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Microbial Analysis of Indian Flying Fox (*Pteropus giganteus*) Ejecta Collected from Two Public Parks in Lahore, Pakistan

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ABSTRACT

Microbial analysis of Indian flying fox, *Pteropus giganteus* ejecta roosting at Jinnah and Lalazar Gardens, Lahore was carried out from January, through December, 2011 and a total of twelve fungal and twelve bacterial genera were isolated. Four fungal (*Candida, Fusarium, Penicillium* and *Saccharomyces*) and two bacterial genera (*Klebsiella* and *Nocardia*) were isolated from bolus only, three fungal (*Cryptococcus, Histoplasma* and *Trichophoton*) and six bacterial (*Acaligenes*, *Azotobacter*, *Bartonella, Nitrsomonas, Pseudomonas* and *Salmonella*) genera were isolated from guano while five fungal (*Alternaria, Aspergillus, Chrysosporium, Exophila*and *Scopulariopsis*) and four bacterial (*Bacillus, Corynebacterium, Listeria* and *Streptomycete*) genera were common in bolus and guano samples. Seasonal variations were recorded in occurrence of various fungal and bacterial genera. From bolus samples, two fungal *Aspergillus* and *Fusarium* and one bacterial *Bacillus* genera were recorded throughout the year while from guano *Bacillus* was the only genus with year round occurrence. Microbial analysis shows that Indian flying fox ejecta are an amalgam of beneficial and pathogenic microbes and its pH (6.7 to 7.4), high concentration of phosphorus (4.50% and 4.33%) and nitrogen (3.26% and 2.37%) favor seed germination, enhance root growth and soil fertility.

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Authors' Contributions
TLG collected and analyzed the data, and wrote the article. SD helped in preparation of media, purification and staining of bacterial isolates. MS helped in identification of bacteria and fungi. Irfan helped in physico-chemical analysis of ejecta samples. SMH helped in interpretation of data. AJ supervised the study.

Key words
Fruit bat, Pathogen, Bolus, Guano.

INTRODUCTION

Bats (Order: Chiroptera) are the only mammals capable of true flight and can cross the barriers other mammals cannot (Willson *et al*., 1989). They are present everywhere except Antarctica (Hutson *et al*., 2001) and are divided in two major groups, the Megachiroptera and the Microchiroptera. The Megachiroptera are frugivorous bats and can cover a distance of 50 km in search of food in a single night (van der Pijl, 1957). In Pakistan, fruit bats are represented by three genera and four species, the short-nosed fruit bat *Cynopterus sphinx*, the Indian flying fox *P. giganteus*, the Egyptian fruit bat *Rousettus aegyptiacus* and the fulvous fruit bat *R. leschanaultii*. However, they are least study in the country (Roberts, 1997; Shahbaz *et al*., 2014).

The fruit bats are important reservoirs of many pathogens, some of which have been reported to be associated with many diseases like rabies (Paez *et al*., 2003), European lyssa virus (Fooks *et al*., 2002), Hendra (Halpin *et al*., 2000) and Menangle (Bowden *et al*., 2001) in Australia, Nipah and Tioman viruses in Malaysia (Chua *et al*., 2002a, b) and hantaviruses in Korea (Kim *et al*., 1994; Chua *et al*., 2005). Ejecta of the fruit bats supports a great diversity of organisms including arthropods, fungi, bacteria and lichens (Ferreira and Martins, 1998) and are most common sources of pathogenic and other mycofauna distribution. The differences in composition of bats' ejecta suggest that bats in different feeding guilds may affect ecosystem structure and dynamics (Justin and Roark, 2007).

