Distribution and periodicity of sandflies (Diptera: Phlebotominae) along different altitudes in Asir Region, Southwest of Saudi Arabia

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Abstract

Asir Region in the southwest of Saudi Arabia has been a subject for expansion of agricultural projects, urbanization, which presumably have impact on distribution of phlebotomine sandflies. Few reports are available on sandflies in this region which is an important focus of cutaneous leishmaniasis. Therefore, this study aimed at updating the species composition, distribution and periodical fluctuation of sandflies in this region. Specimens were monthly collected by the Center for Disease Control light traps for one year in four localities representing different altitudes. In five other, collections were twice during the year period. Ten species (six *Phlebotomus* and four *Sergentomyia*) were identified, of which *P. arabicus* (32%) was the most common followed by *P. bergeroti* (29%) and *P. sergenti* (15%). Of the reported species, *S. palestinensis* is considered a new record from Asir. Sandflies were more common and maximum biodiversity was observed in lowlands and not in high altitudes. At different altitudes, the two commonest species were more active during spring. Sandfly density (sandfly/trap) was directly related to temperature and inversely related to altitude, relative humidity (RH) and wind velocity ($P<0.05$). To sum up, the distribution and abundance of sandflies in Asir are influenced by a combination of different factors: temperature, RH, wind velocity and altitude.

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Introduction

Phlebotomine sandflies (Diptera: Psychodidae) are widespread in the tropics and subtropics (Lane, 1993). They include many potential vectors for both cutaneous leishmaniasis (CL) and visceral leishmaniasis (VL). CL is an endemic disease in many areas of Saudi Arabia including the Western Provinces (Al-Qurashi *et al*., 2000; El-Badry *et al*., 2008). The disease is more common on the foothills and high plateau of Asir Region in the southwest of the Kingdom (Al-Zahrani *et al*., 1989; Abdelwahab & Abdoon, 2005). VL infections also recovered from the south-west of the Kingdom (Al-Zahrani *et al*., 1988a; Ibrahim *et al*., 1995; Al-Jaser, 2006).

Several reports are available on the distribution of the sandfly fauna in several regions of Saudi Arabia (Lewis & Büttiker, 1980, 1982; Killick-Kendrick *et al.*, 1985; Morsy & Al Seghayer, 1992; Abu-Zinada, 1999; Aldawood *et al*., 2004; Al Barrak, 2005; El-Badry *et al*., 2009; Doha & Samy, 2010) revealing the presence of 25 species with the dominance of *P. papatasi* (Mustafa *et al*., 1994). In only three occasions (Lewis & Büttiker, 1982; Abdelwahab & Abdoon, 2005; Alahmed *et al*., 2010), the sandflies were surveyed in Asir with 22 species identified.

During the past few decades, Saudi Arabia has viewed tremendous efforts in social development and urbanization in all provinces, and insect fauna, particularly sandflies was affected. The expansion of agricultural projects, urbanization and development of water resources, lead to creation of new breeding sites for sandflies (Alahmed *et al*., 2010).

The Asir area has also been a subject for various development and tourist projects as well as resettlement programs which presumably have impact on distribution of sandflies. However, few reports (Lewis & Büttiker, 1982; Abdelwahab & Abdoon, 2005; Alahmed *et al*., 2010) described sandflies in Asir region although CL has long been recognized as an important public health problem in this area (Al-Zahrani *et al*., 1988b). Therefore, this study aimed at identifying and updating the sandfly species composition, their geographical distribution, periodical abundance and species diversity in some localities representing different altitudes in Asir Region as one of CL-endemic areas in Saudi Arabia. The study could be important for implementing any large-scale control project.

