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Bombus tunicatus, *Anthidium florentium*, *Polistes nimpha* and *Eristalis cerealis* as important pollinators among the diversity of insect visitors on buckwheat (*Fagopyrum esculentum*) in Kargil, Ladakh

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Abstract
Pollinator species are usually limited, although flowers of some distylous plants are visited by diverse animals. During the present investigations, the pollination biology of buckwheat (*Fagopyrum esculentum*), a distylous crop mainly pollinated by honey bees in other countries like China, Nepal, Canada, America and visited by many insect species was studied. However, in this survey honey bees were not recorded and wild bees such as bumble bee were pre dominantly visiting and foraging on this crop in high altitude region of trans Himalaya. Other than bumble bees, *Anthidium florentinum, Andrena* sp., *Halictus* sp., *Polistes nimpha, Formica fusca,* hoverflies, some Lepidoptera and Coleoptera were also recorded from the buckwheat flowers. This paper is the first to show buckwheat pollinator diversity from Kargil region.

Keywords: Buckwheat, pollinators, Kargil, diversity, trans-Himalaya

Introduction

Buckwheat (*Fagropyrum esculentum* Moench; Polygonales: Polygonaceae) is an ancient crop thought to have its origins in China [1]. It was later introduced to the New World by European settlers during the 17th century [2]. Common buckwheat, scientifically known as *Fagopyrum esculentum*, holds global significance as a pseudo-cereal, covering an approximate harvested area of two million hectares [3,4]. The flowers of common buckwheat are distylous, meaning they contain pin and thrum morphs, which necessitates the pollination by insects carrying compatible pollen, specifically pollen from a different flower morph.

Buckwheat is a significant traditional crop grown in the high-altitude temperate zones of Ladakh, Jammu, and Kashmir. It used to be a staple food for the Ladakhi people until recently. Among the popular food items made from buckwheat flour in Kargil is "kiseer" or "giziri," which resembles plain dosa. However, despite its high medicinal and nutritive value, the cultivation of this nutritious crop is now on the brink of extinction. In Kargil district, buckwheat is known as "Rgyam brass" while in other regions of the district, it is called "Brow or Brow fhay." It is cultivated after the spring crop, after harvesting the first field crop barley and bajra. The sowing of buckwheat seeds begins in July, and flowering typically occurs from around 15th August until the seeds ripen, which takes about 10-12 weeks [5].

The crop holds significant economic importance mainly because of its protein and carbohydrate-rich grains, the resilience of its plants, rapid growth cycle, and the use of its foliage as a green vegetable. Additionally, buckwheat serves as a valuable resource for livestock and poultry feed, produces buckwheat honey, and acts as a beneficial cover for wildlife.

While honeybees are commonly recognized as the primary pollinators of buckwheat, various species of insects, including non-bee arthropods, might also have the potential to play a role as pollinators [6-9]. Insects play a vital role in the conservation and cultivation of buckwheat. They are major visitors to buckwheat flowers, relying on their nectar and pollen as a source of food and energy. Additionally, these insects assist in the crucial process of pollen transport from one flower to another, thereby facilitating the successful reproduction of buckwheat.

In this paper, we explore the pollinators and their diversity in the high-altitude region of trans-Himalayan Kargil district in Ladakh, focusing on the cultivated fields of buckwheat. Although buckwheat cultivation is relatively uncommon in this district, there has been a significant increase in production over the past decade. Preserving and promoting its cultivation is crucial for both the ecosystem and human livelihood.

Materials and methods

Study area

Field surveys were carried out in the Kargil district of Ladakh (Fig. 1). Observations were done from the second week of August 2020, 2021 and 2022 (Fig.2). Kargil is located within the geographic coordinates of approximately 30 to 35 degrees North latitude and 75 to 77 degrees East longitude. The district is strategically situated at the confluence of several river valleys. To the north and south, it is bordered by the Suru river valley, while the southeast leads to the Wakha Rong valley, which connects to Leh district. On the eastern side lies the Sodh Valley, connecting to the Indus Valley which eventually leads to Skardu. This advantageous positioning makes Kargil a vital crossroad linking Kashmir, Ladakh, and Baltistan. Kargil is positioned at an average elevation of 2,676 meters (8,780 feet) above sea level, Being a part of the Himalayas, Kargil experiences a temperate climate. Summers are characterized by hot days and cool nights, while winters are prolonged and chilly, with temperatures frequently plummeting below −20 °C (−4 °F).