Due to the close proximity of bats with humans and domestic animals, it is possible that they had important role in the epidemiology and zoonoses. The contact of bats with humans and domestic animals are either direct or indirect, for example through many hematophagus arthropods such as mosquitos, ticks (Pavlovsky, 1996) and cone-nosed bugs (Albuquerque and Barreto, 1968) feeds on bats, domestic animals and man. They are thought to be transferring half of the communicable diseases in man and act as a reservoir, intermediate host or vector of various pathogens (Freitas *et al*., 1960). The fungi related to bat excreta are mostly limited to the places where bat guano

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is frequently abundant (Darling, 1906). The association between bats and pathogenic fungi was first reported by Emmons (1958) who isolated *Histoplasma capsulatum* from soil contaminated by bat guano in Maryland. Over the next two decades, approximately 30 chiropteran species were identified as hosts for pathogenic fungi (Carvajal, 1977; Reis and Mok, 1979).

Bat guano had also reported to contain beneficial fungi and bacteria, which act as a natural fungicide to protect plants from diseases. Bacteria and fungi play important role to maintain soil health. Bacteria are necessary for plant growth on new fresh sediments. Bacteria fix atmospheric nitrogen and carbon, produce organic matter and immobilize enough nitrogen and other nutrients to initiate nitrogen cycling process in the soil (Lane and Diver, 2000). The clay is the most abundant of all the minerals in the fresh guano while others include quartz and traces of dolomite and calcite. The most abundant elements in bat guano are nitrogen and phosphorus. The total nitrogen ranges between 8–12% and P_2O_5 ranges between 2–7%. Other elements include calcium, magnesium, potassium, aluminum, iron and sulphur that are present in quantities lower than 5% each (Ruth *et al*., 2004). Differences in community structure of the microbes inhabiting guano may be due to differences in guano composition of frugivorous (*P. rodricensis*), sanguivorous (*Desmodus rotundus*), and insectivorous (*Tadarida brasiliensis*) bats. *Desmodus* guano contained more carbon than *Pteropus* guano. The latter contained less nitrogen, and the former contained less phosphorous than guano of the other two species. *Pteropus* guano had a higher C to N ratio, and *Desmodus* guano had higher N to P and C to P ratios than the other two species. These differences in guano composition suggest that guano from bats in different feeding guilds may affect ecosystem structure and dynamics differently (Justin and Roark, 2007).

There is a loophole of specific studies on degradation of species specific bat guano, in general the most abundant organisms in soil that contribute to organic matter decomposition are bacteria and fungi. Marinkelle and Grose (1972) documented infectious pathogenic micro-organisms isolated from bats that affected man or domestic animals. These include *Salmonella* spp., *Spirocheaeta* spp. and *Leptospira* spp. Numerous micro-organisms like *Bartonella rochalima* and *Grahamella* spp. *etc.* are also reported from the bats which are considered harmless to man and domestic animals or reported as doubtful pathogens due to the fact they are apparently not potential pathogens. There are number of medically significant fungal species isolated from the excreta of bats *viz*. *Cryptococcus neoformans, C. laurentii*, *Histoplasmaca psulatum* and *Sporothrix schencki* (Reis and Mok, 1979; Takashi *et al*., 2005). Keeping in view the clinical, economical, and environmental significance of fungi, bacteria and minerals found in bat ejecta the present study was designed to ascertain microbial load and mineral composition of bolus and guano of *P. giganteus* roosting in urban areas of Lahore.

MATERIALS AND METHODS

Study area

Present study, extending from January, 2011 to December, 2011 was conducted in two public parks *i.e*., Jinnah garden (35°55' north latitude and 74°33' east longitudes) and Lalazar garden (31°28' north latitude and 74°14' east longitudes) in Lahore, the second most populated city of Pakistan. The city experiences extreme summer and winter seasons, the summer season is followed by rainy and humid monsoons season. The Jinnah garden covers an area of 176 acres (0.71 km^2) with Indian flying fox, *P. giganteus* population ranging from 3000 to 4000 individuals while at Lalazar garden which is smaller in size and stretched over an area of 04 acres (0.02 km^2), the populations ranges from 800 to 1000 individuals. Both the public parks are permanent open day roosts of the Indian flying foxes (*P. giganteus*) and governed by Pakistan Horticulture Authority (PHA).