Materials and methods

Study area

Asir Region (19°00′N, 42°00′E to 19°00′N, 43°00′′E) in the south-

west of Saudi Arabia has an area of 81,000 km² and a population of 1,913,392 (2010 Census). It is a mountainous area divided into 3 distinct topographical zones: i) Sarawat Asir which is a mountain range extending north-south along the coastal plains of the Red Sea and that rise to almost 3000 m at Jebel Sawdah near Abha *the capital*; ii) Asir Plateau and iii) Tehamah Plain (Tehama) which is a narrow sandy coastal strip of lowlands at sea level. The Region receives more rainfall than the rest of the country and falling in two seasons: spring (March and April) and summer (June to August). Temperature in Asir highlands is generally lower than in the other part of the Region and the rest of the kingdom as well. The coastal plain zone is generally characterized by lower rainfall, high temperature and relative humidity (RH). The study included nine localities representing different altitudes.

Collection of sandflies

Sandfly collections were carried out monthly (June 2009 to May 2010) in Abha, Bishah, Muhayil and Tanomah (Al Namas), and twice during the year period in the Sarat Abidah, Rejal Almaa, Balqarn, Al Bark and Tathleth. In each locality, 2-3 collection sites were selected to represent different habitats: wadies (valleys), farms, dumpsites and sheep raising farms. Sandflies were collected by the Center for Disease Control (CDC) miniature light traps (John W Hock Co, Gainesville, FL, USA). The traps (3 traps/site/night) were set before sunset and collected after sunrise next morning. Collected phlebotomines were preserved in 70% alcohol, cleaned in chloral hydrate: phenol (1:1 vol/vol) and then mounted in Puri's medium for identification using different keys (Lewis, 1982; Lane, 1986; Kakarsulemankhel, 2009, 2010). Along with fly collections, the elevation of the collection site above sea level, the weather temperature, RH and wind velocity (km/h) were recorded.

Periodical abundance

The periodic abundance of the common *Phlebotomus* spp. was examined in localities representing lowlands, moderately altitude areas and highlands.

Statistical analysis

The relations of sandfly density (No/trap) to the locality altitude and weather conditions (temperature, RH and wind velocity) were examined respectively by simple and multiple regression analysis. The slopes of the regression equations were tested for deviation from 0 by t-test. For the four localities that were monthly surveyed during the year period, the diversity of the sandfly based on the Simpson (1-D) and Shannon (H) indices was examined. The Paleontological statistics (Past) software ver. 2.08 was used for data analysis (Hammer *et al.,* 2001).

Results

Species composition and relative abundance

A total of 972 sandflies (454 males and 518 females) of two genera were collected: *Phlebotomus* which was more common (621 fly: 63.89%) and *Sergentomyia* (351 fly: 36.11%). Six hundred forty flies (246 males and 394 female) were identified into 10 species (Table 1): 6 *Phlebotomus* and 4 *Sergentomyia*. *Phlebotomus arabicus* was the most common species (32.19%) followed by *P. bergeroti* (29.38%), *P. sergenti* (15.00%), *P. orientalis* (11.09%), *P. papatasi* (5.94%), *S. clydei* (3.44%), and *P. alexandri* (2.03%). *S. africana, S. palestinensis* and *S. christophersi* were rare (0.31% each).

Based on the present and previous surveys a complete list of sandfly species (8 *Phlebotomus* spp. and 15 *Sergentomyia* spp.) reported in Asir is presented in Table 2.

Geographical distribution

The species composition varied among the surveyed localities (Figure 1). The reported species were encountered in all altitudes, except *S. christophersi* and *S. africana* which were reported only in low lands (<500 m) and *P. alexandri* and *S. palestinensis* which were collected only from highlands (>2000 m). Sandflies were more common (8 species) and more abundant in lowlands (48.10% of collected flies) than in moderately altitude areas (27.99%) or in high lands (23.9%) (Table 3). Regression analysis indicated that fly density (fly/trap) was inversely related to the altitude (b=–0.003, P<0.05).

Species diversity

The diversity for the sandfly sampled monthly in the four localities was examined. The results revealed maximum diversity in Muhayil (low altitude) with the highest Simpson index (1-D=0.69) and Shannon index (H=1.48). On the other hand, Abha (high altitude) represented the site with the minimum diversity indices (1-D=0.59 and H=1.10). The rest of the localities exhibited medium biodiversity indices (Figure 2).

