

**DIVERSITY OF GROUND DWELLING ANTS (HYMENOPTERA:  
FORMICIDAE) IN JITPURPHEDI, TARKESHWOR OF KATHMANDU,  
NEPAL**



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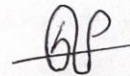
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**Submitted to:**  
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APRIL, 2022

## DECLARATION

I hereby declare that the work presented in this thesis entitled "**DIVERSITY OF GROUND DWELLING ANTS (HYMENOPTERA: FORMICIDAE) IN JITPURPHEDI, TARKESHWOR OF KATHMANDU, NEPAL**" 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 author(s) or institution(s).

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## LIST OF ABBREVIATIONS

Abbreviated form	Detailed of Abbreviations
asl	abovesealevel
B	Baittraps
cm	centimeter
GPS	Global Positioning System
H	Handcollection
km	Kilometer
P	Pitfall traps
Sp	Species
Spp	Morespeciesof same genus

## ABSTRACT

Ants (Hymenoptera: Formicidae) are the most diverse and successful insects on the Earth. Ant diversity is likely high, but there have been few studies on the ant diversity distribution in Nepal. This study evaluated the ants diversity habitats wise and seasonally through different methods in Jitpurphedi, Tarkeshwor. Samples were collected in three different habitats viz. forests, cultivated land and grassland in rainy (2019), autumn (2019) and winter (2019 and 2020) using pitfall traps, bait traps and opportunistic manual collection. Altogether 1748 ants were collected including five subfamilies, 19 genera and 27 morphospecies. Subfamily Formicinae was the most dominant and Dorylinae was the least. *Camponotus* was the most abundant genus as well as most specious genus in all seasons. Among the three different habitats, the similarity index (0.77) was found highest between cultivated land and grassland. Forests were the species rich habitat with Shannon diversity index 3.11 and evenness index 1.1. Similarly, highest number of ant species was collected in autumn season with Shannon diversity (2.96) and evenness index (0.9). The One-way ANOVA concluded that relationships between habitats and ant diversity as well as with seasons were statistically insignificant as the  $p < 0.05$ . Pitfall traps was the most effective technique for ant collection over bait traps and manual collection in all habitats and seasons.

# 1. INTRODUCTION

## 1.1 Background

Ants (Hymenoptera: Formicidae) are the most dominant insect group representing both ecologically and numerically 10% - 15% of the entire animal biomass in terrestrial ecosystems (Beattie and Hughes 2002). Ants are found everywhere, except in Iceland, Greenland and Antarctica (Holdobler and Wilson 1990). Ants occupy a large variety of food niches both on the ground and in the vegetation which makes their local diversity in most habitats exceed that of other social insects (Holdobler and Wilson 1990). Some ants are key predators, ecosystem engineers, seed dispersers and biocontrol agents (Wetterer 2017) while some are notorious pests of household, agriculture and forests. Ants are considered as a key component of terrestrial ecosystems directly or indirectly providing ecological services including seed dispersion control of herbivore population, acceleration of forest recovery or plant succession, organic matter decomposition and nutrient cycling in the soil through their nest building and maintenance (Del-Toro et al. 2012). These significantly affect the structure of different terrestrial ecosystems, mainly due to their abundance, different food habits, population establishment and efficiency of feeding (Holdobler and Wilson 1990). Most ants are beneficial to human beings as food (Defoliart 1999) while others produce compounds with pharmaceutical and biomedical applications (Reddy et al. 2011). Ants are responsible as model organisms for a wide range of scientific research including studies in behavior, ecology and evolutionary biology (Andersen et al. 2003). Ants are an important taxon for comparing habitat diversity and monitoring environmental changes because numerous species have habitat preference and respond quickly to disturbances to their environment (Andersen 1990; Alonso 2000).

Ants are an important and ubiquitous component of the insect fauna on all terrestrial habitats, except the poles with about 140 million years of evolutionary history (Moreau and Bell 2013). There are over 13,911 valid ant species belonging to 17 subfamilies and 338 genera in the world (Bolton 2021). Over 87% of the ant species described fall under four subfamilies viz. **Myrmicinae**, **Formicinae**, **Ponerinae** and **Dolichoderinae** (Guenard 2013). **Global Ant Biodiversity Informatics (GABI)** is the first comprehensive global database with ant species records in available publications and existing databases (Guenard et al. 2017). Ants are ideal candidates in monitoring ecosystem conditions because they occur everywhere and are numerically abundant in both intact and disturbed

habitats (Majer 1983), can be easily sampled (Agosti and Alonso 2000) and are sensitive as well as rapid responses to environmental variables (Majer 1983 and Alonso 2000). Environmental factors are key drivers for high taxonomic and functional diversity of ground dwelling ants in Continuous forests (Santos et al. 2019). Ants were considered to be very sensitive to habitat transformation and disturbances so, ants are extensively used as indicator species (Hoffmann and Andersen 2003). Indeed, ants have been used extensively for several decades in Australia as bioindicators of habitat change (Anderson 1990; Boulton et al. 2005). Many methods are used to sample ants including baiting, pitfall trapping, quadrant sampling, direct sampling, surface digging and litter techniques (Agosti et al. 2000). To decrease biases of sampling methods and to increase species richness, a complementary set of methods is recommended for ant surveys (Majer 1997). The relationship between biodiversity and ecosystem function is a topic that has generated intensive research and controversy in recent years (Hooper et al. 2005).

Nepal has been home to 133 ant species belonging to 48 genera and eight subfamilies (Subedi et al. 2020). Ninety two percentages ant species of Nepal are from four main subfamilies i.e. Myrmicinae (49%), Formicinae (28%), Ponerinae (10%) and Dolichoderinae (4.63%) by representing (0.9%) of the global ant diversity (Subedi, et al. 2020). Nepalese ants have received occasional attention from scientists after the first descriptions of two ant species from Nepal, *Aphaenogaster pachei* and *Myrmica pachei* (Forel 1906). The study of ants in Nepal is still in its infancy. There were no focused on systematic survey of Nepalese ants and publications. The high diversity of ants in the forests of Nepal needs to be assessed with further exploration using multiple sampling methods covering all seasons and forest types (Subedi et al. 2021).

## **1.2 Objectives**

### **1.2.1 General objective**

To explore the diversity of ground dwelling ants in Jitpurhedi, Tarkeshwor, Kathmandu, Nepal.

### **1.2.2 Specific Objectives**

1. To evaluate the ant variation in different habitats.
2. To compare ant diversity in rainy, autumn and winter seasons.
3. To assess the effectiveness of different collection methods.

## **1.3. Rationale of the study**

Ants are important components of ecosystem not only because they constitute a great part of the animal biomass but also because they act as ecosystem engineers. Ants are relatively easy to collect, diverse even in a small habitat and easily identifiable (Hölldobler and Wilson 1990; Agosti et al. 2000). Human activities in natural ecosystems result in fragmentation of ecosystems and biodiversity loss. Thus, it is important to protect ant diversity. However, a lack of taxonomic expertise and equipped infrastructure in the country pose a challenge in the study of ants. Majority of the current information about Nepalese ants was based on hand collected specimens (Subedi et al. 2021) so, surveys using multiple collection methods need to be conducted and many areas are still unexplored. Study was conducted to determine ant diversity, species richness and evenness index in different habitats by using different methods in three seasons. This research will create a preliminary base for the study of the ground dwelling ants in the future for the other researchers and provide useful information on environmental monitoring, conservation, evaluation and ecological research.

## 2. LITERATURE REVIEW

### 2.1. In the Context of Nepal

Nepal occupies the Central part of the Himalayas and offers unique ecological and species diversity. Considering the unique geographical and ecological diversity, many ant species are yet to be discovered and their ecology.

