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PCA Study of the Factors in the Soil That Affect the Population Density of a Collembolan *Bilobella braunerae* (Dhervang 1981) in the Evergreen Forest Southern Western Ghats, India during Various Seasons

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Abstract: The objective of the current study was to determine how soil physic-chemical characteristics affected the population density and vertical distribution pattern of the soil collembolan *Bilobella braunerae* in the Southern Western Ghats' evergreen forest. The findings showed that the post-monsoon season (43.9) had the highest mean abundance of collembola, followed by the monsoon (23.15) and pre-monsoon (17.0). During the post-monsoon season, there was a significant population density of organisms in the 0–10 cm layer of soil. During the pre-monsoon season, a vertical migratory pattern was noted along with an increase in soil temperature and a decrease in moisture content. During monsoon season owing to heavy rainfall, organism showed a vertical migration to 10-20 cm depth and thus lesser number of organisms were seen in 0-10 cm layer of soil. Seasons exerted a strong effect on the abundance of *B. braunerae* in Evergreen Forest. To determine whether soil components have a direct impact on population, Principal Component Analysis (PCA) was applied which revealed that the population density of *B. braunerae* was significantly affected by soil pH, organic carbon, moisture, temperature, phosphorus, and potassium.

Keywords: *Bilobella braunerae*, Physico-chemical parameters, Population density, Vertical migration, Principal Component Analysis (PCA)

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Introduction

According to studies by Hodkinson *et al*. (1996), Andrés (1999) and Kautz *et al*. (2006), soil microarthropods are cosmopolitan in distribution, found from the equator to the poles, and have the ability to adapt to the soil, making them useful tools for assessing the health of ecosystems and studying changes in ecosystems. Soil fauna are thought to be useful indicators of soil quality as they are sensitive to changes in land management and are involved in many soil functions (Stork and Eggleton, 1992; Paoletti and Hassall, 1999; Yeates, 2003). Total soil organic matter influences soil compatibility and soil water-holding capacity while aggregated soil organic matter has major role in regulating air and water infiltration, conserving nutrients, and influencing soil permeability and erodibility (Carter, 2002). The soil quality indicators are the measurable soil factors that influence the ability of soil to sustain crop production and perform several other environmental function (Arshad and Martin, 2002).

Acarina (mites) and Collembola (springtail) are the most diverse and abundant arthropods (Wallwork, 1976). Among soil biota, soil arthropods are considered useful tool as bio-indicator (Menta *et al*., 2018). Acari and Collembola are the two groups which are generally considered in soil quality evaluation approach (Menta and Remelli, 2020). The rainforest conversion into plantation systems is associated with marked shifts in the structure of trophic niches in soil and litter collembolans with potential consequences for ecosystem functioning and food-web stability. There are about 8600 described species of Collembola worldwide (Luxton, 1982). Indian fauna of Collembola represents 299 species under 103 genera and 18 families (Janssens, 2007). The distribution of collembola in the subsoil below 30 cm depended on root biomass and total soil organic content. The density of collembolans correlated significantly with root biomass both in the upper and lower parts of the soil profile which suggests that soil fauna are involved in deep soil carbon cycling largely via grazing on root-associated micro-organisms (Potapov *et al*., 2017). Frouz (2018) reported that soil fauna consumes substantial amounts of litter and during gut litter modifications there is increase in the proportion of lignin, and decrease in soluble polyphenols and carbon: nitrogen (C: N) ratios. The soil fauna affects microbial activity in soil by leaching and the release of particulate organic matter and at a larger scale it can change the soil profile, soil properties, and even the plant community. This type of faunal effects seen in ecosystems in primary or secondary succession stage (Heneghan and Bolger, 1998; Frouz, 2018). During marshland restoration soil surface-dwelling Collembola recovered faster than eu-edaphic species, due to parthenogenesis and fast dispersal between epiedaphic and hemiedaphic species (Dou *et al*., 2019).

The objective of the current study was to determine how soil physic-chemical characteristics affected the population density and vertical distribution pattern of the soil collembolan *Bilobella braunerae* in the Southern Western Ghats' evergreen forest, India.