Sampling strategy

The ejecta (bolus and guano) of Indian flying foxes were collected by spreading a polythene sheet of 1 m × 1 m (length × width) under the roosting sites of *P. giganteus*. Out of total 47 plots at Jinnah garden, the *P. giganteus* was roosting in four plots. Four polythene sheets, one in each plot were placed at Jinnah garden while one polythene sheet was spread under the roosting canopies at Lalazar garden once a month for the whole year. Each sheet remained spread for 10 h *i.e*. from 2000 h Pakistan Standard Time (PST) till 6000 h PST and was removed on the subsequent day. The ejecta were randomly collected and were placed in polythene bags along with the tags indicating garden, roost number, plot number and date (Mahmood-ul-Hassan *et al*., 2010).

Twelve monthly samples of bolus and guano were lumped together into four seasonal samples. The February, March and April samples were named as spring sample. All the remaining monthly samples were also lumped together in the same way and designated as summer (May, June, July), autumn (August, September, October), and winter (November, December, January) samples, respectively. From each of the three monthly samples, 333.3 mg of bolus and guano was used for microbial analysis in such a way that the combined seasonal sample weighed 1 g. The pH of each seasonal sample was observed on pH meter (Mahmood-ul-Hassan *et al*., 2010).

Fungal analysis

Fumigated incubators, sterilized glass-wares and autoclaved apparatus were used to prevent environmental contamination. One gram of sample was transferred in 10 ml (v/w) normal saline solution to prepare five concentrations of serial dilution ranging from 10^{-1} up to 10^{-5} which were used for further identification of fungi present in the bolus and guano samples.

Sabouraud dextrose agar (16.25 g) and agar agar (5 g) media were diluted with 250 ml of distilled water, shaken, boiled, auto-claved for 45 min and poured in Petri dishes. 0.1ml of each dilution was spread on media in Petri dishes and left for incubation at room temperature for 72 h. The fungal growth was then monitored and were counted manually, petri plate with more than or equal to four fugal colonies were processed further for purification and identification while the others were neglected.

Fungal colonies were purified by picking the growth with platinum loop from each different type of colony and were placed in separate petri plates and again incubated for 72 h. The slides were then prepared from each purified colony and stained with Congo red and sealed with DPX. These permanent fixed slides were then observed under microscope (ML 5100) for identification. Macroscopic and microscopic characters of colonies were observed and genera were identified following Emmons *et al*. (1977).

Bacterial analysis

Seven grams of nutrient agar and 0.25 g agar agar was diluted with 250 ml of distilled water and shaken well, autoclaved for 45 min and were poured in 8 Petri plates. 0.1 ml of each dilution of the bolus and guano was spread separately on media in Petri plates and were left for incubation for 24 h at room temperature. The bacterial growth was then checked and bacterial colonies were counted on bacterial counter (Keunzahlgerat BZG 28). Those Petri plates in which bacterial colonies ranged from 30 to 300 were further processed for purification and identification while the remaining were neglected. Bacterial colonies were purified by picking the growth with platinum loop from each different type of colony and were three way streaked in separate Petri plates and again incubated for 24 h. The slides were then prepared from each purified colony, stained by gram staining method and were observed under microscope (ML 5100) for identification (Aaronson, 1970; Cruickshank *et al*., 1975).

Mineral composition

Each lumped seasonal sample (10 g) of both guano and bolus was incubated at room temperature in a pan for 24 h. The samples were analyzed after incubation to determine the level of different minerals present in them. The samples were weighed and dried in an oven at 105°C for 24 h and placed in separate plates. A small part of the dry matter (5 g) was ashed at 650°C for 4 h in respected crucibles (of each season) in an electric furnace. 10 ml of nitric acid was added in each of these 5 g ashed samples then the flasks were kept in water bath at 65°C for 15 min. Perchloric acid ($HClO_4$), (5 ml) was then added to the flask and kept in water bath at 75°C for 15 min. The samples were then dried on hot plate till the fluid reached to 0.5 ml. After proper filtration, the samples were raised to 50 ml solution by repeated washing (Elaroussi *et al*., 1994). The mineral composition was estimated using atomic absorption spectrophotometer.