Forel (1906) first reported Nepalese ants; *Aphaenogaster pachei* and *Myrmica pachei*.

Collingwood (1970) published the first list of 34 species of ants of Nepal from the collections of the 1954 British Museum expedition to the Khumbu Himal Region. He collected 34 species of ants from different parts of Nepal from the altitudes ranging 850m to 4500m asl. Out of which, 12 were generally distributed over the whole Himalayan region, 12 were eastern Himalayan, eight were western Himalayan and two were endemic to Nepal.

Elmes and Radchenko (2009) recorded the two different species of *Myrmica*: *M. webri* and *M. alperti* from Makalu-Barun National Park, Nepal.

Adhikari et al. (2016) documented 30 genera and 70 morphospecies in three habitats and two seasons in Lahachowk, Kaski. Forests was the the most diverse habitats and spring season reported the higher species richness, species diversity and species evenness than autumn season. Sixteen genera using bait traps, pitfall traps and leaf litter method in Shivapuri Nagargun National Park, Nepal were recorded and reported new genera; *Pachycondyla* and *Echinopola* to Nepal (Neupane and Subedi 2018). Species richness was higher in winter than spring as well as pitfall method was found the most effective method.

Subedi et al. (2020) updated checklist of Nepalese ants that included 128 named species in 48 genera and 8 sub-families (*Myrmicinae*, *Formicinae*, *Ponerinae*, *Dolichoderinae*, *Dorylinae*, *Pseudomyrmicinae*, *Leptanillinae* and *Amblyoponinae*) with 33% of total species where *Myrmicinae* was the largest with 53% of total species followed by *Formicinae*.

Adhikari et al. (2020) recorded 12 genera using pitfall traps, using bait traps and opportunistic hand collection in altitudinally and seasonally in Phulchowk hill, Lalitpur, Nepal. The higher species richness, species diversity and abundance were highest in autumn season.

Pokhrel (2020) studied ant diversity along an elevational gradient in Champadevi, Hill, Central Nepal in two seasons by using pitfall traps and all-out search methods and

concluded species richness was maximum during spring season than autumn season and canopy cover do not show a significant effect on ant composition.

Subedi et al. (2021) recorded six sub-families and 36 genera from 70 ant species along the north and south belt transects in eastern, central and western Nepal by using vegetation beating, sweeping and hand collection methods in selected forest types. Forest ant species richness was high in western forests of Nepal.

## 2.2. In the Global context

Anu and Sabu (2006) analyzed leaf litter ants in the Wayanad region of Western Ghats and collected 22 species from 16 genera. Subfamily Formicinae was highly specious in evergreen forests. Hazra (2018) also reported Formicinae, the most dominant family in terms of species richness followed by Myrmicinae, Pseudomyrmicinae, Dolichoderinae and Ponerinae.

According to Mahalakshmi and Channaveerappa (2016), Manikandan et al. (2018) sampled Myrmicinae, the most dominant subfamily followed by Formicinae, Ponerinae and Pseudomyrmicinae. Likewise, the diversity of outdoor ants in a college campus of Kajrat city, Ahmednagar district, Maharashtra, India was studied (Khan 2018) where nine species were recorded belonging to subfamily Myrmicinae, Formicinae and Dolichoderinae.

Furthermore, Andrade and Del-claro (2007) investigated the variety of ant diversity on the ground of an ecological reserve between April 2005 and February 2006 and found 77 ant species distributed in 22 genera and 6 subfamilies where *Camponotus* and *Pheidole* were the most common genera. Similarly, Chavan and Pawar (2011) explored *Crematogaster*, *Pheidole* and *Camponotus* the most dominant species.

Species richness and composition of ant's assembling in adjacent mountain forest and secondary grassland habitats in the central plateau of Madagascar was evaluated (Fisher and Robertson 2002) where they collected 31 species in grassland and 26 species in forest. Patkar and Chavan (2014) compared ground-dwelling ants in undisturbed and disturbed habitat of Great Indian Bustard Wildlife Sanctuary in Maharashtra state, India and found more in undisturbed site. Manikandan et al. (2018) studied diversity of ants in three different sites of Thiruthangal, Sivakasi taluk, Virudhunagar, India and noted totally eleven species of ants belonging to four subfamilies where seven species found in

residential sites, five ant species were found in industrial sites and eleven species found in agricultural sites. Similarly, habitat disturbance, species richness, equitability and abundance of ants in the Fall-Line Sandhills, at Fort Benning, Georgia were examined (Graham et al. 2004). They collected 48 species of ants belonging to 23 genera over four years of sampling and noted that highly disturbed areas had fewer species and greater number of ants than did moderately or lightly disturbed areas.

Sunil et al (1997) reported the ant species richness generally increases with the increase in vegetation. Likewise, diversity and distribution of ground-dwelling ants in lowland rainforest in Southeast Cameroon was studied (Deblauwe and DeKoninck 2007) where they studied the effect of vegetation type on ant species density, activity and composition. Differences in species density, activity and composition between vegetation types were explained by developmental stage of vegetation type.

Furthermore, Forsy and Allen (2005) explored the relationship between sprawl and biodiversity using a data set of ant species collected from 46 habitat patches located in the increasingly suburbanized Florida Keys, USA and found that neither the overall number of native species nor the number of rare native species were significantly affected by the amount of development or proximity to roads and the number of non-native species was significantly correlated with the amount of development. Santoandre et al. (2019) found opposite environmental similarity gradient between natural habitats and plantation ages.

Levings (1983) examined patterns of species distributions in ground ants on Barro Colorado island (BCI), Republic of Panama where more species and more individuals were collected in wet than dry seasons using either methods. He found that moisture availability is an important contributor to these patterns of among site and among season variation as well as moisture availability may affect the distribution of suitable nest sites. Lindsey and Skinner (2001) observed ant composition and activity patterns in three different habitats in summer and winter in the semi-arid Karoo, South Africa and found ant abundance was greater during summer than winter. Similarly, Bharti et al. (2009) studied seasonal patterns of ants in five seasons in Punjab Shivalik Range of North West Himalayan. The number of species was least (5) during winter and maximum (40) during summer season. Likewise, Diversity of ant nests in Anjac Campus was studied by using pitfall traps and all search out methods and found that species richness was high in dry season than wet season (Anusuyadevi and Sevarkodiyone 2018).

In **W**estern Ghats, India, Basu (1997) recorded 29 species in spring and 13 species in late autumn. El Keroumi et al. (2012) recorded 13 species, seven genera in eight Argan forest, **L**ahssinate, **M**orocco. They recorded higher abundance and richness in the spring and summer seasons than autumn.

Yamane and Hashimoto (1999) estimated abundance and diversity of ants concluding that a combination of various sampling methods produce better results in the evaluation of ant species. Majer and Delabie (1994) compared leaf litter and soil fauna which had shown that a combination of pitfalls, litter shifting, baiting and hand sorting increase the efficiency of specimens captures in comparison to any single method by itself.

Furthermore, Gadagkar et al. (1993) studied ant species richness and diversity in some selected localities in western Ghats, India by sampling methods using traps and all-out-search method and found use of trapping method more successful (six subfamilies, 31 genera and 120 species) than all-out-search method (six subfamilies, 27 genera and 101 species). Similarly, ant diversity and abundance were studied along the elevation gradient in **P**hillipines using pitfall and hand collection methods where pitfall method recorded 51 species of ants and hand collection yielded 48 species (Samson et al. 2006). Barech et al (2016) sampled ants in the saline Dry Lake Chott El Hodna in **A**lgeria, a **R**amsaar **C**onservation **W**etland where a total of 928 ant individuals were collected (682 by pitfalls, 246 by hand).