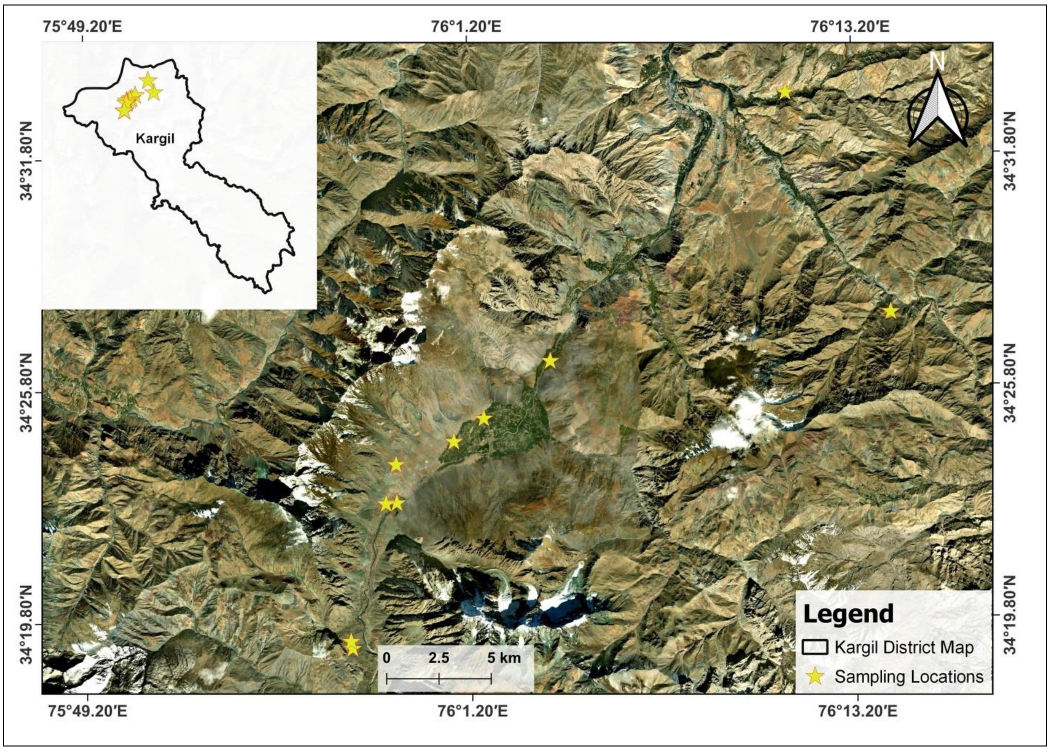

Fig 1: GIS map showing Kargil district in the left corner, rest are zoomed of the google earth image with sampling locations which are denoted by yellow stars in both images.

Fig 2: Study sites during flowering period of buckwheat (*Fagopyrum esculentum*) in the month of August

Collection and preservation

During the flowering season, an entomological hand net was used to sweep and collect pollinators from buckwheat during mid-August. Collection was made every two hours from early morning, around 9:30 am, until evening at 5:00 pm. Highest insect visitation was observed between 9:30 am and 1:00 pm during present investigation. The frequency of insect visits depended on sunny weather and low wind velocity. Field observations were carried out for 3 years consecutively. During field tour possible insect photography was done with the help of Nikon 3500D camera and mobile phone Redmi note pro5.

After collecting the insects with the sweep net, these were transferred to ethyl acetate fumigation jars for about 10-15 minutes before placing them on a thermocol sheet for stretching and pinning purposes. The stretching process was followed by a three-day drying period in an open room.

Once the insects were completely dry, each specimen was labeled and transferred to an entomological box for identification and preservation. The preservation process was carried out in the parent department of Zoology at Panjab University, Chandigarh.

Identification of species

Identification was carried out with the help of taxonomic keys, previous literature and confirmation was done through experts of different group of insects, such as bumble bees identified from "Desert Regional Centre, Zoological Survey of India" located in Jhalamand, Pali Road, Jodhpur, Rajasthan", non *Apis* bees confirmed from Department of Entomology, College of Agriculture and Sericulture, University of Agriculture Sciences, Bengaluru. Dipteran insects got confirmed from Diptera Section, Zoological Survey of India, Kolkata and butterflies insect got confirmed from Forest Research Institute Dehradun.