Materials and Methods

Study Area

Soil samples were randomly collected from 20 different sites of 5X5 cm2 area, from different places in Evergreen Forest Thannithode Forest Upper Zone 90 28’22” N, 76055’ 31” E of Pathanamthitta District, Kerala using a soil auger. The soil through the study area is sandy loam. The soil samples were collected bimonthly for a period of one year from October 2019 to September 2020. Soil samples were collected from 0-10. 10-20 and 20-30 cm layers of the soil pit random collected from each study area during different seasons. Temperature was recorded at site using soil thermometer. The soil samples were collected in a polythene bag, tied using rubber band, properly marked and taken to the laboratory. Samples were pooled, air dried, sieved through 2 mm sieve. The sand, silt and clay contents were analyzed by Particle size analysis, pipette method. Soil pH was measured by pH meter in water suspension (1:2 ratios). Organic carbon content was measured by Walkley Black acid digestion method, total Nitrogen by Kjeldahl distillation method; Phosphorus content by Molybdate-Stannous chloride method; Potassium content by flame photometry method , Exchangeable Acid, Exchangeable base, Calcium and Magnesium by the method of Jackson (1958). Soil temperature was measured using a soil thermometer. Twenty soil samples were collected from study sites during pre-monsoon (February to May), monsoon (June to September) and post-monsoon season (October to January). Mean value along with

Table 1: Mean Population Density of *Bilobella braunerae* in Evergreen Forest in different seasons (Density/25 cm^3)

Seasons	**Evergreen Forest (EG) Mean±SD**
Pre-monsoon	17±0.01205
Monsoon	28.1±0.0095
Post-monsoon	43.95±0.0084

standard deviation was calculated then converted in to area (cm^2).

Population Density of Soil Fauna

With the use of a soil auger, random soil samples of 25 cm2 and up to 30 cm in depth were taken. For an entire year, samples were taken to the laboratory in labeled polythene covers twice a month in the morning, between the hours of 8:00 and 10:00. The Berlese Tullgren funnel was used to extract *B. braunerae* from collected soil samples, and the samples were left overnight in a medium containing picric acid.

Data Analysis

In a 25 cm^2 area, the collembolan density was estimated. The population mean value across 20 sites was calculated along with the standard deviation. To investigate changes in the collembola population density between sites and seasons, a two-way ANOVA was used. PAST 3 software was used to perform PCA of soil.

Results

Population Density and Distribution Pattern of B. braunerae

The post-monsoon season (43.95±0.0084) had the largest population density in the Evergreen Forest habitat, followed by the monsoon season (28.1±0.0095) and the pre-monsoon season (17±0.01205) (Table 1). According to the results of the two-way ANOVA, there was a significant difference between the seasons (F= 3.8E+07, F critical= 3.24482; P<0.05) but no difference in population density between the sites (F= 1.15496, F critical= 1.86733; P>0.05).

Vertical distribution pattern of Bilobella braunerae in Evergreen Forest

During the post-monsoon and monsoon seasons in the evergreen forest, the highest concentration of creatures were observed in the 0–10 cm soil profile; in contrast, during the pre-monsoon, the majority of organisms were found in the 10–20 cm layer and less in the 0–10 cm layer. The population of collembolan is lower in the 20–30 cm layer of soil in all seasons (Fig. 1). The findings of the two-way ANOVA demonstrated a significant difference between the seasons (F=7.87345, F crit = 6.944272; P<0.05) and the vertical distribution pattern of *Bilobella braunerae* (F=9.87726, F crit = 6.944272; P<0.05).

Soil Physico-chemical Parameters

The physic-chemical parameters of soil were considered for the present study to find out the factor which affects the distribution of *B. braunerae* in each season (Table 2). The soil parameters studied were moisture, temperature, pH, OC (organic carbon), EA (exchangeable acid), EB (Exchangeable Base), Sand (Sa), Silt (Si), Clay (Cla), Nitrogen (N), Phosphorous (P), Potassium (K), Magnesium (Mg), and Calcium (Ca). Soil moisture was high in monsoon to post-monsoon and less during pre-monsoon season. It ranged from 78% -89% in seasons. The soil temperature was at an average of 21.39°C during monsoon and 28.33°C in pre-monsoon season The soil of Evergreen Forests in the monsoon season was extremely acidic (3.7875**±**0.024) and acidic in pre-