**H**owever, Fischer (2004) surveyed diversity patterns of ants on **M**onts Doudou in south eastern Gabon using the following methods: litter sifting, sweeping, beating yellow pan traps, pitfall, **M**alaise traps and hand collecting method where Leaf litter methods captured the greatest number of species followed by sweeping, beating and general collecting. Fisher and Robertson (2002) and **M**artelli et al. (2004) concluded that litter sifting was the single most efficient method.

### 3. MATERIALS AND METHODS

#### 3.1. Study Area

The study was carried out in Jitpurphedi (27°46'55" N, 85°17'14"E) of Tarkeshwor municipality of Kathmandu, Nepal. Jitpurphedi is located on northern part of Kathmandu valley below Tarkeshwor temple and has subtropical climate. The distance from Jitpurphedi to Kathmandu is 9.5km. The study was conducted in three habitats viz forest, cultivated land and grassland (approximately 100x70m per site ) and lies with the elevation range of 1400m-3000m in rainy (June-July) 2019, autumn (September-October) 2019 and winter season (January-February) 2020. The average temperature of rainy season was 24°C, autumn was 24°C and winter was 15°C. Different types of vegetation and various human activities are seen in different habitats.

**Forests:** Sub-tropical forests are found in this habitat. Two types of forests of (approximately 100 70m per site ) were selected i.e. first mixed forests where the dominant floral species are *Azeretina Adenophora*, *Reinwartina indica*, *Schima wallichii*, *Castonopsis indica* etc and another pine forests dominated by pine trees. The average pH value of soil is 7. Less human disturbances and protected against grazing are seen in this site.

**Cultivated land:** Kitchen garden land and another farming field were selected for the research in cultivated land . *Brassica species*, *Triticum aestivum*, *Eleusine corocana*, *Oryza sativa*, etc were the crops according to the seasons. The average pH value of soil is

6.5. Human activities like tillaging, harvesting etc are available in this habitat. **Grassland:** Two plots of grassland are made. *Glycine max*, *Cynodon dactylon*, *Imperata cylindrical*, *Oplimemus* etc of grass species are found. The average pH value of soil is 7. Grazing of cattles like goats, cows etc are present in this habitat.

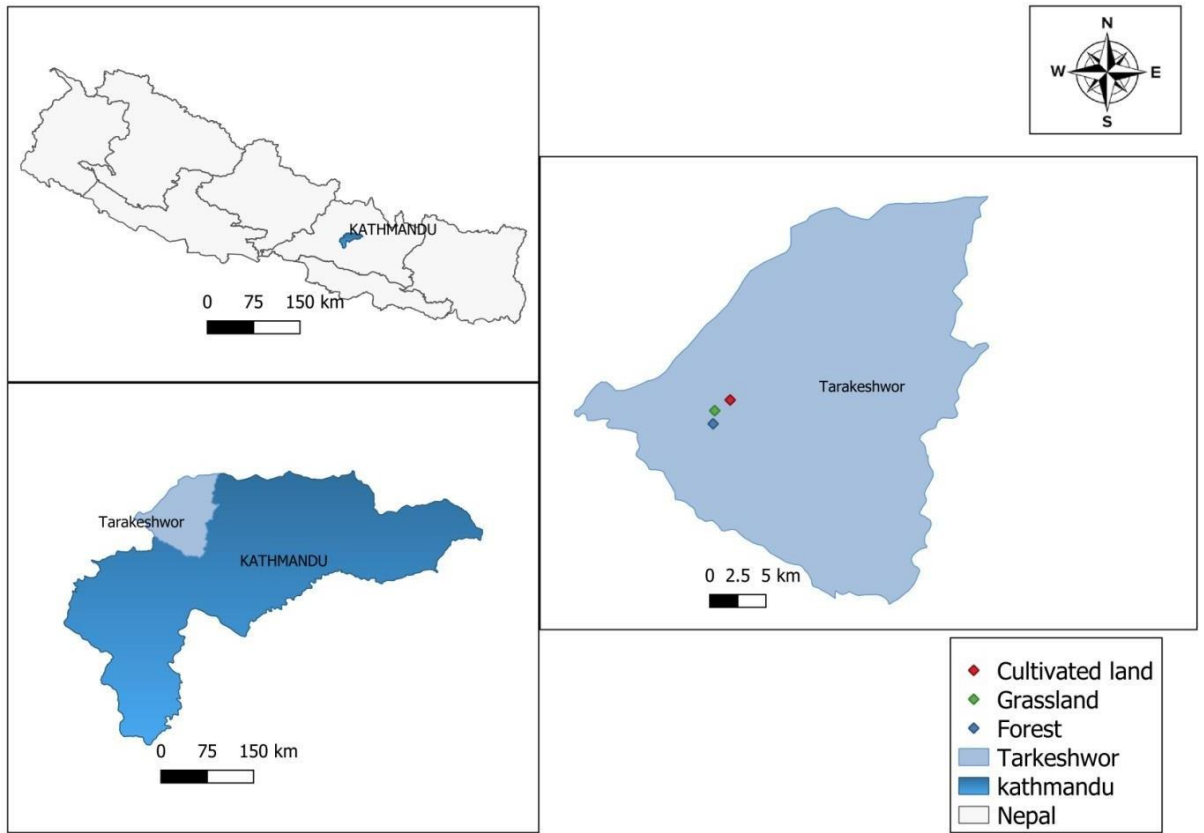


Figure 1: Map of study area

### **3.2. Materials Required:**

1. *Equipments:* GPS, PH soil meter, measuring meter, plastic cups, feather-weight forceps, note book, digger, pen, scale, vials, entomological pins, identification keys and stereo-microscope
2. *Chemicals:* Soapy or detergent water, sugar, breads, biscuits, 70% ethyl alcohol

### **3.3. Methods**

#### **3.3.1. Sampling methods**

Samples were collected three times in every season: rainy (June- July) 2019, autumn (September-October) 2019 and winter season (January-February) 2020. Ants were collected at three sites: forests, cultivated land and grass land using pitfall traps, bait traps and hand collection method (Agosti and Alonso 2000). For removal of sampling errors, three different ant collection methods to collect maximum number of ant species from study area. The trapped ants of all this methods were preserved in vials containing 70% ethanol.

##### *1. Pitfall traps*

The pitfall traps consisted of 0.5 liter soapy or detergent water with an opening of 12 cm in diameter, buried at ground level. Each trap was placed in 10m distance apart and kept for 48 hours. The trapped ants were collected in vials with forceps and preserved in 70% ethanol.

##### *2. Bait traps*

Bait traps consists of opening (lids) of bottle one cm length and eight cm in diameter. Twenty bait traps were placed in 100m area with the distance of 10m apart. Bread and sugar solution were used in the form of bait and kept for 30 minutes and were placed near the pitfall traps. The ants roaming around the baits were collected in the vials with the help of feather with forceps and preserved in 70% ethanol.

##### *3. Opportunistic manual collection*

In this method, the ants were collected from the ground by the hand or the forceps at the time of walking after laying the baits under the stones, logs, barks etc in sampling period and preserved in 70% ethanol for 30 minutes.

#### **3.3.2. Identification of ants**

The collected specimens were identified up to morphospecies with identification keys (Bolton 1994; Holdobler and Wilson 1990) using Stereo-zoom trinocular microscope tallying with specimens and photographs (AntWeb 2021 and AntWiki 2021)). The

photographs were taken by digital Oppo mobile placing ants in slide with scale at Stereo- zoom trinocular microscope. Collected specimens were deposited to the Zoology Department of Amrit Campus.