RESULTS AND DISCUSSION

During present study, a total of twelve fungal genera representing nine families were isolated from bolus and guano samples of *P. giganteus*. These genera included *Alternaria*, *Aspergillus, Candida, Chrysosporium, Cryptococcus, Exophiala, Fusarium, Histoplasma, Penicillium, Saccharomyces, Scopulariopsis* and *Trichophyton*. Nine fungal genera were isolated from bolus and eight from the guano of Indian flying fox. Genus *Aspergillus* (n=7) was most recorded while *Histoplasma* (n=1) and *Trichophyton* (n=1) were least recorded genera. Four of the isolated genera *viz*. *Candida, Fusarium, Penicillium* and *Saccharomyces* were isolated only from bolus of the Indian flying foxes whereas *Cryptococcus*, *Histoplasma* and *Trichophyton* were observed from isolated from guano samples only (Table I). Goveas *et al*. (2006) observed that *P. giganteus* bolus samples contain more fungi while the guano. Takashi *et al*. (2005) reported *Candida lusitaniae* and *Debaryomyces hansenii* from bat guano.

Seasonal variations had been observed among the nine fungal genera isolated from bolus samples of the Indian flying fox during all the four seasons *viz*. spring, summer, autumn and winter. *Alternaria* and *Chrysosporium* were the genera isolated only in spring, *Scopulariopsis* was isolated only in autumn whereas rest of the genera were isolated in two seasons as *Candida* and *Fusarium*in spring and autumn, *Penicillium* in spring and winter, *Exopphiala* in summer and autumn and *Saccharomyces* was isolated in summer and winter season. *Aspergillus* was the only genus recorded in all of the four seasons. Seelan *et al*. (2008) observed 23 species of bats out of which 13 (56.5%) species were found to contain 17 fungal isolates of the genus *Aspergillus*. Maximum numbers of fungal colonies (6.0 × 105 cfu/gm) from bolus were counted in spring season while minimum number of colonies (4.0×10^3cfu/gm) was counted in summer season (Table II). Goveas *et al*. (2006)

Table I.- Macroscopic and microscopic characters of fungal genera extracted from ejecta of *Pteropus giganteus* roosting at Jinnah and Lalazar gardens, Lahore.

