and flight behavior. In: Spaethe, J., Tautz, J. and Chittka, L. eds. Proceeding of the national academy of science, 27 March, 2001, United State of America: 3898-3903.

Steffan-Dewenter, I. and Tscharntke, T. 1999. Effects of habitat isolation on pollinator communities and seed set. Oecologia, **121**(3): 432–440.

Stevens, G. C. 1992. The elevational gradient in the altitudinal range: An extension of Rapoport's latitudinal rule to altitude. American Naturalist **140:** 893–911.

Stone, G.N., Raine, N.E., Prescott, M., Willmer, P.G. (2003) Pollination ecology of acacias (Fabaceae, Mimosoideae). Australian Systematic Botany **16**:103–118

Stout, J.C., Allen, J. A., Goulson, D. 2000. Nectar robbing, forager efficiency, and seed set: Bumblebees foraging on the self-incompatible plant *Linaria vulgaris* (Scrophulariaceae). Acta Oecologica **21** (4-5): 277−283.

Streinzer, M., Chakravorty, J., Neumayer, J., Megu, K., Narah, J. Schmitt, T. et al. 2018. Species composition and altitudinal distribution of bumblebees (Hymenoptera: Apidae: Bombus) in the East Himalaya Arunachal Pradesh, India.

Svensson, B., Lagerlof, J. and Svensson, B.G. 2000. Habitat preference of nest seeking bumblebees (Hymenoptera: Apidae) in an agricultural landscape. Agriculture, Ecosystem, and Environment **77(3)**: 247-255.

Thompson, H.M., and Hunt, V. 1999. Extrapolating from honeybee to bumblebee in pesticide risk assessment. Entotoxicology **8**:147-166.

Van den Eijnde, J. de Ruijter, A. and Van der Steen, J. 1991. Method for rearing *Bombus terrestris* continuously and the production of bumblebee colonies for pollination purposes. *Acta Horticulturae* **288**: 154–158.

Velthuis HHW, van Doorn A. 2006. A century of advances in bumblebee domestication and the economic and environmental aspects of its commercialization for pollination. Apidologie **37**(4):421–451.

Walther-Hellwig, K., and. Frankl, R. 2000. Foraging habitats and foraging distances of bumblebees, Bombus spp. (Hymenoptera, Apidae), in an agricultural landscape. Journal of Applied Entomology **124**: 299–306.

Waser, N. M. 1986. Flower constancy: definition, cause, and measurement. American Naturalist **127**: 593–603.

West, E. L., Laverty, T. M. 1998. Effect of floral symmetry on flower choice and foraging Behaviour of bumblebees. Canadian Journal of Zoology **76**: 730- 739.

Westphal, C., Steffan-Dewenter, I. and Tscharntke, T. 2003. Mass flowering crops enhance pollinator densities at a landscape scale. Ecology Letters **6**(11): 961–965.

Williams, I. H. 2002. Insect Pollination and Crop Production: A European Perspective. A report submitted to the Ministry of Environment, Brasília.

Williams, I.H. 1994. The dependence of crop production within the European Union pollination by honey bees. Agricultural zoology Review **6**: 229-257.

Williams, P. H. 1985. A preliminary cladistic investigation of relationships among the bumblebees' (Hymenoptera, Apidae). Systematic Entomology **10**: 239-255.

Williams, P. H. 1991. The bumblebees of the Kashmir Himalaya (Hymenoptera: Apidae, Bombini). Bulletin of the British Museum Natural History (Entomology), **60**(1): 1-204.

Williams, P. H. 1996. Mapping variations in the strength and breadth of biogeographic transition zones using species turnover. In Williams, P. H eds. Proceedings of the Royal Society of London Series B: Biological science, 22 May 1996, London, U. K. 579-588

Williams, P. H., and Osborne, J. L. 2009. Bumblebee vulnerability and conservation world-wide. Apidologie, **40:** 367-387.

Williams, P. H., Brstriakova, N., Huangi, J., Miao, Z., and Ani, J. 2015. Bumblebees, climate and glaciers across the Tibetan plateau (Apidae: Bombus Latreille). Systematics and Biodiversity **13**(2): 164-181.

Williams, P. H., Cameron, H. M., Cederberg, B., and Rasmont, P. 2008. A simplified subgeneric classification of the bumblebees (genus Bombus). Apoidologie, **39**(1): 46-74.

Williams, P. H., Huang, J. and An, J. 2017. Bear wasps of the middle kingdom: a decade of discovering China's bumblebees. Antenna **41** (1): 21-24.

Williams, P. H., Ito, M., Matsumura, T., and Kudo, I. 2010. The bumblebees of the Nepal Himalaya (Hymenoptera: Apidae). Insecta Matsumurana, **66:** 115-151.

Williams, P.H. 1985. A preliminary cladistic investigation of the relationship among the bumblebees (Hymenoptera, Apidae). Systematic Entomology **10**: 239-255.

Williams, P.H. 1986. Environmental changes and the distribution of British bumblebee (*Bombus* Latr.). Bee World **67**: 50s-61.

Williams, P.H. 1998. Bull. Natural History Museum. London UK. Entomology **67:** 79-152.

Williams, P.H. 2007. The distribution of the bumblebee color patterns worldwide: possible significance for thermoregulation, crypsis, and warning mimicry, Biological Journal of the Linnean Society **92**: 97-118.

Williams, P.H., and Osborne, J.L. 2009. Bumblebee vulnerability and conservation worldwide. Apidologie **40**: 367-387.

