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## Taxonomic Morphology of *Phlebotomus (Paraphlebotomus) sergenti* Parrot (1917) and *Phlebotomus alexandri* Sinton (1928) (Diptera, Psychodidae) of Pakistan

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**Abstract:** During entomological surveys conducted by the author in the whole of the Balochistan Province in 1996-2001, *Phlebotomus sergenti* Parrot (1917) (N=140) and *Phlebotomus alexandri* (1928) (N=82) were collected from 11 and five localities, respectively. These localities appear to be the new record in the literature to date. Taxonomic characters previously remained un-described, unmeasured and unfigured are described, measured and illustrated in the present paper. Pakistanese specimens are compared with the published data of these species from other countries. Differential diagnosis of these species is also given. A key is constructed for the identification of these two Pakistanese species.

**Key words:** *Phlebotomus sergenti*, *Phlebotomus alexandri*, sandfly, taxonomic characters

### INTRODUCTION

*Phlebotomus sergenti*: Parrot<sup>[1]</sup> has been reported from Dera Ismail Khan by Newstead and Sinton<sup>[2]</sup> and from Quetta by Sinton<sup>[3]</sup>. Lewis<sup>[4]</sup> reported it from some parts of Pakistan and gave a very brief note "The cibarium of both sexes is very like that of *Ph. papatasi* but the teeth are smaller and less scattered. The labral crest of the male is like that of *Ph. alexandri*". Aslamkhan and Rafiq<sup>[5]</sup> reported it from Sibi and Lehri towns of Balochistan. Burney and Lari<sup>[6]</sup> in their paper mentioned "According to report of the Entomologist, R.F.M. College, Rawalpindi, sandflies were widely distributed in Balochistan all over Sibi to Khuzdar area and from Mastung to Quetta, Bostan, Zhob and beyond distributed in rural and urban areas. A total of 3450 specimens were collected and identified as *Ph. sergenti*, *Ph. papatasi*, *Sergentomyia squamipleuris*". Unfortunately locality wise species distribution was not given in the said paper. Aslamkhan<sup>[7]</sup> and Aslamkhan *et al.*<sup>[8,9]</sup> reported it from Barkhan and Kahan towns of Balochistan. However, none of the above authors did provide detailed description with illustration of taxonomic characters of Pakistanese *Phlebotomus sergenti*.

*Phlebotomus alexandri*: Sinton<sup>[10]</sup> (♂) were collected originally from Amara in Mesopotamia by Newstead<sup>[11]</sup> who described it under the name *Ph. sergenti* var. which resembled *Ph. sergenti* in the general morphology of the terminalia but differed in the shape of the style and length of antennal segments. The styles of these males were

oblong and oval and its antennal segments were short. He described these males as *Ph. sergenti* var. He observed the different shape of the style and the presence of one terminal spine and one sub terminal spine was markedly displaced towards the center and also the short 3<sup>rd</sup> antennal segment. The other character observed was the longer brush of hairs on the basal process of coxite. Sinton<sup>[10]</sup> collected similar males in Waziristan and noted that they are identical with the males from Iraq and named them *Ph. sergenti* var. *alexandri* after the Alexander the Great, because the boundaries of his empire were the same as the distribution of the new variety. Parrot<sup>[12]</sup> found var. *alexandri* in Algeria and observed that females of this variety differ considerably from those of *Ph. sergenti* and justify considering the variety as a species, this has been generally accepted. Theodor<sup>[13]</sup> compared the wing length of ♀ *Ph. alexandri* and *Ph. sergenti* and observed that the wings are shorter than the body in all forms of *Ph. alexandri* which differentiate it from females of *Ph. sergenti*. *Ph. alexandri* is an uncommon species with a discontinuous distribution in Pakistan and Lewis<sup>[4]</sup> reported it from a single male taken from Parkutta (Northern area of Pakistan) and gave measurements of antennal segment 3, labrum, genital pump and gave figure of labrum. However descriptions, measurements and figures of other taxonomic characters of ♀ and ♂ of *Ph. alexandri* from Pakistan were still unknown in the literature prior to the present study. Moreover, previously *Ph. alexandri* has never been recorded from Balochistan Province.

To fill the gap of knowledge, a taxonomic study was conducted by the author in the whole of Balochistan Province in 1996-2001 and 2013 sandflies were collected comprising of the genera *Phlebotomus*, *Sergentomyia* and *Grassomyia*<sup>[14]</sup>. In view of the insufficient description of Parrot, Sinton and Lewis (loc. cit.), *Phlebotomus sergenti* and *Phlebotomus alexandri* are re-described in the present paper. Morphology of taxonomic characters of these two species are measured and illustrated also.