### **3.4. Data processing and statistical analysis**

The collected ant specimens' data were entered in MS-Excel sheet and the complete count of species presented in each habitat, method and season were done for specimens' composition and structure indices. The diversity index of each sampling plot was first calculated with the presence data of species richness and the frequency of each species by using

Shannon-Weiner Diversity index ( $H'$ ) =  $-\sum (P_i) * (\ln P_i)$

This was used to calculate the species diversity indices in a community. Pielou's Evenness

Index  $J' = H' / H_{max}$

This evenness index was used to know the closeness of species of ant in type of habitats, method and seasons and to find the stability of an ecosystem.

To measure the similarity between two community samples, coefficient of Sorensen was used as the following equation.

$QC = 2a / (2a + b + c)$

One-way ANOVA was used to calculate the relation between the habitats heterogeneity and species richness as well as to find out the association between the seasons and species richness.

#### 4. RESULTS

Total of 1,748 ants were reported including five subfamilies (Formicinae, Myrmicinae, Ponerinae, Pseudomyrmicinae and Dorylinae), 19 genera (*Camponotus sp*, *Polyrachis sp*, *Lepisiota sp*, *Oecophylla sp*, *Paratrechina sp*, *Nylanderia sp*, *Aphaenogaster sp*, *Crematogaster sp*, *Pheidole sp*, *Meranoplus sp*, *Monomorium sp*, *Lophomyrmex sp*, *Tetramorium sp*, *Leptogeny sp*, *Brachyponera sp*, *Odontoponera sp*, *Odontomachus sp*, *Dorylus sp*, and *Tetraponera sp*) and 27 morphospecies through three different techniques (Table 1). Out of five subfamilies, Formicinae represented the most abundant subfamily where Dorylinae have the least abundance (figure 2)

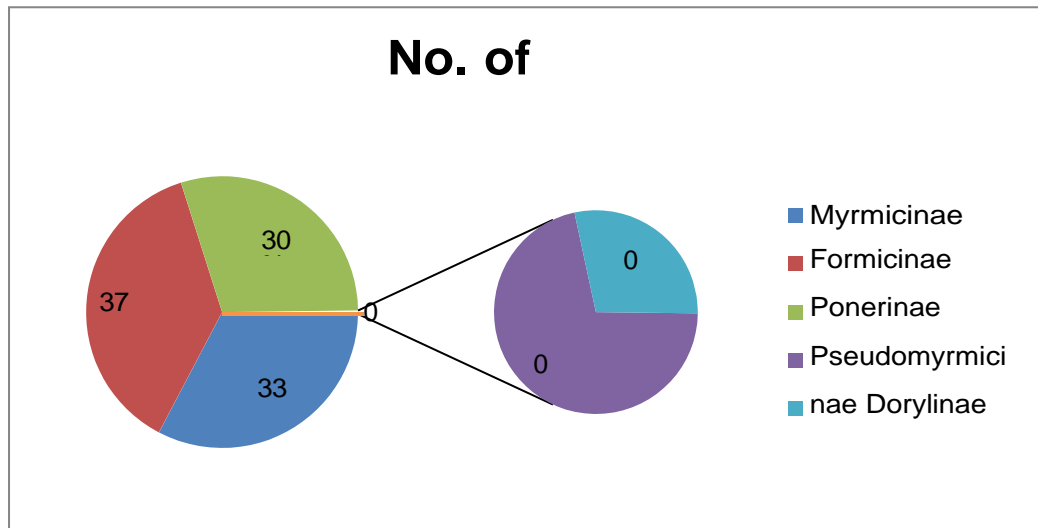


Figure 2: Subfamily wise ant abundance

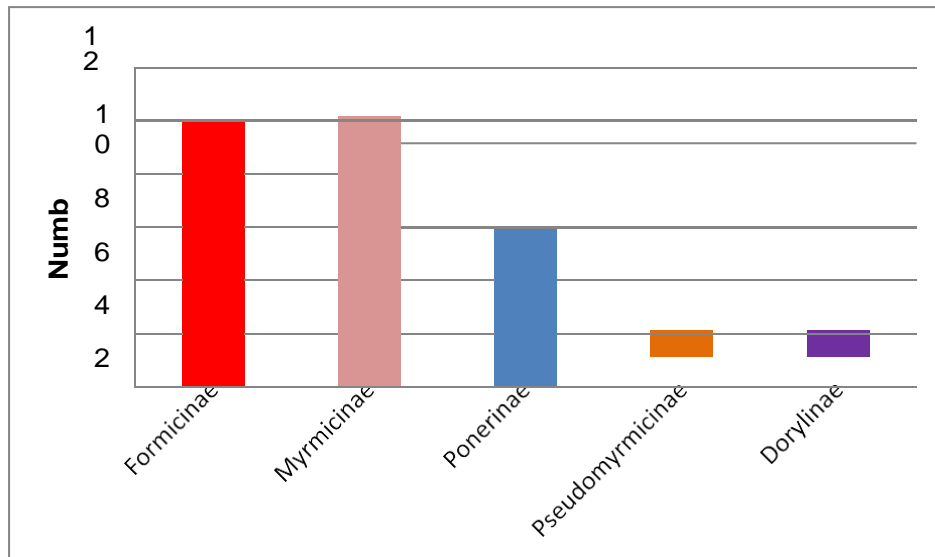


Figure 3: Subfamily wise ant morphospecies

#### 4.1. Species richness

Five subfamilies (Myrmicinae, Formicinae, Ponerinae, Pseudomyrmicinae and Dorylinae), 19 genera (*Camponotus sp*, *Polyrachis sp*, *Lepisiota sp*, *Oecophylla sp*, *Paratrechina sp*, *Prenolepis sp*, *Aphaenogaster sp*, *Crematogaster sp*, *Pheidole sp*, *Meranoplus sp*, *Monomorium sp*, *Lophomyrmex sp*, *Tetramorium sp*, *Leptogeny sp*, *Brachyponera sp*, *Odontoponera sp*, *Odontomachus sp*, *Dorylus sp*, and *Tetraoponera sp*) and 27 morphospecies were reported (Table 1). Formicinae bear six genera with ten morphospecies followed with Myrmicinae including seven genera and nine morphospecies, Ponerinae with four genera and six morphospecies, Pseudomyrmicinae and Dorylinae with single genera and single morphospecies (Table 1).

Table 1: Ants reported from Jitpurphedi, Tarkeshwor

S.N.	Subfamilies	Genus	Morphospecies
1	Myrmicinae	<i>Aphaenogaster</i> Mayer, 1853	1
		<i>Crematogaster</i> Lund, 1831	2
		<i>Pheidole</i> Westwood, 1839	2
		<i>Monomorium</i> Mayer, 1855	1
		<i>Lophomyrmex</i> , Emery, 1897	1
		<i>Tetramorium</i> Mayer, 1855	1
		<i>Meranoplus</i> Smith, 1853	1
2	Formicinae	<i>Camponotus</i> Mayer 1861	4
		<i>Polyrachis</i> Smith, 1857	1
		<i>Paratrechina</i> Motschulsky, 1863	2
		<i>Prenolepis</i> Mayer, 1861	1
		<i>Oecophylla</i> Smith, 1860	1
		<i>Lepisiota</i> Santschi, 1926	1
3	Ponerinae	<i>Leptogeny</i> Roger, 1861	2
		<i>Brachyponera</i> Emery, 1900	2
		<i>Odontoponera</i> Mayer, 1862	1
		<i>Odontomachus</i> Latreille, 1804	1
4	Pseudomyrmicinae	<i>Tetraponera</i> Smith, 1852	1
5	Dorylinae	<i>Dorylus</i> Fabricius, 1793	1
Total	5	19	27

#### 4.2. Diversity of ants in different habitats

The study was conducted out in three habitats: forest, cultivated land and grassland.