Genera	Macroscopic features	Microscopic features	Source	No. of isolates
Alternaria	Grows rapidly and the colony size reaches a diameter of 3 to 9 cm. The colony was flat, downy to woolly and covered by grayish, short, aerial hyphae in time. The surface, greyish white at the beginning which later darkens and becomes greenish black or olive brown with a light border. The reverse side, typically brown to black due to pigment production.	Septate, brown hyphae. Conidiophores septate and brown in color, occasionally producing a zigzag appearance, bearing simple or branched large conidia (7-10 x 23-34 µm) which had both transverse and longitudinal septations. Conodia, ovoid to obclavate, darkly pigmented, muriform, and smooth or roughened. The end of the conidium nearest the conidiophore is round while it tapers towards the apex. This gives the typical beak or club-like appearance of the conidia.	Bolus and guano	4
Aspergillus	Colonies are downy to powdery in texture. The surface initially white, quickly becoming black with conidial production. Reverse is pale yellow and growth may produce radial fissures in the agar.	Hyphae septate and hyaline. Conidial heads radiate initially, splitting into columns at maturity. Conidia brown to black, very rough, globose, and measure 4-5 µm in diameter**.** Conidiophores are long (400-3000 µm), smooth, and hyaline, becoming darker at the apex and terminating in a globose vesicle (30-75 µm in diameter). Metulae and phialides cover the entire vesicle.	Bolus and guano	7
Candida	The colonies are cream colored, grow rapidly and mature in 3 days. The textures of the colonies are pasty and glistening.	Abundant, branched pseudohyphae and true hyphae with blastoconidia are present. The blastoconidia are formed in grape-like clusters along the length of the hyphae.	Bolus	2
Chrysosporium	Colonies grow moderately rapidly at 25°C. The morphology of the colonies was variable, granular, woolly, cottony and flat, or raised and folded in appearance. From the front, the color was white cream, yellow to tan to pale brown. The reverse color white to brown.	Hyphae septate while the conidia hyaline, broad-based, one-celled, and smooth- or rough-walled. Conidia are broader than the vegetative hyphae and occur terminally on pedicels, along the sides of the hyphae, or in intercalary positions. Arthroconidia, on the other hand, abundant and larger than their parent hyphae in diameter.	Bolus and guano	3
Cryptococcus	Colonies, fast growing, soft, glistening, usually mucoid, and cream to beige in color.	Produced round, budding yeast cells. No true hyphae and pseudohyphae are absent.	Guano	2
Exophiala	Initially yeast-like, moist, and brownish to greenish black in color, becomes velvety due to development of short, aerial grayish hyphae. The front color, olivaceous-black and the reverse was black in mature colonies	Septate hyphae which bear conidiogenous cells (annelides). The annelids, tubular and rocket-shaped and typically taper to form a narrow elongated tip. Ellipsoidal, conidia (1-3x3-6 µm) produced from the annelides.	Bolus and guano	3
Fusarium	Grow rapidly on Sabouraud dextrose agar at 25°C and produce woolly, cottony, flat, spreading colonies. The front color of the colony, cream to yellow orange. The reverse side colorless.	Hyphae are septate and hyaline. Conidiophores are simple, Macroconidia are moderately curved, stout, thick-walled, usually 3-5 septate and are borne on short conidiophores that soon form sporodochia. Microconidia are borne from long monophialides, are one to three-celled and occur in false heads only. Chlamydoconidia are present.	Bolus	2
Histoplasma	Thermally dimorphic fungus, colony was creamy, slow growing moist and yeast-like. The color was white initially and became buff brown with age. From the reverse, a yellow to yellowish orange in color.	Narrow-based, ovoid, budding yeast cells were formed	Guano	1
Penicillium	The colonies of *Penicillium* are rapid growing, flat, filamentous and velvety in texture. The colonies were initially white and become blue green with time. The plate reverse, pale to yellowish	Septate hyaline hyphae, branched conidiophores, metulae, phialides, and conidia were observed. Phialides were typical, formed brush-like clusters. The conidia were round, unicellular, and visualized as unbranching chains at the tips of the phialides.	Bolus	2
Saccharomyces	Colonies grow rapidly flat, smooth, moist, glistening, and cream to tannish cream in color.	Blastoconidia was observed, unicellular, globose, and ellipsoid to elongate in shape. Multilateral (multipolar) budding is typical. Hyphae absent.	Bolus	2
Scopulariopsis	Colonies grow moderately rapidly and matured within 5 days, granular in texture. The front color, white initially and became light brown tan with time. Reverse color, tan with brownish center.	Septate, hyphae, conidiophores, annelides, and conidia were observed. Conidiophoreswas hyphae-like and branched. Conidia are one-celled, globose to pyriform, smooth, but more commonly rough-walled, spiny, truncate, and forming basipetal chains.	Bolus and guano	2
Trichophyton	The growth rate was slow to moderately rapid. Texture was waxy, the front color was white to bright yellowish beige. Reverse was pale yellowish brown.	Septate, hyaline hyphae, conidiophores, microconidia, macroconidia, and arthroconidia were observed. Conidiophores poorly differentiated from the hyphae. Miroconidia (microaleuriconidia), one-celled and round. Macroconidia (macroaleuriconidia), multicellular (2- or more-celled), smooth, thin- or thick-walled, cylindrical and clavate.	Guano	1

documented *Fusarium* and *Penicillium* as common fungal genera in bolus and guano of Indian flying fox.