Effect of weather conditions

Regression analysis indicated that the fly density (fly/trap) was directly related to temperature (b=0.413, P<0.01) and inversely related to RH (b=–0.002, P<0.05) and wind velocity (b=–0.170, P<0.05).

Periodic abundance

The seasonal activity patterns of the two common species (*P. arabicus* and *P. bergeroti*) across the different altitudes were examined (Figures 3

Table 1. Relative abundance of reported sandfly species in Asir Region.

Genus	Species	No.	%
Phlebotomus	*P. (Phlebotomus) bergeroti* Parrot	188	29.38
	P. (Phlebotomus) papatasi (Scopoli)	38	5.94
	P. (Paraphlebotomus) alexandri Sinton	13	2.03
	P. (Paraphlebotomus) sergenti Parrot	96	15.00
	P. (Adlerius) arabicus Theodor	206	32.19
	P. (Larroussius) orientalis Parrot	71	11.09
Sergentomyia	*S. (Parrotomyia) africana* (Newstead)	2	0.31
	S. (Parrotomyia) palestinensis (Adler & Theodor)	2	0.31
	S. (Sintonius) christophersi (Sinton)	2	0.31
	S. (Sintonius) clydei (Sinton)	22	3.44
	Total No. (Male/Female)	640 (246/394)	
	Sex ratio M:F	1:1.60	

and 4). Generally, sandflies were more active during spring months (March to May; mean temperature=30°C, RH=24%, wind=6 km/h) with moderate activity during summer (June to August; mean temperature=36°C, RH=35%, wind=7 km/h) and autumn months (September to November; mean temperature=29°C, RH=33%, wind=9 km/h) and very low density or absent during winter months (December to February; mean temperature=20°C, RH=47%, wind=14 km/h).

Discussion and conclusions

The present study is a report of the results of an entomological survey of sandflies in Asir region.

During the study, 10 sandfly species were identified: 6 belong to *genus Phlebotomus* and 4 to genus *Sergentomyia*. *Phlebotomus* spp. were dominating (*ca* 96%), which is a Palaearctic feature (Lewis & Büttiker, 1980), its distribution is in agreement with the previous observation (Alahmed *et al.*, 2010). All the encountered species in this study were reported earlier from Asir region (Lewis & Büttiker, 1982; Abdelwahab & Abdoon 2005; Alahmed *et al*., 2010) except *S. palestinensis*, which is considered a new record from this region, and it occurs only at high altitude (Tanomah). The species was previously reported from Al Baha, Makkah, Al Madinah and Jizan (Lewis & Büttiker, 1982).

Of the identified *Phlebotomus* species (612 fly), *P. arabicus* (*ca* 34%) and *P. bergeroti* (*ca* 31%) were the commonest species followed by *P. sergenti, P. orientalis*, *P. papatasi* and *P. alexandri*. *P. bergeroti* was reported as a rare species in Saudi Arabia (Nadim *et al.,* 1979; Büttiker *et al.,* 1982), however, Abdelwahab & Abdoon (2005) and Alahmed *et al.* (2010) indicated that in Asir, *P bergeroti* is the most abundant species while *P. sergenti* is second in abundance. In Al Baha, *P. bergeroti* is also the most abundant constituting 41.7% of the collected flies (Doha & Samy, 2010). *Phlebotomus alexandri* revealed also very low density (Abdelwahab & Abdoon, 2005; Alahmed *et al.,* 2010). *Phlebotomus ara-*

Table 2. A list of sandfly species reported in Asir Region during the present (1) and previous surveys (2. Lewis & Büttiker, 1982; 3. Abdelwahab & Abdoon, 2005; 4. Alahmed *et al.*, 2010).