In forests, *Camponotus* sp.1, *Camponotus* sp.2, *Odontoponera* sp, *Leptogeny* sp1, *Aphaenogaster* sp and *Crematogaster* sp.1, were among the most abundant species. *Camponotus* sp3, *Oecophylla* sp, *Crematogaster* sp2, *Meranoplus* sp, *Leptogeny* sp, *Odontoponera* sp, *Odontomachus* sp, *Tetraponera* sp was also reported (Appendix II).

In cultivated land, *Polyrachis sp* was the most abundant species whereas *Dorylus sp* was the species reported in this habitat only (Appendix II).

In grassland, *Odontoponera sp*, *Leptogenysp1*, *Brachyponera sp1* and *Brachyponera sp2* were collected maximum in this habitat (Appendix II).

The highest similarity index was between cultivated land and grassland (0.77) while the lowest similarity index was between the forests and cultivated land (0.68) (Figure 6).

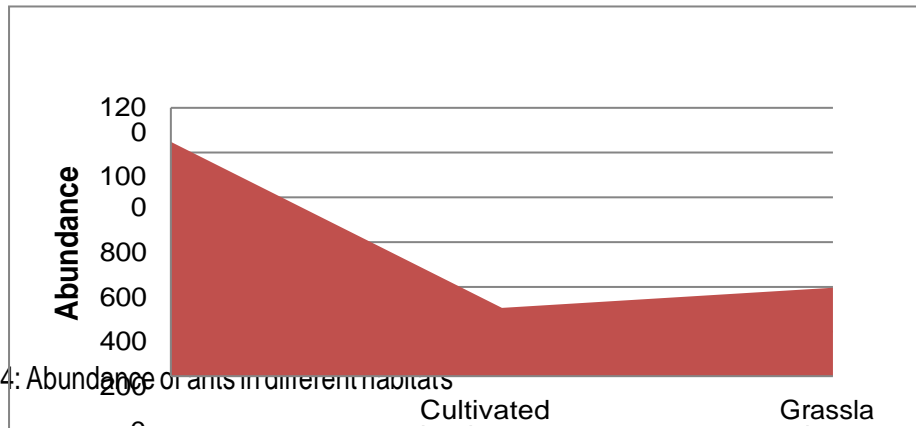


Figure 4: Abundance of ants in different habitats

Table 2: Data analysis of ant diversity in different habitats

Diversity index/sites	Forests	Cultivated land	Grassland
Species richness (S)	17	13	12
Shannon Diversity (H)	3.11	2.17	1.46
Evenness Index (J)	1.09	0.84	0.58
Number of ants collected	1048	305	395

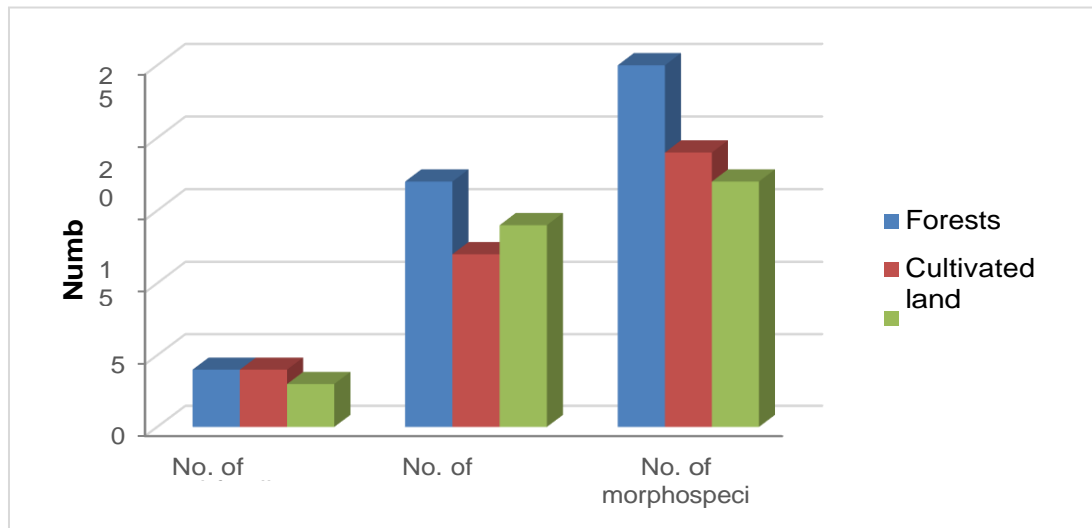


Figure 5: Subfamilies, genera and morphospecies in different habitats

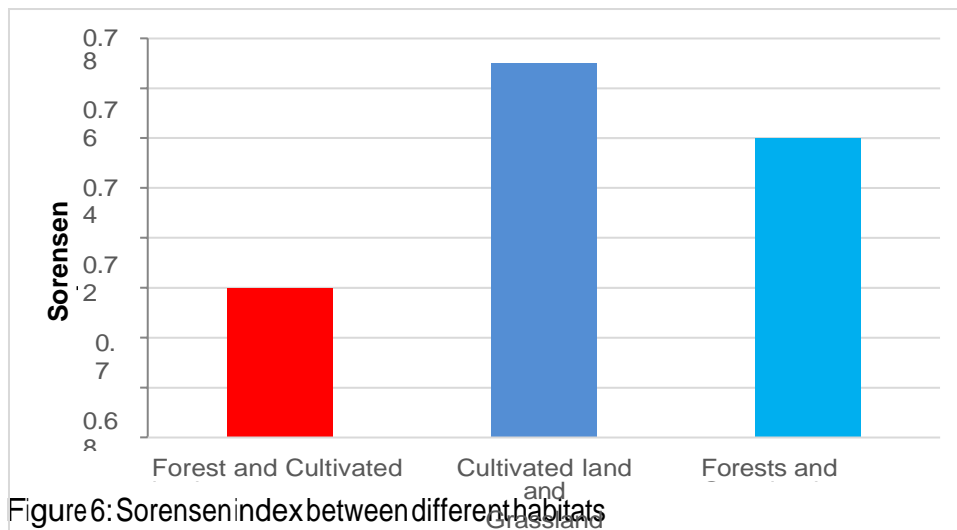


Figure 6: Sorensen index between different habitats

In one-way ANOVA, it is proved that significant differences between ant species composition in various habitats at  $p < 0.05$  level of significance ( $p$ -value=0.002), F-value (6.71)

### 4.3. Seasonal variation in diversity of ants

This research was conducted during rainy, autumn and winter. Four subfamilies, Fifteen genera, 23 morphospecies were collected in rainy season, five subfamilies, 19 genera and 27 morphospecies were collected in autumn season and three subfamilies, 12 genera, 12 morphospecies were collected in winter seasons (Appendix III). Maximum species richness (27) was recorded in autumn as compared to rainy (15) and winter (12).

Similarly, the Shannon index of species richness ( $H'$ ) was recorded as the highest during autumn season as  $H'=2.96$  than in rainy  $H'=2.88$  and winter  $H'=2.38$  as a greater number of individuals was collected in autumn (874), rainy (575) and winter (299) (Appendx 4). Higher species richness (18) in forests was recorded in autumn as compared to rainy (15) in forests and winter (10) in forests (Table 4)

The ANOVA comparisons of ant species richness across the three season's detected significant differences between the seasons at  $p<0.05$  level of significance ( $p$ -value=0.003), F-value (6.33).