Seasonal variations were also noted regarding occurrence of fungal genera in guano samples of *P. giganteus*. A total of eight genera were isolated from guano during all the four seasons. Out of these, *Exophiala* and *Histoplasma* were isolated during spring season only; *Scopulariopsis* was isolated during summer and *Trichophyton* during autumn only. *Chrysosporium* and *Cryptococcus* were isolated in summer and winter season while *Alternaria* and *Aspergillus* were recorded in spring, autumn and winter. No genus was recorded in all the four seasons. Maximum number of fungal colonies (4.0×10^4cfu/gm) from guano were counted in spring while minimum number of colonies (3.0×10^4cfu/gm) were counted in autumn season (Table II). Seelan *et al*. (2008) documented the diversity of *Aspergillus* species by isolating six species of *Aspergillus* from 13 species of bats. The abundance and diversity of isolated fungal genera is also correlated with food sources and the roosting site of the bats. Yamamoto *et al*. (1995) investigated that the bat guano may mediate the exchange of pathogenic fungi just as pigeon excreta mediate the exchange of *Cryptococcus neoformans*, the causative agent of cryptococcosis. Apart from that, fruits consumed by these frugivore bats are also important factor in understanding the ecology of bats. Sometimes the infected fruit may contain pathogenic micro-organisms that may be present during the fruit decay process (Sepiah, 1985). So, this would be a key factor how the fungi are transmitted to bats since frugivorous bats consume fruits as their main diet.

Table I describes macroscopic and microscopic characters of all the twelve genera. Twelve bacterial genera namely *Acaligens*, *Azotobacter*, *Bacillus, Bartonella, Corynebacterium, Klebsiella, Listeria, Nitrsomonas, Nocardia, Paeudomonas, Salmonella* and *Streptomycete* were isolated from ejecta of *P. giganteus. Bacillus* was the only bacterial genus recorded in all the seasons from ejecta while *Azotobater, Bartonella*, *Nitrosomonas* and *Sallmonella* were recorded once a year. Six out of ten genera isolated from only guano included *Acaligens*, *Azotobacter*, *Bartonella, Nitrsomonas, Paeudomonas* and *Salmonella* whereas two genera *Klebsiella,* and *Nocardia* were isolated from bolus samples only. Four genera *Bacillus,Corynebacterium, Listeria* and *Streptomycete* were represented in both bolus and guano samples of Indian flying fox (Table II). The bacteriological examination of bolus and guano of the Indian flying fox done by Goveas *et al*. (2006) revealed the presence of *Alcaligenes* and *Pseudomonas* in guano, and *Bacillus*, *Klebsiella* and *Proteus* in bolus. Among actinomycetes, *Streptomycetes* were common in guano and *Micromonospora* in bolus.

Mineral composition of bolus of Indian flying fox was analyzed in four seasonal samples. The pH of fruit bat bolus is near acidic to neutral ranges between 6.7 and 7.4. The most abundant elements in bolus are phosphorus and nitrogen whereas potassium is less abundant. The total nitrogen ranges between 2.28% and 4.10% in bolus which are higher than that in guano. Total phosphorus ranges between 3.50% and 5.0% and total potassium ranges between 0.6% and 0.74% (Table II).

pH of bat guano varied from 7.1 to 7.4 with nitrogen and phosphorous contents ranging from 2% to 3.30% and 3.10% to 5.20% respectively (Table II). The studies of Goveas *et al*. (2006) revealed higher nitrogen, phosphorus and potassium in the bolus than the guano (3.3:4.3:0.7 vs 2.6: 4.2:0.6) of the *P. giganteus*. Ruth *et al*. (2004) documented that nitrogen and phosphorus were the most abundant elements in bat guano. The total nitrogen ranges between 8–12% and P_2O_5 ranges between 2–7%. Other elements include calcium, magnesium, potassium, aluminum, iron and sulphur that are present in quantities lower than 5% each.

In this study, the NPK values, fungal and bacterial load in guano and bolus of the Indian flying foxes were analyzed. The result showed higher nitrogen percentage in bolus than guano whereas phosphorus and potassium percentages were higher in guano of these bats. Nitrogen in guano is known to enhance crop growth while phosphorus provokes root development, shoots, budding, multiple branches and flowering in plants. Like other bat guano studies the present study clearly indicated that ejecta of the Indian flying foxes could be significantly used with appropriate ratios with soil to increase the growth, dry matter and productivity of plant and crops.