Analyzing of diversity data

Data analysis of the results was done with the help of the following diversity indices.

a. Shannon-Wiener Diversity Index: To quantify species diversity, used the Shannon-Wiener Diversity Index (H) [10]:

$$H = -\Sigma (Ni/N) \ln (Ni/N) \; i=1$$

Where: **Ni** = Number of individuals of species i

N = Total number of individuals across all species

b. Evenness Index: The Evenness Index, as described by [11] is calculated as follows:

$$E = H / \ln S$$

Where: **S** = Total number of species

N = Total number of individuals across all species H = Diversity Index

c. Margalef's Index: For a straightforward assessment of species richness, [12] was utilized:

Margalef's Index = **(S-1) / ln N**

Where: **S** = Total number of species

N = Total number of individuals in the sample

ln = Natural logarithm

d. Simpson index of diversity (1-D): Simpson's Diversity Index, introduced by [13], is a widely used and straightforward method for estimating the likelihood that two randomly selected entities from a given dataset belong to the same category or type. The Simpson index takes into account both the number of different categories (species) and their respective levels of dominance within the dataset. The resulting index value ranges from 0 to 1. When the value approaches 0, it indicates a low level of diversity, whereas values near or above 1 suggest high diversity.

$$D = \sum n_i(n_i-1)/N (N-1)$$

D = Simpson's diversity

Ni = total number of individuals of a particular species

N = total number of individuals of all species

Results

In present study 23 species of buckwheat pollinators were recorded these belonged to four orders of class Insecta including Hymenoptera comprising 8 species, Diptera with 9 species, Lepidoptera with 5 species and Coleoptera comprising of only single species (Table-1). On the basis of families in each recorded order the highest number was recorded in Hymenoptera (40%) followed by Diptera (33%) Lepidoptera (20%) and then Coleoptera (7%) (Fig.3). Highest number of species percentage was recorded by Diptera (39%) followed by Hymenoptera (35%), Lepidoptera (22%) and lowest by Coleoptera (4%) (Fig.4). Diversity indices of each recorded insect visitor of buckwheat were calculated. Highest Shannon Wiener index was recorded by Hymenoptera (1.93) followed by Diptera (1.91), Lepidoptera (1.47) and lowest by Coleoptera (). Highest species richness recorded by Diptera (1.51) followed by Hymenoptera (1.17), Lepidoptera (0.85) and then Coleoptera. Evenness was highest in Hymenoptera (0.92) followed by Lepidoptera (0.91), Diptera (0.87) and Coleoptera. Simpson index of diversity was highest in Hymenoptera (0.83) followed by Diptera (0.82), Lepidoptera (0.74) (Table-2). Number of genera was recorded equal in both Hymenoptera and Diptera order seven in each order followed by Lepidoptera four genera and Coleoptera recorded single genus reported (Fig.5).

Table 1: List of flower visiting insect pollinators of buckwheat (*Fagopyrum esculentum*) in Kargil, Ladakh.

S.N.	Common name	Scientific name	Family	Order
1	Bumblebee	*Bombus tunicatus (*Smith 1987*)*	Apidae	Hymenoptera
2	Bumblebee	*Bombus cryptarum* (Fabricius, 1775)	Apidae	Hymenoptera
3	Mining bee	*Andrena* sp.	Andrenidae	Hymenoptera
4	Sweat bee	*Halictus* sp.	Halictidae	Hymenoptera
5	Wool carder bees	*Anthidium florentinum*(Fabricius,1775)	Megachilidae	Hymenoptera
6	Ant	*Formica fusca* (Linnaeus,1758)	Formicidae	Hymenoptera
7	Eusocial paper wasp	*Polistes nimpha* (Christ 1791)	Vespidae	Hymenoptera
8	German yellow jacket	*Vespula germanica* (Fabricius, 1793)	Vespidae	Hymenoptera
9	Drone fly	*Eristalis tenax* (Linnaeus, 1758)	Syrphidae	Diptera
10	Drone fly	*Eristalis cerealis* (Fabricius, 1805)	Syrphidae	Diptera

11	Tachinid flies	*Tachina fera* (Linnaeus, 1761)	Tachinidae	Diptera
12	Tachinid flies	*Tachina ursina* (Meigen, 1824)	Tachinidae	Diptera
13	Hornet Hoverfly	*Volucella zonaria* (Poda, 1761)	Syrphidae	Diptera
14	Bee fly	*Anthrax aperta* (Walker, 1852)	Bombyliidae	Diptera
15	Bee fly	*Thyridanthrax* sp. Osten-Sacken, 1886	Bombyliidae	Diptera
16	Waisted Beegrabber	*Physocephala aurantiaca* (Brunetti,1923)	Conopidae	Diptera
17	Red-tailed flesh fly	*Sarcophaga pernix* (Harris, 1780)	Sarcophagidae	Diptera
18	Seven-spot ladybird	*Coccinella septempunctata* (Linnaeus, 1758)	Coccinellidae	Coleoptera
19	Cabbage butterfly	*Pieris brassicae* (Linnaeus, 1758)	Pieridae	Lepidoptera
20	Painted lady butterfly	*Vanessa cardui* (Linnaeus, 1758)	Nymphalidae	Lepidoptera
21	Common Blue butterfly	*Polyommatus icarus* (Rottemburg, 1775)	Lycaenidae	Lepidoptera
22	Common Meadow Blue butterfly	*Polyommatus stoliczkana* (Smith 1989)	Lycaenidae	Lepidoptera
23	Hill Hedge Blue butterfly	*Celastrina argiolus* kollari (Linnaeus, 1758)	Lycaenidae	Lepidoptera