## MATERIALS AND METHODS

For collection, preservation, dissection and examination of external and internal parts of the body of the sandfly, the conventional methods especially those used by Johnson *et al.*<sup>[15]</sup>, Lewis<sup>[16]</sup>, Killick-Kendrick<sup>[17]</sup>, Lawyer *et al.*<sup>[18]</sup> and Killick-Kendrick *et al.*<sup>[19]</sup> were generally followed. Standard taxonomic keys furnished by Lewis<sup>[4,20,21]</sup> and Artemiev<sup>[22]</sup> were followed. All the diagrams were drawn with a camera lucida and are to the given scales and the measurements are in millimeter (mm), unless otherwise indicated. All the specimens are housed with the Author's collection of Sandflies, Department of Zoology, University of Balochistan, Quetta, Pakistan.

## RESULTS

*Phlebotomus (Paraphlebotomus) sergenti* Parrot<sup>[1]</sup>.

*Phlebotomus sergenti*<sup>[1]</sup>, Bull. Soc. Path. Exot., 10: 564;<sup>[3]</sup>, Indian J. Med. Res., 11: 814;<sup>[10]</sup>, Indian J. Med. Res., 16: 307.

*Phlebotomus (Phlebotomus) sergenti*<sup>[23]</sup>, Archs. Inst. Pasteur Alger, 18: 310, *Phlebotomus (Paraphlebotomus) sergenti* Parrot<sup>[24]</sup>, Bull. ent. Res., 39: 97;<sup>[4]</sup>, Bull. Brit. Mus. Nat. Hist. (Ent.), 19: 17;<sup>[25]</sup>, 236;<sup>[26]</sup>, Medskaya Parazit., 43: 159-160;<sup>[22]</sup>, 16;<sup>[20]</sup>, Bull. Brit. Mus. Nat. Hist. (Ent.), 37: 236.

**Female:** 32 specimens were examined (Fig. 1). Head 0.52 (0.48-0.576) long, 0.228xlength of wing, 0.552 (0.512-0.60) broad, eyes 0.26 (0.24-0.28) long, 0.5xlength of head, distance between eyes 0.22(0.20-0.24), Wing (Fig. 1A) 2.28 (2.0-2.42) long, 0.686 (0.588-0.753) broad,  $\alpha=0.413$  (0.357-0.465),  $\beta=0.33$  (0.30-0.36),  $\delta=0.12$  (0.02-0.16), in 3 females from Quetta, a greater  $\delta$  0.187 and 0.232 are observed, respectively,  $\gamma=0.42$  (0.32-0.52),  $\pi=0.108$  (0.08-0.12), in 1 female from Quetta a smallest  $\pi$  of 0.075 is observed; alar index 1.15-1.29. Palp (Fig. 1B) 0.70-0.81 mm long, palpal formula 1,4,2,3, 5, but also 1,2-4,3,5, 1,2,4,3,5, relative length 1,3.5, 4.31, 3.2, 6.9; P3, was having about 15-20 Newstead's sensilla at the basal third of the segment, other segments have none. A3 (Fig. 1C) 0.282 (0.263-0.30) long, 0.123xlength of wing, 0.989xlength of labrum,

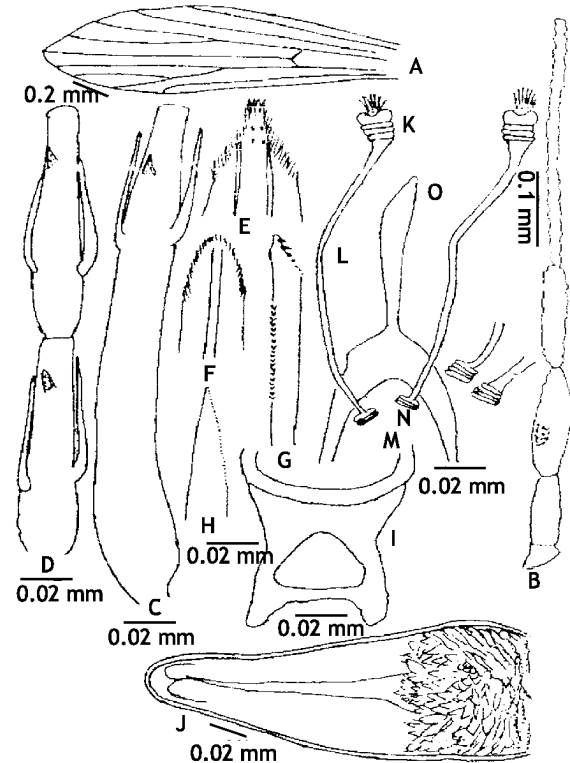


Fig. 1: Camera Lucida drawings of ♀ *Phlebotomus sergenti* from Balochistan (Pakistan) showing: wing (A), palp (B), the third (C), fourth (D, lower) and the fifth (D, upper) antennal segments, labrum (E), hypopharynx (F), maxilla (G), mandible (H), cibarium (I), pharynx (J), spermatheca (K), ducts (L), genital atrium (M), base of the ducts (N), genital fork (O)