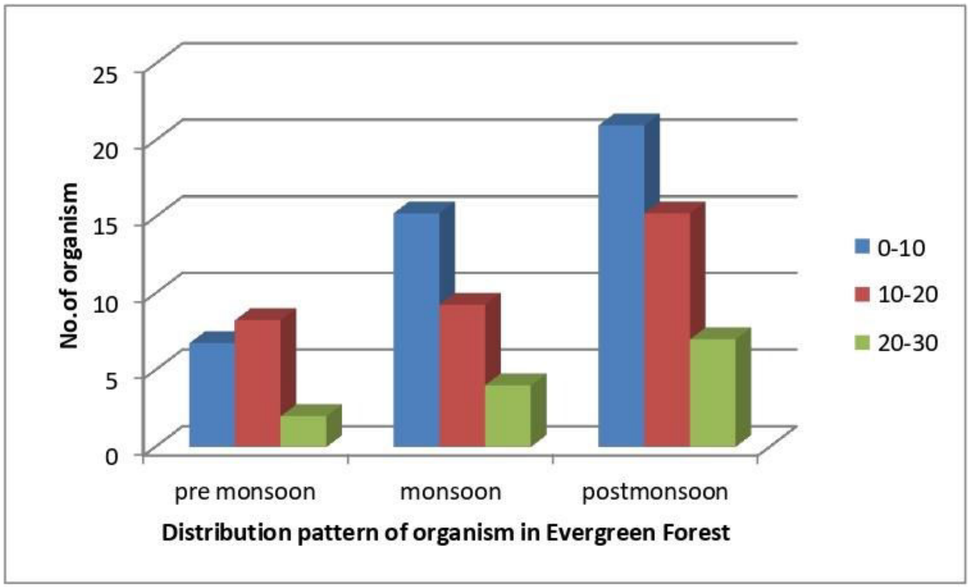

Fig. 1: Vertical distribution pattern of *Bilobella braunerae* in evergreen forest (25 cm ^{2} area)

Table 2: Soil properties during various seasons in Evergreen Forest of Thannithode Forest

Soil Parameters	Pre-Monsoon Mean±SD	Monsoon Mean±SD	Post-Monsoon Mean±SD
Moisture (%)	78.56±0.407	89.23±0.330	84.01±0.42
Temperature (^{0}C)	28.13±0.164	21.39±0.175	24.36±0.18
pH	4.794±0.0154	3.7875±0.024	4.9±0.02
OC	4.71±0.022	4.87±0.053	5.15±0.01
Sand (%)	71.03±0.615	74.90±0.569	75.52±0.461
Silt (%)	9.76±0.137	12.70±0.095	13.09±0.332
Clay (%)	19.20±0.341	12.32±0.149	11.26±0.225
EA (%)	3.658±0.007	3.705±0.031	1.43±0.025
EB%	5.777±0.025	5.35±0.088	3.4±0.02
N (ppm)	5646.4±0.666	4675.05±0.670	3755±0.74
P (ppm)	8.83±0.0376	6.60±0.343	4.726±0.018
K (ppm)	267.975±0.639	292.69±0.584	152.43±0.28
Ca (ppm)	436.633±0.191	514.16±0.642	143.08±0.39
Mg (ppm)	35.6025±0.161	67.49±0.232	48.301±0.18

monsoon (4.794±0.0154) and post-monsoon (4.9**±**0.02). Organic carbon content was high in Evergreen Forest soil and it was above 4 at all the sites in all seasons and it ranged between 4.71-5.15. Exchangeable acid was found in the range of 1.43 % to 3.7% and exchangeable base between 3.4 % to 5.7%. The soil texture was found to be sandy loam base on the percentage of sand, silt and clay. As the percentage of sand and clay is high there is absence of luxuriant vegetation and community has degraded into grassland. The average value of Nitrogen in the Evergreen Forest was highest during pre-monsoon (5646 ppm) followed by monsoon (4675 ppm) and post-monsoon (3755 ppm). The Phosphorous content was high in pre-monsoon (8.83 ppm) followed by monsoon (6.60 ppm) and post-monsoon (4.726 ppm) in Evergreen Forest soil. The Potassium