Table 2: Buckwheat (*Fagopyrum esculentum*) pollinator diversity, Richness and Evenness index and Simpson index

Order	No. of family recorded	No. of species	Species Diversity	Species Evenness	Species richness	Simpson index
Hymenoptera	6	8	1.93	0.92	1.17	0.83
Diptera	5	9	1.91	0.87	1.51	0.82
Lepidoptera	3	5	1.47	0.91	0.85	0.74
Coleoptera	1	1				

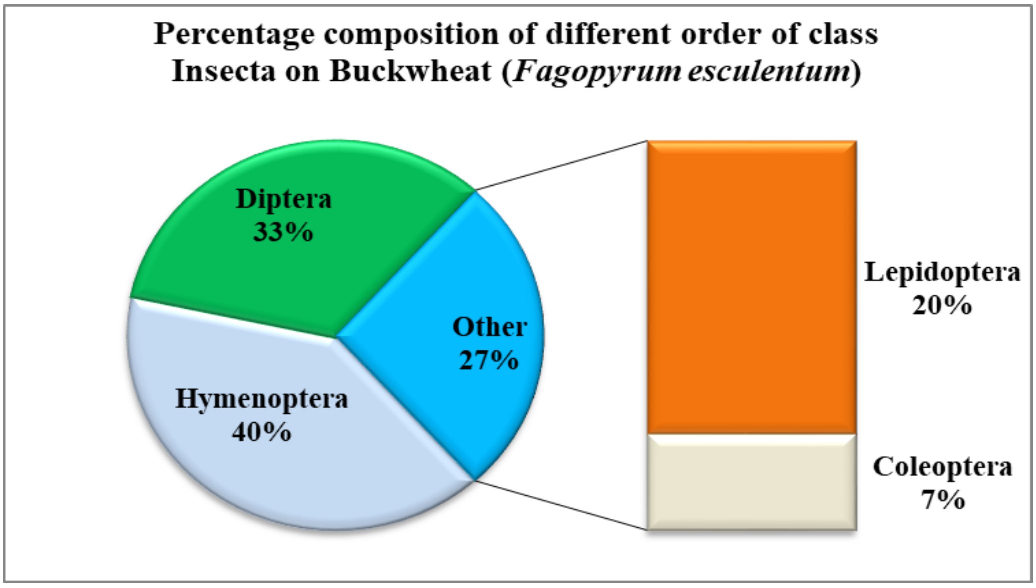

Fig 3: Number of families in each recorded order.