Species	Source			
	1	2	3	4
Phlebotomus (Phlebotomus) papatasi (Scopoli)	X	X	X	X
P. (Phlebotomus) bergeroti Parrot	X	X	X	X
P. (Paraphlebotomus) alexandri Sinton	X	X	X	X
P. (Paraphlebotomus) kazeruni Theodor & Mesghali		X		
P. (Paraphlebotomus) saevus Parrot & Martin		X		
P. (Paraphlebotomus) sergenti Parrot	X		X	X
P. (Larroussius) orientalis Parrot	X	X	X	X
P. (Adlerius) arabicus Theodor	X	X	X	X
Sergentomyia (Sergentomyia) antennata (Newstead)		X		X
S. (Sergentomyia) fallax (Parrot)		X		X
S. (Sergentomyia) taizi Lewis		X		
S. (Sergentomyia) schwetzi (Adler, Theodor & Parrot)				X
S. (Sintonius) tiberiadis (Adler, Theodor & Lourie)		X		X
S. (Sintonius) adleri (Theodor)		X		X
S. (Sintonius) calcarata (Parrot)		X		X
S. (Sintonius) christophersi (Sinton)	X	X		X
S. (Sintonius) clydei (Sinton)	X	X		X
S. (Parrotomyia) palestinensis (Adler & Theodor)	X			
S. (Parrotomyia) africana (Newstead)	X	X		X
S. (Parrotomyia) magna (Sinton)				X
S. (Grassomyia) dreyfussi Parrot		X		X
S. (Grassomyia) squamipleuris (Newstead)				X
S. (Neophlebotomus) sonyae Lewis				X

Table 3. Distribution of sandfly species in different altitudes of the monthly surveyed localities.

	Lowlands <500 m	Moderate altitude >1000 m	Highlands >2000 m	
Locality	Muhayil	Bishah	Tanomah	Abha
No. of species	8	6	6	4
Collected flies: No. (%)	165 (48.10)	96 (27.99)	34 (9.91)	48 (13.99)

bicus is rare (Abdelwahab & Abdoon, 2005), in contrast to our findings and that of Alahmed *et al*. (2010). *Phlebotomus orientalis* was considered rare (Abdelwahab & Abdoon, 2005), but represented 5.85% of the collected flies in Abha (Alahmed *et al.,* 2010) and 12% in the present study. Among the *Sergentomyia* species (28 sandflies), *S. clydei* was dominating (*ca* 79%), while *S. christophersi*, *S. africana* and *S. palestinensis* each represented about 7%, numbers similar to previous findings. *S. christophersi, S. clydei, S. africana* numbered 37, 31 and 24, respectively out of 558 collected sandflies (Alahmed *et al.,* 2010).

Several investigators (Büttiker *et al.,* 1982; Lewis & Büttiker, 1982; Aldawood *et al.,* 2004; Al Barrak, 2005; El-Badry *et al.,* 2008) reported *P. papatasi* as a dominant species in Saudi Arabia. However in this study, *P. papatasi* was not common and represented only about 6% of the collected sandflies. Only 13 specimens (2.38%) (Alahmed *et al.,* 2010) and 19 specimens (0.95%) (Lewis & Büttiker, 1980) of *P. papatasi* were previously collected in Asir. This confirms the marked affinity of sandfly fauna of Asir (Abdelwahab & Abdoon, 2005) as in southern Sinai, Egypt (El Sawaf *et al.,* 1987) where *P. bergeroti* and *P. sergenti* constituted the highest percentage of collected sandflies whereas *P. papatasi* was a poorly represented species.