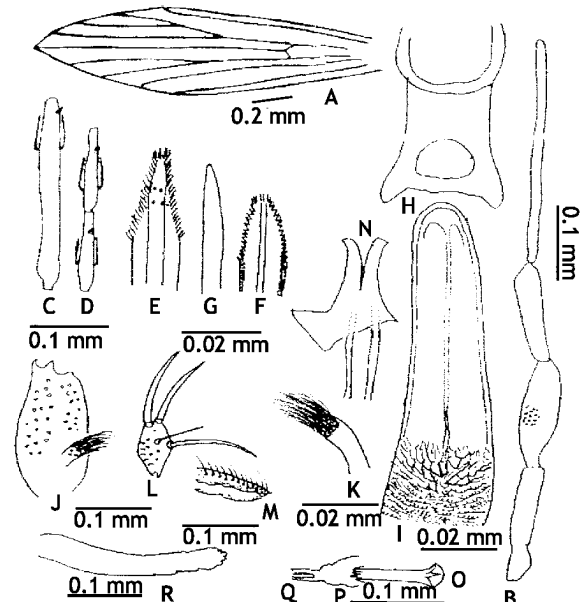


Fig. 2: Camera Lucida drawings of ♂ *Phlebotomus sergenti* from Balochistan (Pakistan) showing: wing (A), palp (B), the third (C), fourth (D, lower) and the fifth (D, upper), labrum (E), hypopharynx (F), maxilla (G), cibarium (H), pharynx (I), coxite (J), basal process (K), style (L), paramere (M), aedeagus (N), genital funnel (O), pump (P), filament (Q), surstyle (R)

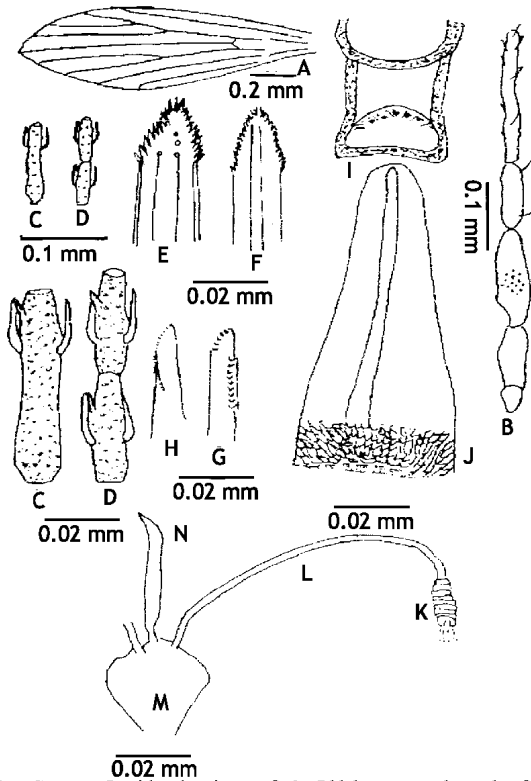


Fig. 3: Camera Lucida drawings of ♀ *Phlebotomus alexandri* from Balochistan (Pakistan) showing: wing (A), palps (B), the third (C), the fourth (D, lower) and fifth (D, upper) antennal segments, labrum (E), hypopharynx (F), maxilla (G), mandible (H), cibarium (I), pharynx (J), spermatheca (K), ducts (L), genital atrium (M), genital furca (N)