Table 3: Ants diversity reported in different seasons with habitats

Seasons	Habitats	Species richness(S)	Shannon Index (H)	Evenness index (J)	No. of ants collected
Rainy	Forests	15	1.957	0.722	345
	Cultivated land	11	0.627	0.261	91
	Grassland	11	0.922	0.384	139
Autumn	Forests	18	2.036	0.704	526
	Cultivated land	13	0.735	0.286	165
	Grassland	13	0.858	0.334	183
Winter	Forests	10	1.589	0.690	177
	Cultivated land	5	0.604	0.375	50
	Grassland	9	0.851	0.387	72

#### 4.4. Sampling efficiency of different methods

Pitfall method collected maximum ant individuals (1149) followed by bait method (526) and opportunistic hand collection (73) (Figure 7). Majority of ants collected by pitfall method included *Camponotus sp1*, *Camponotus sp2*, *Camponotus sp3*, *Camponotus sp4*, *Aphaenogaster sp*, *Polyrachis sp*, *Leptogeny sp1*, *Brachponera sp1*, *Pheidole sp1* and *Odontoponera sp*. *Camponotus sp2*, *Camponotus sp4*, *Meranoplus sp*, *Braachyponera sp*, *Odontomachus* and *Dorylus sp* were collected only through this method. *Paratrechina sp* and *Tetramorium sp* were collected through bait traps and *Oecophylla sp*, and *Tetraponera sp* were collected through hand collection only.

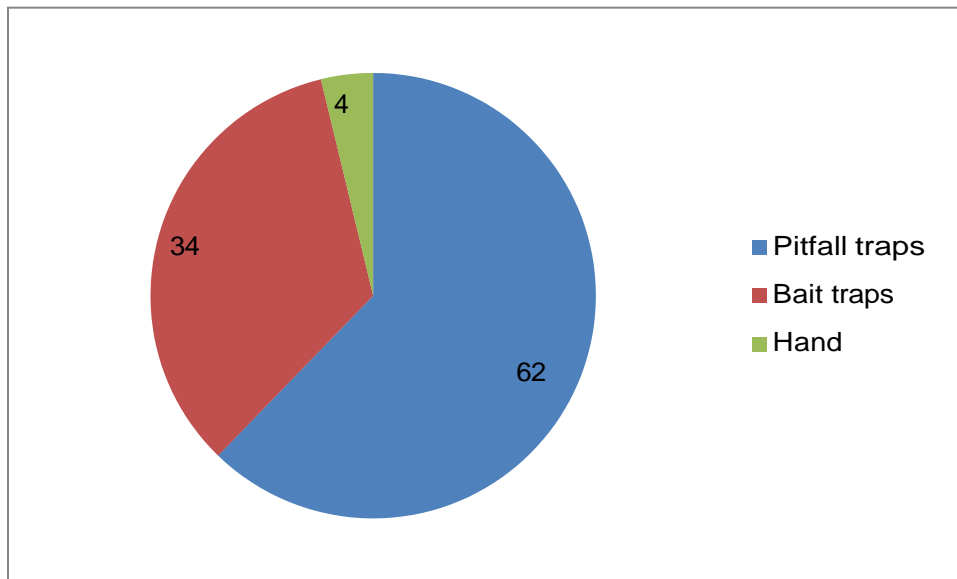


Figure 7: Abundance of ants collected in different sampling methods

Table 4 : Ants collected through different methods

Season		Rainy			Autumn			Winter		
Genus	Site	Forest s	Cultiva ted land	Gras slan d	Forest s	Cultivat edland	Grassl and	Fores ts	Culti vated land	Grassla nd
<i>Camponotous sp1</i>		P,H	P	P	P,H	P	P,H	P,H	P	P,H
<i>Camponotous sp2</i>		P	-	P	P,H	P	P	-	-	-
<i>Camponotous sp3</i>		P	-	-	P	-	-	P	-	-
<i>Camponotous sp4</i>		P	-	-	P	-	-	-	-	-
<i>Polyrachis sp1</i>		P	P,H	P,H	-	P,H	P,H	-	P,H	-
<i>Paratrechina sp1</i>		B	-	-	P,B, H	P,H	B	-	-	B
<i>Lepisiota sp1</i>		-	-	P,B	B	B	-	-	-	-
<i>Prenolepis sp1</i>		B	B,P	-	B	B	-	-	-	-
<i>Oecophylla sp1</i>		-	-	-	H	-	-	-	-	-
<i>Aphaenogaster sp1</i>		P,B	-	P,B	H	B	-	-	P,B	-
<i>Crematogaster sp1</i>		B	B	P,B	P,B, H	B	P,B	H	-	-
<i>Cematogaster sp2</i>		P	-	-	P	-	-	P	-	-
<i>Tetramorium sp1</i>		B	B	-	B	B	-	-	-	-
<i>Pheidole sp1</i>		P,B	B	P	P,B	B	-	-	-	-
<i>Pheidole sp2</i>		B	P,B	-	P,B	B	-	-	B	-

<i>Monomorium sp1</i>	P,B	B	-	P,B	P,B	B	-	-	-
<i>Meranoplus sp1</i>	-	-	-	-	H	-	-	-	-
<i>Lophomyrmex sp1</i>	P,B	B	-	P,B	B	-	-	-	-
<i>Leptogeny sp1</i>	P,H	-	P,H	P,H	-	P,H	P	-	P,H
<i>Leptogeny sp2</i>	P,H	-	P	P,B, H	-	P	P	-	P
<i>Brachyponera sp1</i>	P,B, H	-	P,B ,H	P,B, H	P	P,B, H	P,B	-	P
<i>Brachyponera sp2</i>	-	-	P,B	P	P	P,B	-	-	P,B
<i>Odontomachus sp1</i>	P	-	-	P	-	-	P	-	-
<i>Odontoponera sp1</i>	P,B, H	-	P	P,B, H	-	P	P,B	-	-
<i>Tetraponera sp1</i>	H	-	-	H	-	-	-	-	-
<i>Dorylus sp1</i>	-	-	-	-	P	-	-	-	-

## 5. DISCUSSIONS

### 5.1. Species richness

The present study recorded ant specimens representing five subfamilies (**Myrmicinae**, **Formicinae**, **Ponerinae**, **Pseudomyrmicinae** and **Dorylinae**), 19 genera and 27 morphospecies in Jitpurphedi, Tarkeshwor (Figure 5). Out of the five subfamilies, **Formicinae** was the most dominant family followed by **Myrmicinae** and **Ponerinae** (Figure 2). This result was agreed with the study of ant species diversity at **Contai Municipality, Purbi, Medinipur West Bengal, India** (Hazra 2018; Neupane and Subedi 2018; Adhikari et al. 2020). This outcome was also somehow coincided with the diversity and distribution of forest ants in **Nepal** (Subedi et al. 2021) while **Mahalakshmi and Channaveerappa (2016), Manikandan et al. (2018)** sampled **Myrmicinae**, the most dominant subfamily followed by **Formicinae, Ponerinae** and **Pseudomyrmicinae** whereas the diversity of outdoor ants in a college campus of **Kajrat city, Ahmednagar district, Maharashtra, India** was studied (Khan 2018) where nine species were recorded belonging to subfamily **Myrmicinae, Formicinae** and **Dolichoderinae**.