Sandflies were more common and abundant in lowlands respect to higher altitudes. The lowest abundance was observed in highlands in agreement with the previous observation (Büttiker *et al.,* 1982). In Asir region (Abdelwahab & Abdoon, 2005), the highest abundance of *Phlebotomus* sandflies was found in Tehamah foothills in lowland and in the coastal plain while the lowest fly densities were reported at higher altitudes in Sarawat Mountains and Asir plateau. This was confirmed in the present study by regression analysis, which indicated that the density of flies inversely related to the altitude ($P<0.05$). Meanwhile, the results revealed maximum diversity indices in Muhayil (lowland) and minimum ones in Abha (highland) due to the low richness of the species in this locality (n=4 spp). This may indicate that the lowlands are the most favourable sites for the breeding and activity of sandflies. Lower fly abundance at higher altitudes may be due to low temperature (annual mean=21.6°C), fog and strong winds (annual mean=11.92 km/h) in comparison with lowlands (annual mean of temp=32.03°C and of wind velocity=6.43 km/h)

It was reported (Abdelwahab & Abdoon, 2005) that the species composition of the sandflies is not affected by altitude, although it was found (Büttiker *et al.,* 1982) that the species spectrum shows a tendency for more species to occur at higher altitudes than in the Tehamah districts (lowlands). Moreover, in Sinai, Egypt (El Sawaf *et al.,* 1987), a remarkable difference in sandfly species composition at different altitudes was observed. The present study indicated that although most sandfly species were encountered in all altitudes, some species were found restricted to certain altitudes: *S. christophersi* and *S. africana* in lowlands and *P. alexandri* and *S. palestinensis* in highlands. This agrees with previous research (Büttiker *et al.,* 1982) that is *P. papatasi* and *P. bergeroti* prefer lower altitudes whereas *P. orientalis* and *P. arabicus* are species of higher altitudes.

The knowledge of the seasonal activity of sandflies is of importance in predicting the period of maximum risk of *Leishmania* transmission and for carrying out an effective control program. Results indicated that in different altitudes, *Phlebotomus* flies were more active during spring, with moderate activity during summer and autumn and very low density or absent during winter seasons. Almost similar observations

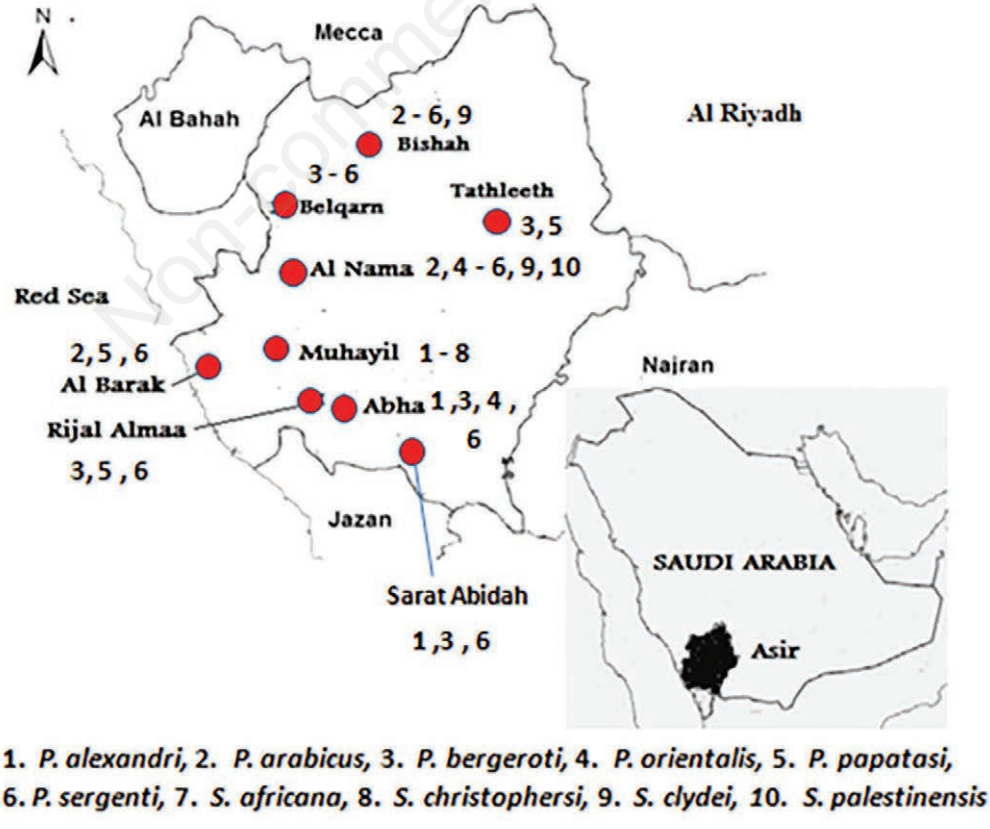

Figure 1. The surveyed localities and sandfly species distribution in Asir Region.