The fungal and bacterial analyses brought forward the presence of some useful and some pathogenic genera in bolus and guano of the Indian flying fox. Among 12 fungal genera *Aspergillus, Penicillium* and *Saccharomyces* are used in drug production, food making and fermentation. The maximum species of *Candida, Cryptococcus, Fusarium*and *Scopulariopsis* are harmless and are useful soil microbes. While the genera *Alternaria, Chrysosporium, Exophila, Histoplasma* and *Trichophyton* are infectious and pathogenic either to humans, animals or plants. Among bacterial genera, *Alcaligenes, Azotobacter, Nitrosomonas*and *Corynebacterium*have economical and industrial significance and are also important in Nitrogen-cycle and N_2 fixative bacteria in environment. Some members of *Streptomycetes* are pathogens while two third of them are important in medicine production and also plays important role in decaying vegetation. *Nocardia*

Table II.- Seasonal variations in occurrence of various fungal and bacterial genera from bolus and guano samples of Indian flying fox (*Pteropus giganteus*) roosting at Jinnah and Lalazar garden, Lahore.

Season	Fungal cfu/gm	Fungal Genera	Bacteria cfu/gm	Bacterial Genera	Mineral Composition			Fungal cfu/gm	Fungal Genera	Bacteria cfu/gm	Bacterial Genera	Mineral Composition		
		BOLUS			N%	P%	K%		GUANO			N%	P%	K%
Spring	6.0×10^5	*Alternaria* *Aspergillus* *Candida* *Chrysosporium* *Fusarium* *Penicillium*	7.6×10^5	*Bacillus* *Corynebacterium*	4.10	4.60	0.60	4.0×10^4	*Alternaria* *Aspergillus* *Exophiala* *Histoplasma*	6.7×10^6	*Alcaligenes* *Azotobacter* *Bacillus* *Corynebacterium*	2.50	4.10	0.60
Summer	4.0×10^3	*Aspergillus* *Exophiala* *Saccharomyces*	3.5×10^5	*Bacillus* *Listeria* *Nocardia*	3.36	3.50	0.66	3.0×10^7	*Chrysosporium* *Cryptococcus* *Scopulariopsis*	8.9×10^4	*Alcaligenes* *Bacillus* *Listeria* *Nitrosomonas* *Salmonella* *Streptomycete*	3.10	3.10	0.56
Autumn	5.0×10^5	*Aspergillus* *Candida* *Exophiala* *Fusarium* *Scopulariopsis*	8.0×10^7	*Bacillus* *Corynebacterium* *Klebsiella* *Listeria* *Nocardia* *Streptomycete*	2.28	4.90	0.70		*Alternaria* *Aspergillus* *Trichophyton*	4.0×10^4	*Bacillus* *Bartonella* *Pseudomonas* *Streptomycete*	2.00	4.90	0.64
Winter	4.0×104	*Aspergillus* *Penicillium* *Saccharomyces*	7.6×105	*Bacillus* *Corynebacterium* *Klebsiella*	2.28	4.90	0.70	3.0×107	*Alternaria* *Aspergillus* *Chrysosporium* *Cryptococcus*	5.6 × 106	*Alcaligenes* *Bacillus* *Corynebacterium* *Pseudomonas*	3.30	5.20	0.70

species constitute major useful oral micoflora while some are pathogens. Species of *Bartonella, Klebsiella, Listeria* and *Salmonella* are infectious and pathogens. The members of *Pseudomonas* are important bio-control and bioremediation agents whereas some species are pathogenic.

CONCLUSION

It can be concluded from the present study that ejecta of the *Pteropus giganteus* are composed of beneficial and pathogenic microbes. However, it might be useful in enhancing fertility of the soil.

Statement of conflict of interest

Authors have declared no conflict of interest.

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