1.137xlength of A4+5, ascoid on A3 0.06 (0.05-0.07) long and 0.212xlength of segment; A4 (Fig. 1D, lower) 0.124 (0.112-0.134) long, ascoid on A4 0.066 (0.060-0.070) long, 0.532xlength of segment, A5 (Fig. 1D, upper) 0.124 (0.11-0.134) long, ascoid on A5 0.066 (0.06-0.070) long, 0.532xlength of segment. In almost all female flies, ascoids reach the upper end of antennal segments. Antennal segments III, IV and V have a single prominent papilla (Fig. 1C-1D). The positions of the papilla on the segments are: AIII, 0.807, AIV, 0.724, AV, 0.727. The positions of the ascoids on the segments are: AIII, 0.692, AIV, 0.344 and AV, 0.272. There are two ascoids on segments III to XV. Labrum (Fig. 1E) 0.285 (0.265-0.31) long, 0.038 broad, with four median, terminal sensillae stout and longer whereas about 15 adorals, sensilla depth 0.056, labrum is 0.125xlength of wing. Hypopharynx (Fig. 1F) 0.025 broad, with 3 apical pointed teeth and with 16 curved teeth on each side, dental depth 0.040. Maxilla (Fig. 1G) 0.012 broad with seven lateral (two large and more prominent, other 2 rounded and less prominent, remaining three very minute dot like) and 18 ventral teeth, dental depth 0.088. Mandible (Fig. 1H) 0.009 broad with about 35 teeth, dental depth 0.048, 3 curved teeth per 0.004. Proboscis 0.285

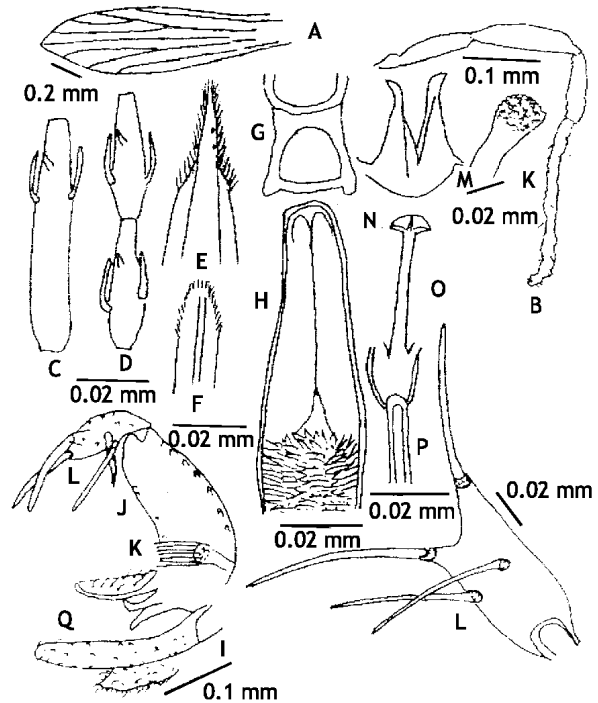


Fig. 4: Camera Lucida drawings of ♂ *Phlebotomus alexandri* from Balochistan (Pakistan) showing: wing (A), palp (B), the third (C), fourth (D, lower) and fifth (D, upper) antennal segments, labrum (E), hypopharynx (F), cibarium (G), pharynx (H), male terminalia (I), coxite (J), basal process of coxite (K), style (L), aedeagus (M), genital funnel (N), genital pump (O), filament (P), surstyle (Q)

(0.265-0.31) long, 0.125xlength of wing. Cibarium (Fig. 1I) 0.054 (0.044-0.065) broad, the side walls of cibarium bear 5-6 small spicules and 3-4 minute chitinous dots scattered at antero-median part of cibarium, chitinous arch well developed, pigment patch and anterior process both absent. Pharynx (Fig. 1J) 0.238 (0.22-0.256) long, pharynx is 1.31-1.42 times as long as broad and its posterior portion is 3.87-3.9 times as wide as the narrowest anterior part. The anterior edge of armature forms an almost straight line. The posterior part of the armature is composed of several broad and long leaves like spicules and basal part consisted of straight transverse edges with minute teeth whereas more distally pharyngeal armature consists of long broad spicules directing obliquely towards the center. Female genitalia: spermatheca (Fig. 1K) 4-5 segmented, global apical segment 0.021 (0.011-0.032) long, 0.015 (0.014-0.016) broad and remaining segments 0.006 (0.007-0.008) long, 0.013 (0.012-0.014) broad, hair like tubules of apical segment 0.012 (0.013-0.014) long, spermathecal ducts (Fig. 1L) 0.025 (0.024-0.026) long, with separate openings into genital atrium which is 0.048 B (Fig. 1M), the distal portion of the duct in the genital atrium appear like asymmetrical bell with shrunken edges (Fig. 1N), genital fork (Fig. 1O) 0.11-0.12 long.