Table3: Eigen value of PC in Evergreen Forest Habitat during pre-monsoon

PC	Eigen value	% variance
1	2.82505	18.834
2	2.50802	16.72
3	1.96892	13.126
4	1.57207	10.48
5	1.3253	8.8354
6	1.08922	7.2615
7	0.886867	5.9124
8	0.655479	4.3699
9	0.567401	3.7827

content was high in monsoon (292.69 ppm) followed by pre-monsoon (267.975 ppm) and post-monsoon (152.43 ppm) in Evergreen Forest soil. Nitrogen, phosphorous and potassium content was found to be high in soil and so luxuriant vegetation was seen. As soil in forests was extremely acidic and sandy loam in nature, soil calcium content was average and was less than 610 ppm.The magnesium content was optimum in forest soil as it was below 77 ppm. The soil texture of Konni Evergreen Forest was loamy sand, sandy loam and loamy sand, respectively based on the percentage of sand, silt and clay.

PCA of Soil Parameters

A PCA of the soil's chemical and edaphic parameters was conducted in order to identify the primary factor influencing the population density of *B. braunerae* in Evergreen Forest. The most significant principal components are those with eigen values greater than one. Five components in the premonsoon season's PCA of the soil have eigen values greater than one (Table 3). 74.11% of the total variance of the original data was accounted for by the percentages of the total variances of the five extracted main components. These five derived components thus account for nearly all of the variance in the original data**.**

Evergreen Forest
Pre-monsoon *Season*:

The PCA analysis of soil parameters during premonsoon season from Evergreen Forest habitat is based on three princiale component factors (PC1, PC2, and PC3). These three components when accumulated they account for 77 % of the total variance. Primary factors under PC1 which influenced the distribution of *B.braunerae* in Evergreen Forest during pre-monsoon period were, Moisture (0.67018), Temperature (0.4962), pH (0.292), OC (0.18305), Sand (0.6597), Silt (0.4764), EA (0.5388), EB (0.50806), Nitrogen (0.21475), Potassium (0.32504) and Ca (0.61854) (Table 4). PC1 analysis of fourteen soil parameters for collembolan abundance during pre-monsoon yielded eleven components having positive values and three components with negative values. Under PC2, factors like EB (0.39125), silt (0.2738), clay (0.1872), Nitrogen (0.64772) and Magnesium (0.39137) were found to influence collembolan distribution during pre-monsoon period. Under PC3, factors such as pH (0.4727), sand (0.078), silt (0.2597), clay (0.46325), Nitrogen (0.33127), Phosphorous (0.1107), and Potassium (0.57089) were found to influence the distribution in soil during pre-monsoon season (Table 4). Only two components Nitrogen and silt had positive values in PC1, PC2, and PC3.

Monsoon Season

In the PCA analysis of soil parameters during monsoon season, in the Evergreen Forest, the first

Table 4: PCA values of various soil factors controlling the density and distribution of *Bilobella braunerae* in Evergreen Forest Habitat during pre-monsoon Season

Variables	PC 1 (eigen value)	PC 2 (eigen value)	PC 3 (eigen value)
Moisture	0.67018	-0.08697	-0.15634
Temperature	0.49662	-0.04196	-0.13263
pH	0.29207	-0.35928	0.47277
Organic Carbon	0.18305	-0.15915	-0.62001
EA	0.5388	-0.57654	-0.24946
EB	0.50806	0.39125	-0.05915
Sand	0.65976	-0.19871	0.078716
Silt	0.47647	0.27381	0.2597
Clay	-0.22054	0.1872	0.46325
Nitrogen	0.21475	0.64772	0.33127
Phosphorous	-0.16591	-0.7687	0.11078
Potassium	0.32504	-0.23991	0.57089
Calcium	0.61854	-0.12965	-0.15136
Magnesium	-0.08703	0.39137	-0.69908
Organism	0.42616	0.68654	0.073815

Table 5: Eigen value of PC in Evergreen Forest Habitat during monsoon

PC	Eigen value	% variance
1	2.72571	18.171
2	2.51295	16.753
3	2.05019	13.668
4	1.65152	11.01
5	1.33347	8.8898
6	1.20833	8.0555
7	0.932667	6.2178
8	0.76006	5.0671
9	0.509872	3.3991