were previously reported (Abdelwahab & Abdoon, 2005) where the highest abundance of sandflies was recorded during the spring and summer (March to September) and the lowest fly abundance was throughout the period from November to February. Morsy *et al*. (1995) found that the greatest number of *P. papatasi* in Riyadh occurred most commonly during the summer season with two peaks in June and September. During the winter season no insects were found and then the population density started to appear again from March. Regression analysis indicated that fly density was directly related to the weather temperature ($P<0.01$) and inversely related to RH and wind velocity ($P<0.05$). Previous analysis (Abdelwahab & Abdoon, 2005) showed a significantly positive correlation between fly density and temperature and negative correlation with RH at Asir foothills.

Cutaneous leishmaniasis is an endemic disease in many areas of Saudi Arabia (Al-Qurashi, 2000; El Hassan, 2013) and is widespread in villagers of the Asir plateau (Al-Zahrani *et al*., 1988b). The disease is more common on the foothills and high plateau of Asir region (Al-Zahrani *et al*., 1989). Visceral Leishamniasis infections occur in Jizan (Al-Zahrani *et al*., 1988a; Ibrahim *et al*., 1995; Al-Jaser, 2006).

Of the reported sandfly species, several are implicated as vectors of leishmaniasis in Asir and other regions of the kingdom.

- *P. bergeroti* is assumed as a secondary vector of leishmaniasis in Asir mountain plateau (Büttiker *et al*., 1982) and as a suspected vector of *L. tropica* (anthroponotic cutaneous leishmaniasis) in Makka (Lewis, 1982) and in some African countries as well as in Egypt, Iran, Oman and Yemen (Maroli *et al*, 2013). Based on dominance of this species and the recorded increase in the number of leishmaniasis cases after few months of its peak of abundance (Abdelwahab & Abdoon, 2005) may highlight its role as a probable vector of *Leishmania* in Asir area. Recently, *P. bergeroti* was found positive for *Leishmania*-like flagellate infection in Al Baha (Doha & Samy, 2010). However, studies are needed to clarify the possible role of this highly abundant species in *Leishmania* transmission in Asir where this disease is widely spread;
- *P. papatasi* is the main vector of CL in many parts of Saudi Arabia (Lewis & Büttiker, 1982; Mondragon-Shem *et al*., 2015), may be the major vector in Al Qassim Region (Lewis & Büttiker, 1980; Al Barrak, 2005) and is a proven vector of *L. tropica* in many countries including Yemen, Iraq, Egypt, Algeria, Iran, Jordan, Morocco and Tunisia (Maroli *et al*., 2013). Moreover, *P. papatasi*, was found to be a proven vector of *L. major* (zoonotic cutaneous leishmaniasis) in the Sinai Peninsula, Egypt (Samy *et al*., 2014). Although, *P. papatasi* is the most widespread and dominant species in all investigated areas of Saudi Arabia (Mustafa *et al*., 1994; Alahmed *et al*., 2010), however, in the present study this species was not common (6% of collected sandflies). This may indicate that this species has little or no role in *Leishmania* transmission in Asir Region. This assumption agrees with what has been found in Kuwait where *P. papatasi* did not appear as an urban species and was not regarded as a possible vector of *leishmania* in that country (Abdelwahab & Abdoon, 2005);
- *P. sergenti* is a proven vector of *L. tropica* in the southern region of Saudi Arabia (Al-Zahrani *et al*., 1988b; Doha & Samy, 2010), and in several other countries (Maroli *et al*., 2013);
- *P. alexandri* and *P. orientalis* are suspected to be the main vectors of *L. donovani* (VL) in the southwestern part of Saudi Arabia (Al-Zahrani *et al*., 1997). The role of the two species in transmitting *L. donovani* is fully documented in other countries. *P. alexandri* is the main vector from North Africa to western China (Lane, 1993) while *P. orientalis* is a proven vector in Sudan (Maroli *et al*., 2013);
- *P. arabicus* was found naturally infected with *Leishmania*-like flagellates in Al Baha (Doha & Samy, 2010).