**Male:** 52 specimens were examined (Fig. 2). Head 0.49 (0.48-0.51) long, 0.24xlength of wing, 0.50 (0.48-0.52) broad, eyes 0.24 (0.224-0.256) long, 0.489xlength of head, eyes 0.175 (0.16-0.19) broad, distance between eyes 0.16 (0.15-0.168). Wing (Fig. 2A) 2.041 (1.842-2.172) long, 0.587 (0.511-0.649) broad  $\alpha=0.36$  (0.303-0.427) long,  $\beta=0.335$  (0.288-0.397) long,  $\delta=0.099$  (0.062-0.147), greatest  $\delta=0.15$  and 0.16 are observed in 2 males from Quetta, a smallest  $\pi=$  of 0.04 is also observed in a specimen from Quetta,  $\gamma=0.384$  (0.32-0.456) mm, alar index 1.052-1.075. Palps (Fig. 2B) 0.85 long, formula 1,2,4,3,5 and also 1,2-4,3,5 and 1,4,2,3,5. relative length 1, 2.75, 3.51, 2.85, 6.36, 2.85, 6.36. Newstead sensilla situated on the middle of segment 3 and number was about 15-20. A3 (Fig. 2C) 0.285 (0.253-0.311) long, 0.139xlength of wing, 1.239xlabrum length, 1.096xlength of A4+5, ascoid on A3 0.05 (0.04-0.06) long, 0.175xlength of segment, A4 (Fig. 2D, lower) 0.13 (0.12-0.14) long, ascoid on A4., 0.05 (0.04-0.06) long, 0.384xlength of segment, A5 (Fig. 2D, upper) 0.13 (0.12-0.14) long, ascoid on A5 0.05 (0.04-0.06) long, 0.384xlength of segment, antennal segments III, IV and V have a single prominent papilla (Fig. 2C-2D). The positions of the papilla on the segments are:

AIII, 0.864, AIV, 0.7 and AV, 0.666. The positions of the ascoids on the segments are: AIII, 0.68, AIV, 0.2 and AV, 0.25. There are two ascoids on segments III to XV. Proboscis 0.265 (0.23-0.29) long, 0.129xlength of wing. Labrum (Fig. 2E) 0.23 (0.20-0.25) long, 0.112xlength of wing, labrum with 2 stout apical sensilla and about 20 long narrow adorals, sensilla depth 0.044. Hypopharynx (Fig. 2F) with about 14 teeth on each side, dental depth of 0.028. Maxilla (Fig. 2G) a sword like structure without teeth. Cibarium (Fig. 2H) 0.048 (0.039-0.057) broad, cibarium as in female but chitinous dots and spicules weak. Pharynx (Fig. 2I) 0.205 (0.19-0.22) long, fore-width 0.045 (0.045-0.050), hind-width 0.063 (0.056-0.070), armature 0.055 (0.050-0.060) height, 0.268xlength of pharynx, 1.4 times as wide posteriorly as anteriorly, pharynx 3.25 (3.14-3.39) times long its greatest breadth. The anterior edge of armature forms an almost straight line. The pharyngeal armature shows proximally a series of transverse ridges with serration and more distally long and broad spicules pointing towards center. Male terminalia: Coxite (Fig. 2J) 0.25 (0.22-0.29) long, 0.098 (0.07-0.110) broad, the basal process (Fig. 2K) is a characteristic feature of this species<sup>[22,27]</sup>, 0.052 (0.044-0.060) long, head of basal process 0.018 (0.016-0.02) broad, head small, slightly elongated with small tapering apex having 13-15 relatively thick hairs, yellow pigmented and slightly curved ventrally, next to head, a little differentiated neck 0.016 (0.014-0.017) broad. Style. (Fig. 2L) short, more or less oval, 0.095 (0.09-0.10) long,

0.046 (0.04-0.05) broad, style with: 2 terminal spines, the longest slightly curved, situated on small tubercles of equal length and thickness and 2 median spines, one of which at the middle of the body of style is short, thin and almost straight 0.077 (0.060-0.086) long, whereas the other one is located closer to the base and lateral border of the style, is the longest 0.125 (0.12-0.13) long, slightly curved and thinner than the terminal spines but relatively thicker than the short thin medium spine. Paramere (Fig. 2M) 0.12 (0.091-0.152) long with flat elliptical upper surface and with about 15 short spinules. Aedeagus (Fig. 2N) 0.063 (0.053-0.073) long, darkly pigmented, almost oval with slightly curved apex. Genital funnel (Fig. 2O) 0.048 long, 0.024 broad, genital pump (Fig. 2P) 0.176 (0.16-0.193) long, genital filament (Fig. 2Q) 0.19 (0.18-0.2) long. Filament/pump=1.079 (1.036-1.125). Surstyle (Fig. 2R) 0.255 (0.24-0.27) long and about the length of coxite.