three principal components account for 71% of the total variance. Primary factors under PC1 during monsoon period were temperature (0.63602), pH (0.61056), EB (0.6298), Nitrogen (0.41968), and Calcium (0.448) (Table 6). PC1 analysis of fourteen soil parameters during monsoon revealed that five components had positive values and nine components with negative values. Under PC2, factors like Moisture (0.5362), Temperature (0.4608), pH (0.4387), OC (0.58407), Sand (0.080831), EA (0.29316), EB (0.51917), Nitrogen (0.16066), and Magnesium (0.66635) were found to influence collembolan distribution during monsoon period. Under PC3, factors such as Moisture (0.30503), Temperature (0.31129), pH (0.30058), OC (0.30181), EA (0.43515), silt (0.24307), clay (0.38257), Phosphorous (0.41814), Potassium (0.52301) and Ca (0.74053) were found to influence the distribution in soil during monsoon season (Table 6). The temperature and pH crucially influence the distribution of organism with positive loadings

Table 6: PCA values of various soil factors controlling the density and distribution of *Bilobella braunerae* in Evergreen Forest Habitat during monsoon season

Variables	PC1 (eigen value)	PC2 (eigen value)	PC3 (eigen value)
Moisture	-0.26168	0.5362	0.30503
Temperature	0.63602	0.4608	0.31129
pH	0.61056	0.43871	0.30058
OrganicCarbon	-0.37865	0.58407	0.30181
EA	-0.2746	0.29316	0.43515
EB	0.62983	0.19179	-0.03483
Sand	-0.17759	0.080831	-0.14931
Silt	-0.15567	-0.65492	0.24307
Clay	-0.41062	-0.01664	0.38257
Nitrogen	0.41968	0.16066	-0.31706
Phosphorous	-0.34008	-0.04721	0.41814
Potassium	-0.30201	-0.45697	0.52301
Calcium	0.4487	-0.00631	0.74053
Magnesium	-0.48229	0.66635	-0.0742
Organism	0.47237	-0.49003	0.39712

Table 7: Eigen value of PC in Evergreen Forest Habitat during post-monsoon

PC	Eigen value	% variance
1	2.51221	16.748
2	2.45498	16.367
3	1.90802	12.72
4	1.61991	10.799
5	1.31247	8.7498
6	1.1198	7.4653
7	0.847767	5.6518
8	0.769327	5.1288
9	0.737631	4.9175

in PC1, PC2 and PC 3.

Post-monsoon Season

During post-monsoon season in evergreen forests, the first three principal components account for 75 % of the total variance. Primary factors under PC1 during post-monsoon period were Moisture (0.004892), EA (0.47185), EB (0.28184), Sand (0.57363), Silt (0.10858), Phosphorous (0.58975), Potassium (0.30809) and Calcium (0.29638) (Table 8). PC1 analysis of fourteen soil parameters during post-monsoon revealed that eight components had positive values and six components with negative values. The PC2, factors Temperature (0.22937), pH (0.41106), OC (0.23168), EA (0.39562), EB (0.5601), Silt (0.72926), Potassium (0.40741), Ca (0.44397) and Mg (0.033295) were found to influence collembolan distribution during post-monsoon period. Under PC3, factors such as Moisture (0.20015), pH (0.74251), EB (0.3478), sand (0.010314), silt (0.105), Phosphorous (0.070634), Ca (0.40395) and Mg (0.22047) were found to influence the distribution in post-monsoon season (Table 8). The soil variables Moisture, EB, Silt, and Calcium, with positive loadings in PC1, PC2, and PC3, play a significant role in the distribution of organisms.

Discussion

Principal Component Analysis (PCA) was conducted to find out soil edaphic and chemical

Table 8: PCA values of various soil factors controlling the density and distribution of *Bilobella braunerae* in Evergreen Forest Habitat during post-monsoon Season

Variables	PC1 (eigen value)	PC2 (eigen value)	PC3 (eigen value)
Moisture	0.004892	-0.26903	0.20015
Temperature	-0.70931	0.22937	-0.15199
pH	-0.05688	0.41106	0.74251
Organic Carbon	-0.44055	0.23168	-0.44077
EA	0.47185	0.39562	-0.58871
EB	0.28184	0.5601	0.3478
Sand	0.57363	-0.00036	0.010314
Silt	0.10858	0.72926	0.105
Clay	-0.27266	0.30382	-0.25591
Nitrogen	-0.15896	0.54111	-0.06066
Phosphorous	0.58975	-0.33561	0.070634
Potassium	0.30809	0.40741	-0.57545
Calcium	0.29638	0.44397	0.40395
Magnesium	-0.73392	0.033295	0.22047