In conclusion, the present findings indicate that the distribution and abundance of sandflies in Asir Region are influenced by a combination of ecological and topographical factors (temperature, RH, wind velocity and altitude). The obtained results could be important for the successful implementation of leishmaniasis control programs. Among the reported species, *S. palestinensis* is considered a new record from Asir.

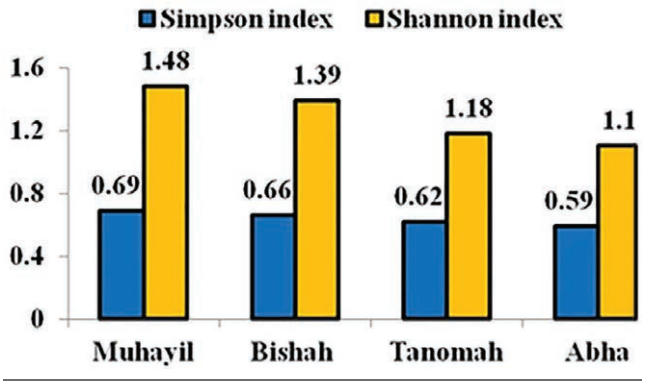

Figure 2. Sandfly diversity indices for monthly sampled localities.

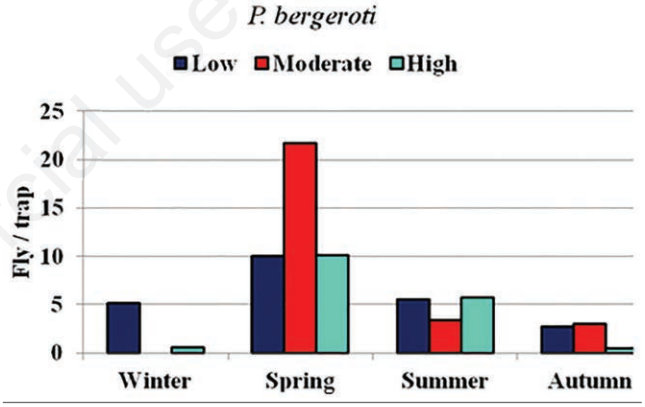

Figure 3. Periodical abundance of *Phebotomus bergeroti* at different altitudes (Low: Muhayil; Moderate: Bishah; High: Tanomah and Abha localities).

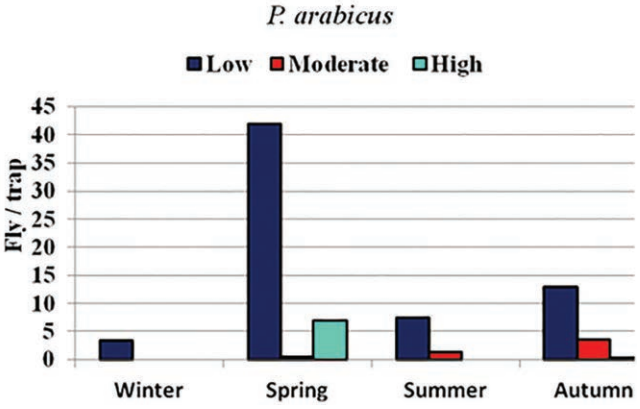

Figure 4. Periodical abundance of *Phlebotomus arabicus* at different altitudes (Low: Muhayil; Moderate: Bishah; High: Tanomah and Abha localities).

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