**Distribution:** Balochistan. New Record, Present survey: Barshore, Chaman, Kohlu, Kuchlak, Muslim Bagh, Ornach, Panjigour, Pishin, Qillah Abdullah, Qillah Saifullah, Ziarat. These localities except Barshore, Kuchlak, Muslim Bagh, Qilla Saifullah and Ziarat are important foci of cutaneous leishmaniasis. Flies were collected from indoors and out doors using mouth aspirators and sticky traps. Barkhan and Bibertek<sup>[7]</sup>, Bostan<sup>[6]</sup>, Kahan<sup>[7]</sup>, Lehri<sup>[5,7]</sup>, Mastung<sup>[6]</sup>, present survey], Quetta<sup>[3,6]</sup> [present survey], Sangsila<sup>[7]</sup>, Sibi<sup>[5,6,7]</sup>, Talli<sup>[7]</sup>, Zhob<sup>[6]</sup>.

*Phlebotomus (Paraphlebotomus) alexandri* Sinton<sup>[10]</sup>.

*Phlebotomus sergenti* var.<sup>[11]</sup>, Bull. ent. Res., 11:309 (♂), *Phlebotomus sergenti* var. *alexandri*<sup>[10]</sup>, Indian J. Med. Res., 16: 308,<sup>[28]</sup> Bull. ent. Res., 21: 533; *Phlebotomus alexandri*<sup>[29]</sup>, Indian J. Med. Res., 20: 58,<sup>[30]</sup> Indian J. Med. Res., 21: 418;

*Phlebotomus (Phlebotomus) alexandri* Sinton, Parrot<sup>[23]</sup>, Arch. Inst. Pasteur Alger, 18:310,<sup>[31]</sup> Arch. Inst. Pasteur Alger, 24: 67, *Phlebotomus (Paraphlebotomus) alexandri* Sinton, Theodor<sup>[32]</sup>, 19,<sup>[33]</sup> J. Med. ent., 1: 290,<sup>[4]</sup> Bull. Brit. Mus. Nat. Hist. (Ent.), 19: 15,<sup>[25]</sup> 241,<sup>[22]</sup> 25,<sup>[20]</sup> Bull. Brit. Mus. Nat. Hist. (Ent.), 37: 235,<sup>[34]</sup> Fauna of Saudi Arabia, 2: 259.

**Female:** 20 specimens were examined (Fig. 3). Head 0.48 (0.464-0.496) broad, 0.271xlength of wing, Eyes 0.256 (0.24-0.264) long, 0.533xbreadth of head, 0.2 (0.192-0.208) broad and distance between eyes 0.224 (0.216-0.232). Wing (Fig. 3A) 1.77 (1.60-1.84) long, 0.536 (0.48-0.60) broad,  $\alpha=0.315$  (0.24-0.424) long,  $\beta=0.249$  (0.20-0.30) long,  $\delta=0.068$  (0.04-0.08), in one specimen from Sibi a largest  $\delta$  is measured to be 0.16,  $\gamma=0.392$  (0.36-0.48) long,  $\pi=0.084$  (0.056-0.12), in