Out of 19 genera most specious genus was *Camponotus* (four morphospecies) followed by *Crematogaster sp1*, *Pheidole sp1*, *Leptogeny sp1* and *Brachyponera sp1* with two morphospecies each. This result also agreed with study recorded *Camponotus*, *Pheidole* and *Crematogaster*, the most prevalent genera globally (Wilson 1976; Ryder Wilkey et al. 2010; Bolton 2020). In this study area, *Camponotus sp1*, *Odontoponera sp*, *Leptogeny sp*, *Brachyponera sp1*, *Aphaenogaster sp*, *Crematogaster sp1*, *Paratrechina sp*, *Polyrachis sp* species were the most abundant genera. This result was co-existence with the study of ants in **Lahachowk, Kaski, Nepal** (Adhikari et al 2020 ).The most frequently encountered ant was *Camponotus* and had the greatest individual numbers which are called carpenter ants because of their nest ing behavior (Chavan 2011). This result also agreed with the literature in which the ants of the *Camponotus* genus were considered ubiquitous subordinate ants that may numerically dominant arboreal vegetation (Davidson 1977; Tadu et al. 2014). **Myrmicinae** occurs in different habitats due to their adaption potential in varying environmental conditions eg: *Pheidole* nests in soil while *Crematogaster* nests on dead wood of trees (Andersen et al. 2002). **Ponerinae** subfamily is more specific about niche and food habitats. They nest in soil and even rotten logs and feeds on wide range of food sources. **Pseudomyrmicinae** in this research was represented by single genus *Tetraponera sp* and was exactly similar with the study of ground dwelling

ant in Phuichowki hill, Lalitpur, Nepal (Adhikari et al. 2020). These are solitary foragers and make them in dead woods and rotten logs (Chavan 2014). *Paratrechina Longicornis*, known as the crazy ant prefers moist habitats for reproduction and the numerical dominance of *Paratrechina* on baits is thought to be principally linked to its foraging speed (Kenne et al. 2005).

### 5.1. Ant diversity comparison among habitats

The forest was slightly richer in ant species (25 morphospecies) than the cultivated land (19 morphospecies) and the grassland (17 morphospecies) (Figure 5). Ant species richness in forest increased with increasing heterogeneity of vegetation and density of trees (Bestelmayer et al. 2001). Calceira et al. (2010) recorded higher species 39 in forest and 29 in grassland of Argentina which was parallel with Jitpurphedi outcome. Similarly, in Amravati City of India, Chavan and Pawar (2011) recorded 30 species of ants in forest, 22 species of ants in human settlement and 15 species in grassland which was agreed with this finding. Lower species richness and evenness of ants in different habitats is due to disturbance (Bruhl et al. 2003). The composition of ant communities may be influenced by variation in resource availability and habitat quality (Palmer 2003; Boulton et al. 2005; Dauber et al. 2005), interspecific competition (Gibb 2005) and temporal changes in activity (Bestelmayer 2000; Albrecht and Gotelli 2001). The presence and relative abundance of ants in some systems is affected by both habitat patch size and edge effects (Bruhl et al. 2003).

The highest Sorensen similarity coefficient was found between grassland and cultivated land (0.77) followed by forests and grassland (0.74). The least Sorensen similarity coefficient was between forests and cultivated land (0.68) (Figure 6). This result was exactly similar with the study of ants in Lahachowk, Kaski, Nepal (Adhikari et al 2020) while Patkar and Chavan (2011) calculated the highest similarity index between forest and human habitat.

In this study areas, human disturbances in cultivated land mostly occurred during tilling period and harvesting period. Similarly, the lowest species richness of grassland could be the consequence of fragmentation, cattle grazing and disturbance and smaller area structures. Likewise, Patkar and Chavan (2014) compared ground-dwelling ants in undisturbed and disturbed habitat of Great Indian Bustard Wildlife Sanctuary in Maharashtra state, India and found more in undisturbed site. Moreover, species richness was negatively affected with grazing and significantly correlated with the plant biomass

or plant richness (Boulton et al. 2005). Graham et al. (2004) examined habitat disturbances, species richness, equitability and abundance of ants in the Fall-Line Sandhills, at Fort Benning, Georgia and noted that highly disturbed areas had fewer species and greater number of ants than did moderately or lightly disturbed areas. Sunil et al (1997) reported the ant species richness generally increases with the increase in vegetation. These are the reasons why ant species richness less in grassland and cultivated land than forests and these all evidences supports Jitpurphedi results.

## 5.2. Seasonal variation

The study was carried in rainy, autumn and winter seasons. Maximum species richness (27) was recorded in early autumn as compared to rainy (15) and winter (12). Maximum species richness (27 morphospecies) was recorded in autumn as compared to rainy (23 morphospecies) and winter (19 morphospecies). This result was exactly co-existence with the study of ground dwelling ant in Phuichowki hill, Lalitpur, Nepal (Adhikari et al., 2020). The study has shown strong seasonal fluctuations within the ant community due to the climatic and abiotic factor. Ants were found to be less active during the coldest and driest time of the year (Rico-Gray et al. 1998). They alter and gradually halt their activities and process to hibernation due to cold as temperature and moisture availability decreases from late autumn. Species richness was high in dry season than wet season (Anusuyadevi and Sevarkodiyone 2018). Overall seasons, fewer species and fewer individuals were collected at drier, sunnier sites and found that moisture availability is an important contributes to these patterns of among site and among season variation as well as moisture availability may affect the distribution of suitable nest sites (Levings 1983). The number of ants specimen in rainy (May-July) has decreased as compared to (August-September) might be the factor of weather. Rain is known to limit foraging activity. Holdobler and Wilson (1990) stated that rainfall may be able to reduce the ants activity because of rainfall washing away pheromones and lose their way to back home. The temperature was approximately 24°C in autumn season which was most favorable temperature for ants to forage so, the species richness was higher in autumn than winter and rainy season. In seasonal pattern of ants in Punjab Shivalik range in North-West Himalaya which concluded that eight species were recorded in winter and 40 species in summer season (Bharti et al 2009). Holdobler and Wilson (1990) noted that ants nest preferentially under flat rocks in cooler climates because these have low specific heat when dry and thus heat up faster than the surrounding soil. Similarly, Dreyer (1932)

observed that temperature gradually decreased in winter season so that ants face problem in gaseous exchange and low respiratory quotient than reduction in metabolic activities and finally, they go to hibernation due to cold temperature. So, species richness varied in different seasons, temperature and moisture availability (Adhikari 2016), precipitation (Barry 2008, Dunn et al. 2009), and contemporary climate between habitats (Hawkins et al. 2003). Thus, species richness is low in winter in comparison to autumn season and rainy season.

### **5.3. Effectiveness of ant collection methods**

The study has shown considerable variations of the ant community due to different methods. The highest number of ants (66%) was collected by using pitfall method than bait traps (30%) and hand collection (4%) (Figure 7). The highest number of ants was collected in pitfall traps because they were kept for longer duration and sample both nocturnal and diurnal ants. This result was similar to the examination of ants in the Fall - Line Sand hills at Fort Benning, Georgia (Graham et al. 2004). Pitfall method was the most successful method of ant collection (22 morphospecies) followed by bait method (14 morphospecies) and then hand collection method (13 morphospecies). This findings was parallel with the outcome of the previous study in Nepal (Neupane and Subedi 2018). This study was also corresponding to the survey of ants performed on 12 localities in Uttara Kanada districts of Karnataka which yielded 31 genera with 120 species and concluded that pitfall traps as the successful method with 28 genus and 91 species collected (Gadagkar et al. 1993). Similarly, ant diversity and abundance were studied along the elevation gradient in Phillipines using pitfall and hand collection methods where pitfall method recorded 51 species of ants and hand collection yielded 48 species (Samson et al. 2006). Barech et al (2016) sampled ants in the saline Dry Lake Chott El Hodna in Alergia, a Ramsaar Conservation Wetland. A total of 928 ant individuals were collected (682 by pitfalls, 246 by hand) representing 24 species belonging to 14 genera and 4 sub families. More species of ants were collected by pitfall trapping (20) than hand sampling (15). So, pitfall traps was the easiest and most cost-effective method but may not be effective for species associated with soil, deep litter and vegetation (Majer 1997). Bait traps are effective for collecting live ants. Hand collecting gives good results for comparative studies of ant assemblages across habitats than litter sampling or pitfall traps because it can be easily sampled ant fauna in different environmentally sensitive sites such as rocks, slopes, human and animal disturbances (Gotelli et al. 2010).