Willmer, P.G., Bataw, A. A. M. and Hughes, J. P. 1993. The superiority of bumblebees to honeybees as pollinators: insect visits to raspberry flowers. Entomological Journal **19**(3): 271-284.

Xie Z. Williams, P. H. and Tang, Y. 2008. The effect of grazing on bumblebees in the high rangelands of the eastern Tibetan Plateau of Sichuan. Journal of Insect Conservation **12:** 695–703.

Yang, D. 1999. The status of species diversity and conservation strategy of bumblebees, a pollination insect in Lancang River Basin of Yunnan, China, Chinese Biodiversity **7:** 170–174.

Annex I: Summary of the collected bumblebee specimens

Subgenus	Species	specimen	Q	W	M	Altitude range(m)
Alpigenobombus	*B. breviceps*	23	-	23	-	1571-2780
	B. genalis	3	3			2184-2349
	B. grahami	10	-	10	-	2241-2726
Melanobombus	*B. eximius*	62	-	62	-	1760-2592
	B. miniatus	3	-	3	-	2592
Festivobombus	*B festivus*	5	-	5	-	2614-2577
Orientalibombus	*B. haemorrhoidalis*	51	20	31	-	597-2592
Pyrobombus	*B. rotundiceps*	12	2	9	1	1656-2780
	B. parthenius	9	-	9	-	2191-2554
Psithyrus	*B. turneri*	9	-	9	-	2096-2592
Pressibombus	*B. pressus*	2	-	2		2737
Sibiricobombus	*B. asiaticus*	7	-	7	-	2066-2739

Abbreviation: Q - number of queens, W - number of workers, M - number of males

Annex II: Plant species with their respective family, flower color and categories (invasive/non-invasive) of the flowering plants visited by the bumblebees

Plant species	**Family**	**Color**	**Categories**
Parochetus communis	Fabaceae	White	Non-invasive
Dahlia pinnata	Asteraceae	Red	Invasive
Imatiens stenantha	Balsaminaceae	White	Non-invasive
Anaphalis contorta	Asteraceae	White	Non-invasive
Anaphalis busua	Asteraceae	White	Non-invasive
Aconogonum molle	Polygonaceae	White	Non-invasive
Rubus nepalensis	Rosaceae	Red	Non-invasive
Cercium verutum	Malvaceae	Purple	Invasive
Corydalis casimiriana	Papaveraceae	Yellow	Non-invasive
Strobilanthes attenuta	Acanthaceae	Purple	Non-invasive
Imatiens scabrida	Balsaminaceae	Purple	Non-invasive
Anemone elongata	Asteraceae	White	Non-invasive
Gaultheria fragrantissima	Ericaceae	White	Non-invasive
Anemone elongata	Ranunculaceae	White	Non-invasive
Bidens pilosa	Asteraceae	Yellow	Invasive
Anaphalis busua	Asteraceae	White	Non-invasive
Elsholtzia fruticose	Lamiaceae	purple brown	Non-invasive
Persicaria nepalensis	Polygonaceae	Purple	Non-invasive
Fagopyrum dibotrys	Polygonaceae	White	Non-invasive
Colquhounia coccinea	Lamiaceae	Red	Non-invasive
Lavatera cachemiriana	Malvaceae	Pink	Invasive
Cucurbita spp.	Fabaceae	Yellow	Non-invasive
Chirita bifolia	Gesneriaceae	Blue	Non-invasive
Jasminum humile	Oleaceae	Yellow	Non-invasive

Ophiopogon sp.	Oleaceae	White	Non-invasive
Prinsepia uitilis	Rosaceae	Yellow	Non-invasive
Viburnum erubescens	Caprifoliaceae	White	Non-invasive
Cercium verutum	Asteraceae	Purple	Non-invasive
Lantena camera	Verbenaceae	red-yellow	Invasive
Viburnum erubescens	Caprifoliaceae	White	Non-invasive
Lindera pulcherrila	Laureceae	Yellow	Non-invasive
Solanum tuberosum	Solanaceae	White	Invasive
Ageratina adenophora	Asteraceae	White	Invasive
Colquhounia coccinea	Lamiaceae	Red	Non-invasive
Viburnum erubescens	Caprifoliaceae	White	Non-invasive
Tagetes spp.	Asteraceae	Yellow	Non-invasive
Trifolium repens	Fabaceae	White	Non-invasive
Dahlia pinnata	Asteraceae	Red	Invasive
Anaphalis griffithii	Asteraceae	White	Non-invasive

Photo Plates

Photo 1 ***B. haemorrhoidalis*** **(queen)**

Photo 2 ***B. haemorrhoidalis*** **(worker)**

Photo 3 ***B. rotundiceps*** **(queen)**

Photo 4 ***B. rotundiceps*** **(male)**

Photo 5 ***B. turneri*** **(worker)**

Photo 6 ***B. festivus*** **(worker)**

Photo 7 ***B. genalis*** **(queen)**

Photo 8 ***B. asiaticus*** **(worker)**

Photo 9 ***B. breviceps*** **(worker)**

Photo 10 ***B. miniatus*** **(worker)**

Photo 11 ***B. grahami*** **(worker)**

Photo 12 ***B. parthenius*** **(worker)**

Photo 13 ***B. pressus*** **(worker)**

Photo 14 ***B. eximius*** **(worker)**

Photo 15 ***B. eximius*** **foraging on** ***Solanum tubersum***

Photo 16 ***B. eximius*** **foraging on** ***Jasminum humile***

Photo 17 ***B. turneri*** **foraging on** ***Jasminum humile***

Photo 18 ***B. haemorrhoidalis*****(Queen)**