one specimen from Sibi  $\pi$  is measured to be 0.16, alar index=1.26 (1.20-1.41). Proboscis 0.266 (0.26-0.27) long. Palps (Fig. 3B) 0.708 long, formula 1,4,2,3,5 with relative length 100:297:390:280:658, with about 15-20 spatulate Newstead's sensilla over middle third of segment 3. A3 (Fig. 3C) 0.125 (0.11-0.14) long, 0.51xlength of labrum, 0.26xbreadth of head, 0.07xlength of wing, 0.469x length of proboscis, 0.925x length of A4 + 5, ascoid on A3 0.03 long. A4 (Fig. 3D, lower), 0.068 (0.066-0.070) long, ascoid on A4 0.028 (0.024-0.03) long and is 0.41xlength of segment. A5 (Fig. 3D, upper) 0.067 (0.064-0.070) long, ascoid on A5 0.029 (0.028-0.030) long, A4+5=0.135 (0.13-0.14) long. Antennal segments III, IV and V had a single prominent papilla (Fig. 3C,D). The positions of the papillae on the segments are: AIII, 0.083, AIV, 0.68 and AV, 0.62. The positions of the ascoids on the segments are: AIII, 0.66, AIV, 0.37 and AV, 0.31. There are two ascoids on segments III to XV. Labrum (Fig. 3E) 0.245 (0.24-0.25) long, 0.030 broad, labrum with three thin apical sensilla, adorals small and narrow and a sensilla depth 0.048, labrum is 0.51x breadth of head, 0.138x length of wing. Hypopharynx (Fig. 3F) 0.024 broad with about three apical and nine teeth on each side and dental depth of 0.04. Maxilla (Fig. 3G) with five lateral and nine ventral teeth and dental depth of 0.056. Mandible (Fig. 3H) narrow, 0.017 broad, with 7-8 re-curved teeth per 0.01. Cibarium (Fig. 3I) 0.042 (0.036-0.049) broad, chitinous arch well developed and darkly pigmented, cibarial ventral plate in the form of convex arch, 7-10 minute rounded dot like denticles scattered at antero-central part of the cibarium and posterior to these are 5-8 triangular denticles in a zigzag row and also relatively larger 4-6 small spicules attached to both sides of the cibarium. Pharynx (Fig. 3J) conical, gradually widening posteriorly, 0.184 (0.16-0.21) long, pharynx was 2.10-2.42 times as long as broad and its widest posterior portion is 1.78-2.0 times as wide as the narrowest anterior part, height of armature 0.039 (0.030-0.050), 0.211xlength of pharynx. The anterior margin of pharyngeal armature forms an almost a straight line, armature darkly pigmented and almost rectangular in shape, broad scales lie one above the other at the sides, armature occupies base of the pharynx, very few punctuate rows of teeth at base of pharynx and some times absent. Female genitalia: Spermatheca (Fig. 3K) 0.029 (0.025-0.032) long, with 7-9 segments, apical segment not expanded and not larger than other segments, anterior segments of spermatheca 0.013 (0.012-0.014) broad, posterior segments 0.013 (0.012-0.014) broad, ducts (Fig. 3L) 0.174 (0.15-0.195) long, without transverse striations and with separate openings into genital atrium (Fig. 3M) which was 0.051 (0.048-0.054) broad, genital furca (Fig. 3N) 0.098 (0.096-0.10) long.

**Male:** 22 specimens were examined (Fig. 4). Head 0.392 (0.384-0.40) broad, 0.251xlength of wing, Eyes 0.18 (0.16-0.20) long, 0.459xbreadth of head, 0.144 broad and distance between eyes 0.164 (0.152-0.176). Wing (Fig. 4A) 1.56 (1.36-1.76) long, 0.42 (0.36-0.48) broad,  $\alpha$ =0.22 (0.176-0.28) long,  $\beta$ =0.18 (0.16-0.20) long,  $\delta$ =0.7 (0.064-0.08) long, but in one specimen from Khuzdar  $\delta$  is noted to be zero,  $\pi$ =0.080 (0.04-0.12) long. Alar index 1.22 (1.1-1.4), however, in 2 specimens: 1 from Sibi and 1 from Khuzdar,  $\alpha$  is found to be equal of  $\beta$ . Proboscis 0.18 (0.16-0.20) long. Palps (Fig. 4B) 0.66 long, formula 1,2-4,3,5 or 1,2,4,3,5 with relative length 100:285:342:285:7.71, with about ten to 15 spatulate Newstead's sensilla in middle third of segment 3. A3 (Fig. 4C) 0.13 (0.12-0.14) long, 0.083x length of wing, 0.33x breadth of head, 0.722xlength of proboscis, 0.866xlength of labrum, 0.822xlength of A4+5, ascoid on A3=0.03 long, 0.23xlength of segment. A4 (Fig. 4D, lower) 0.08 (0.07-0.090) long, ascoid on A4 0.03 long and is 0.375xlength of segment. A5 (Fig. 4D, upper) 0.078 (0.070-0.086) long, ascoid on A5 0.03 long and 0.384xlength of segment. The positions of the ascoids on the segments are: AIII 0.692, AIV, 0.351 and AV, 0.368. There are two ascoids on segments III to XV. Antennal segments III, IV and V have a single papilla (Fig. 4C-4D). The positions of the papillae on the segments are: AIII, 0.769, AIV, 0.648 and AV, 0.657. Labrum (Fig. 4E) 0.15 (0.14-0.16) long, 0.026 broad, with three or 4 apical sensilla and about fifteen laterals, sensilla depth 0.036. Hypopharynx (Fig. 4F) 0.016 broad with three long apical teeth and about eight lateral teeth and the dental depth of 0.024. Cibarium (Fig. 4G) 0.035 (0.030-0.040) broad, chitinous arch developed, anterior process and pigment patch both absent, ventral plate in the form of a convex line, cibarial armature not seen. Pharynx (Fig. 4H) 0.14 (0.158-0.17) long, pharynx is 3.15-4.93 times as long as broad and its greatest breadth is 1.23-1.86 times as broad as its anterior narrowest part, height of armature 0.035 (0.030-0.040), teeth occupies the posterior quarter (0.25) of the pharynx, anterior edge of teeth formed an almost straight line, the posterior part of the armature was in straight or curved punctiform ridges whereas antero-central part is composed of scales pointing backwards. Male terminalia (Fig. 4I): coxite (Fig. 4J). 0.20 (0.18-0.22) mm long, 1.69xlength of style, 1.025xlength of surstyle and 0.10 (0.08-0.12) broad, basal process of coxite (Fig. 4K) 0.032 (0.025-0.04) long with small, thin and almost rounded head 0.023 (0.022-0.025) broad, a differentiated neck (next to head) 0.014 (0.014-0.015) broad, head with about 15-18 pigmented long brush like thick hairs directed obliquely down. Style (Fig. 4L) slender, 0.118 (0.11-0.124) long, 0.042 (0.04-0.046) broad, bearing four spines: 2 median (1 terminal and 1 sub terminal), the 2 median spines are of unequal length stand