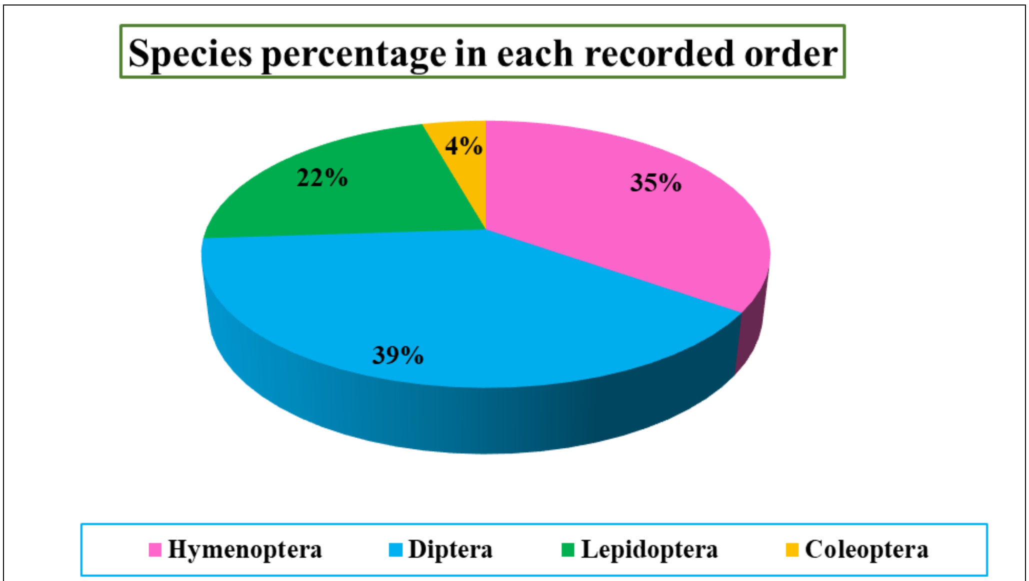

Fig 4: Number of species percentage in each recorded order

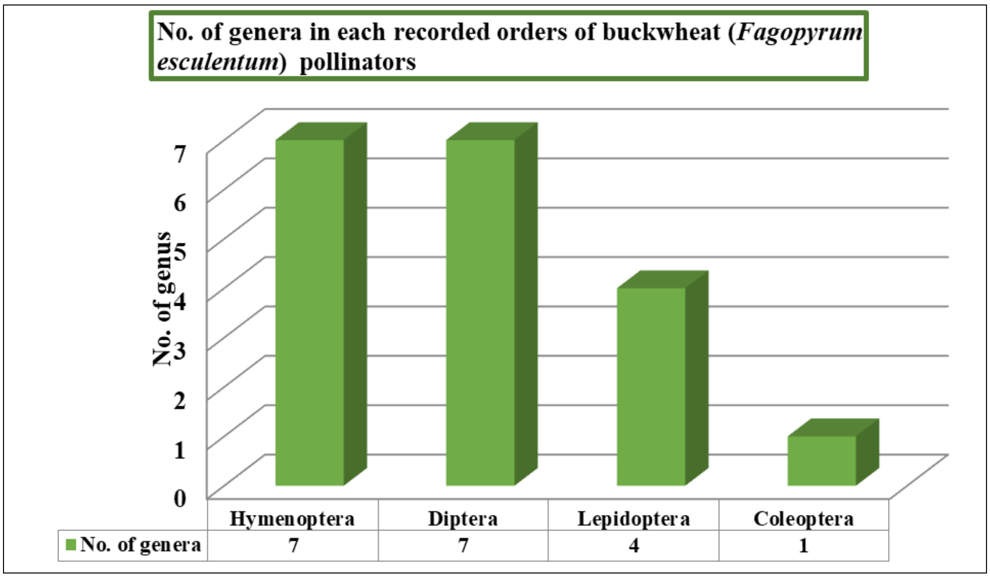

Fig 5: Number of genera in each recorded order

Discussion

Typically, the buckwheat flower lacks the ability to self-pollinate due to its specific structure that prevents automatic contact between pollen and the stigma. However, occasional flowers with pistils and stamens of equal length may exhibit a modest level of self-fertility. Recently, a selection (*F. sagittatum* Gilib.) has been developed, featuring stamens and pistils at the same level, leading to a significantly higher degree of self-fertility. Nonetheless, this particular variety holds no direct commercial value [14]. Since buckwheat pollen is not dispersed by wind, the assistance of insects is essential for the effective transfer of pollen between flowers. The pseudocereal crop established the significance of insect pollination for commercial seed production of buckwheat in the United States [15]. Similarly, several researchers in Russia, where buckwheat is extensively cultivated, have also confirmed this necessity [16-18]. After conducting a comprehensive review of buckwheat pollination, emphasized the importance of conducting controlled cage tests on this crop [19]. These tests are crucial for determining the extent of self-pollination, if any, and for assessing the quantity of seed that can be expected under various pollination conditions. *Bombus tunicatus*, *Andrena* sp. and *Halictus* sp. *Polistes nympha* hoverflies, bee like flies, emerge as the most efficient pollinators for buckwheat due to their strong attraction to buckwheat flowers and the ability to effectively transfer pollen from anthers to stigmas, whether through collecting pollen or nectar.

After examining and analyzing all the recorded data, it was observed that in high-altitude regions, bees (Hymenoptera), flies (Diptera), and butterflies (Lepidoptera) were the most common insect pollinators of buckwheat. The whitish color of the flowers proved highly attractive to various insect pollinators, which actively collect pollen and nectar from them. However, their activities are greatly influenced by the prevailing weather conditions. Sudden changes in weather can also impact buckwheat pollinators in the area. Despite the challenges, buckwheat can be successfully cultivated in these cold climate conditions because of the diversity of pollinators visiting it. These include bumblebees, non-*Apis* bees, wasps, ants, syrphid flies, bee-like flies, various butterfly species, and beetles. These insects play an active role in pollinating the crop, and contributing to its successful reproduction.

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