## 6. CONCLUSION AND RECOMMENDATIONS

### 6.1. Conclusions

Altogether, five subfamilies, 19 genera and 27 morphospecies were recorded from three types of habitats using three methods in three seasons. This study concluded that sub-family Formicinae, Myrmicinae and Ponerinae were the dominant subfamilies. This study showed that the most preferred habitat was forest area followed by grassland and cultivated land. Similarly, Shannon Diversity index and species evenness were highest in forests, cultivated land and grassland respectively as well as there is significant differences between ant richness with habitat heterogeneity and different seasons. More than 44.44% of ant species were overlapped in all habitats, eight species were found in only one habitat and remaining ants were found in two different habitats. In case of seasons, autumn season was the most diverse in comparison to rainy and winter season with highest Shannon diversity index and evenness index. One-way ANOVA concluded that relationships between habitats and ant diversity as well as with seasons were statistically insignificant as the  $p < 0.05$ . Similarly, pitfall method represented the maximum species richness and abundance over bait traps and hand collection. Hence, forests and autumn season were the best habitat and seasons for ant fauna respectively as well as pitfall traps was the most effective, efficient and easiest method for sampling ants in all habitats and seasons.

### 6.2. Recommendations

Future surveys should be directed toward testing the efficiency of the ant survey methods in various habitats.

The research was carried out for three seasons only so, in depth research should be designed to cover more seasons within a year and in between year.

Only three methods were used for data collection in this research, for more exploration of ants other effective methods like Winkler extractor, Berlese extraction, leaf litter shifting and canopy collection (canopy baits, canopy pitfalls) could be used for more exploration of ants.

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## 8. APPENDICES

### APPENDIX I: Photoplates of representative genus recorded in Jitpurphedi



*Tetraponera* (Pseudomyrmicinae)



*Leptogeny* (Ponerinae)



*Odontomachus* (Ponerinae)



*Odontoponera* (Ponerinae)



*Brachyponera* (Ponerinae)



*Monomorium* (Myrmicinae)



*Lophomyrmex* (Myrmicinae)



*Pheidole* (Myrmicinae)



*Crematogaster* (Myrmicinae)



*Camponotus* (Formicinae)



*Oecophylla* (Formicinae)



*Polyrachis* (Formicinae)



*Paratrechina* (Formicinae)



*Dorylus* (Dorylinae)

**AppendixII: Ants reported in different habitats**

S.N	Species	Forests	Shannon index	Cultivated land	Shannon index	Grassland	Shannon index
1	<i>Camponotus sp1</i>	102	0.226742	78	0.348724	45	0.247468
2	<i>Camponotus sp2</i>	28	0.0967825	-	-	8	0.078976
3	<i>Camponotus sp3</i>	18	0.096782	-	-	-	-
4	<i>Camponotus sp4</i>	12	0.05118	-	-	-	-
5	<i>Polyrachis sp</i>	8	0.037215	63	0.325777	24	0.17017
6	<i>Paratrechina sp1</i>	62	0.167276	28	0.219236	18	0.14074
7	<i>Paratrechina sp2</i>	26	0.09171	36	0.252211	23	0.165564
8	<i>Lepisiota sp</i>	1	0.00663	2	0.032965	3	0.037065
9	<i>Nylanderia sp</i>	32	0.106531	6	0.077283	28	- 0.187613
10	<i>Oecophylla sp</i>	3	0.016763	-	-	-	-
11	<i>Aphaenogaster</i>	135	0.263992	36	0.252212	25	0.174684

12	<i>Crematogaster sp1</i>	68	0.17747	4	0.056839	3	0.037065
13	<i>Crematogaster sp2</i>	26	0.091708	-	-	-	-
14	<i>Pheidole sp1</i>	52	0.149023	8	0.095498	18	0.140742
15	<i>Pheidole sp2</i>	43	0.131028	9	0.10396	-	-
16	<i>Monomorium sp</i>	29	0.09926	6	0.077283	16	0.129875
17	<i>Meranoplus sp</i>	2	0.011949	-	-	-	-
18	<i>Lophomyrmex sp</i>	28	0.096782	16	0.154634	12	0.106146
19	<i>Tetramorium sp</i>	23	0.083817	9	0.10396	-	-
20	<i>Leptogeny sp1</i>	52	0.149023	2	0.032965	55	0.274519
21	<i>Leptogeny sp2</i>	21	0.07835	-	-	8	0.078976
22	<i>Brachyponera sp1</i>	49	0.143204	-	-	78	0.320328
23	<i>Brachponera sp2</i>	9	0.040855	-	-	31	0.199726
24	<i>Odontoponera sp</i>	193	0.311589	-	-	-	-

25	<i>Odotomachus sp</i>	21	0.07835	-	-	-	-
26	<i>Tetraponera sp</i>	5	0.303079	-	-	-	-
27	<i>Dorylus sp</i>	-	-	2	0.032965	-	-

### APPENDIX III: Ants collected in different seasons

S.N.	Species	Rainy	Shannon index	Autumn	Shannon index	Winter	Shannon index
1	<i>Camponotus sp1</i>	78	0.270987	104	0.118993	43	0.278887
2	<i>Camponotus sp2</i>	8	0.059477	28	0.032036	-	-
3	<i>Camponotus sp3</i>	3	0.027421	12	0.013729	3	0.04617
4	<i>Camponotus sp4</i>	3	0.027421	9	0.010297	-	-
5	<i>Polyrachis sp</i>	34	0.167221	45	0.051487	16	0.156674
6	<i>Paratrechina sp1</i>	28	0.147166	64	0.073226	16	0.156674
7	<i>Paratrechina sp2</i>	26	0.140005	37	0.042334	22	0.191996
8	<i>Lepisiota sp</i>	-	-	5	0.005720	1	0.019065

9	<i>Nylanderia sp</i>	28	0.147166	37	0.042334	1	0.019065
10	<i>Oecophylla sp</i>		-	3	0.003432	-	-
11	<i>Aphaenogaster sp</i>	56	0.226826	89	0.101830	51	0.30167
12	<i>Crematogaster sp1</i>	29	0.150652	42	0.04805	4	0.057714
13	<i>Crematogaster sp2</i>	9	0.065068	14	0.016018	3	0.04617
14	<i>Pheidole sp1</i>	26	0.140005	52	0.059496	-	-
15	<i>Pheidole sp2</i>	14	0.090459	29	0.033181	9	0.105448
16	<i>Monomorium sp</i>	24	0.132576	27	0.030892	-	-
17	<i>Meranoplus sp</i>		-	2	0.00229	-	-
18	<i>Lophomyrmex sp</i>	24	0.132576	32	0.036613	-	-
19	<i>Tetramorium sp</i>	9	0.065068	23	0.026316	-	-
20	<i>Leptogeny sp1</i>	38	0.179544	49	0.05606	22	0.191996
21	<i>Leptogeny sp2</i>	9	0.065068	17	0.019450	3	0.04617
22	<i>Brachyponera sp1</i>	49	0.209852	39	0.044622	39	0.265680
23	<i>Brachponera sp2</i>	12	0.080754	17	0.019451	11	0.121498
24	<i>Odontoponera sp</i>	62	0.240154	82	0.09382	49	0.296396

25	<i>Odotomachus sp</i>	4	0.03456 0	11	0.012586	6	0.078435
26	<i>Tetraoponera sp</i>	2	0.01969	3	0.003432	-	-
27	<i>Dorylus sp</i>			2	0.002288	-	-

**Appendix IV: Ant species richness with Shannon and Evenness index in seasons**

Diversity index/ Seasons	Rainy	Autumn	Winter
Species richness (S)	15	19	12
Shannon Diversity (H)	2.879	2.958	2.379
Evenness Index (J)	1.063	1.004	0.957
Number of ants collected	575	874	299