factors influencing the distribution of *Bilobella braunerae*. PCA was applied to the 14 soil variables during three seasons. Primary factors under PC1 which influenced the distribution of *Bilobella braunerae* in Evergreen Forest during the pre-monsoon period were Moisture, Temperature, pH, Organic Carbon, Sand, Silt, EA, EB, Nitrogen, Potassium, and Calcium. Furthermore, the first principal component correlates strongly with the moisture, sand, calcium and OC which had strong positive loading. The abundance of Collembola was positively correlated with soil moisture (Choi *et al*., 2006; Shultz *et al*., 2006; Kardol *et al*., 2011; Abbas and Parwez, 2012). During the monsoon season, temperature, pH, EB, Nitrogen, and Calcium were the principal soil components influencing the population density of the collembolan. The soil variables temperature, pH, and EB were having strong positive loadings. In monsoon season moisture content has negative values. The high moisture contents may have a limiting effect on the Collembola and therefore a weak correlation between abundance and soil moisture was noted by Asif *et al*. (2016). Moisture, EA, EB, Sand, Silt, Phosphorous, Potassium, and Calcium have positive correlation especially Sand and Phosphorous were having strong positive values and temperature had a strong negative loading during the post-monsoon season.

Moisture content was high in Evergreen Forests (above 77%), compared to semi-evergreen and moist deciduous forests in all seasons. The moisture content was high during monsoon season and less during the pre-monsoon season. The soil moisture content was always above 77% in the Evergreen Forests during all seasons which support the luxuriant growth of vegetation. The soil temperature was low during the monsoon season (June to September) due to heavy rain caused by the southwest monsoon. The decrease in rainfall during pre-monsoon (February to May) increases the soil temperature by 5-7 ^{0}C compared to the monsoon season. The pH of forest soil was acidic in general due to humid climate and decomposing organic matter. The soil was extremely acidic in the evergreen and semi-evergreen forests whereas moderately acidic in moist deciduous forests. Organic carbon content was high during the post-monsoon season in all forest types and less in pre-monsoon. Among forests, organic carbon percentage was above 4% in Evergreen Forests in all seasons and comparatively less in moist deciduous forest. Generally, forest soil is rich in organic matter, acidic, high in nitrogen and poor in bases, due to heavy leaching and are dark reddish-brown to black with a loamy to silty loam texture (Swarupanandan *et al*., 1998). In ideal soil conditions the proportion of NPK is 3:1:2, which were present in forest soil. The Nitrogen content in Evergreen Forest soil was more than 3282 ppm which was reasonably higher than moist deciduous forest. So, luxuriant growth of vegetation was seen in evergreen forests. The phosphorous content was medium in forest soils as it ranged between 4.72 ppm to 8.83 ppm. The potassium content was high in forest soils as it was between 152.43 to 505.5 ppm. As soil in forests was extremely acidic and sandy loam in nature, soil calcium content was average and was less than 610 ppm.The magnesium content was optimum in forest soil as it was below 77 ppm.

The results of the PCA demonstrate the relevance of the multivariate statistical analysis methods in identifying the primary factor influencing the population of collembolan. During the pre-monsoon season, the soil variables pH, Organic Carbon, EA, EB, Sand, Silt, and Calcium showed positive values. During the pre-monsoon, there was a negative loading for temperature. *Bilobella braunerae* density declines with increasing soil temperature. According to Badejo and Van Straalen (1992), variations in springtail numbers are highly dependent on soil temperature. *Bilobella braunerae* population was positively affected by sand, silt, and calcium during the monsoon. In every habitat, a negative association between pH and organic carbon content was seen during the monsoon season, which led to an increase in organic matter. A negative association was seen in all habitats during the monsoon season between the pH and the amount of organic carbon, suggesting that an increase in organic matter led to a reduction in soil pH. While in the post-monsoon season, moisture, organic carbon, EB, EA, and potassium were the key soil components impacting the density of organisms. The amount of moisture and organic carbon content were observed to positively correlate during the post-monsoon season, which increased the population density of *Bilobella braunerae*. These outcomes resemble those of Tripathi *et al*. (2007). Collembolan species choose their habitats based on a variety of factors, including soil organic carbon, moisture, silt, clay, and calcium (Bisht, 2002).

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