in almost at same position on two small tubercles. Median spines are at 0.58x length of style. One of the median spine short [0.08 (0.07-0.09) long], thin and straight whereas the other median spine located close to the lateral border of the body of the style are longer [0.095 (0.09-0.10)], a little curved and thicker. Both terminal and sub terminal spines are of the same length (0.12), same thickness and curvature but stand on separate tubercle. Terminal tubercle longer 0.038 (0.036-0.040) long, whereas subterminal tubercle short (0.01 long) and is at 0.818 of the length of style. Paramere 0.114 long, with flat elliptical surface covered with short hairs. Aedeagus (Fig. 4M) 0.072 (0.074-0.076) long, darkly pigmented with slightly curved apex ventrally. The shank of the aedeagus terminating into a tip, shaped like a cutting end of a chisel. Genital funnel (Fig. 4N), 0.026 long and 0.016 broad, genital pump (Fig. 4O) 0.115 (0.11-0.12) long, filament (Fig. 4P) 0.17 (0.15-0.19) long, F./P.=1.36-1.58. Surstyle (Fig. 4Q) 0.195 (0.18-0.21) long and 1.0-0.954xlength of coxite.

**Distribution:** Balochistan. New Record, Present survey: Khuzdar, Nal, Sibi, Turbat, Wadh. These localities are important foci of cutaneous leishmaniasis. Flies were collected from indoors and out doors using mouth aspirators and sticky traps.

**Differential diagnosis of *Ph. alexandri*:** The morphology of the pharynx and pharyngeal armature, small and thin basal process of coxite and shorter A3 of *Ph. alexandri* make it differs markedly from other species of *Paraphlebotomus*. It differs from following related species in following features:

***Ph. sergenti*:** *Ph. alexandri* is readily distinguishable from *Ph. sergenti*. Its female differs: in the shape of spermathecae, which have 8-9 segments as against three to four in *Ph. sergenti*, in the antennae and in the pharynx also. The teeth occupy only base of the pharynx and are a little more oblique and extend slightly less forward than in the pharynx of *Ph. sergenti* ♀. Further, *Ph. sergenti* ♀ has longer A3, pharynx bottle shaped, pharynx anterior part narrow and posterior side wall do show a constriction and uniform blunt broad leaf like pharyngeal teeth directing obliquely down. *Ph. sergenti* ♂ differ clearly by having oval style with terminal spines stand on tubercles of equal length, characteristic size and shape of basal process and absence of a marked constriction between head and neck and long genital pump. Whereas *Ph. alexandri* ♂ can be clearly distinguished from *Ph. sergenti* by the character of small and thin head of basal process of coxite and marked constriction between head

and neck, slender and long style, terminal spines of the style stand on the tubercles of unequal length, short genital pump and by the short length of the third antennal segment.

**Key for identification of Pakistani *Phlebotomus sergenti* and *Ph. alexandri*:** Cibarial teeth absent or if present always in the form of spicules not arranged in a definite row, pigment patch absent, hind end of abdominal tergites 2-6 with many erect hairs, style of male with four or 5 spines..... Genus *Phlebotomus*

**Female:** Antenna 3 longer (0.263-0.30), 0.967-0.992xlength of labrum, labrum is longer 0.265-0.31 long, female pharynx bottle shaped, pharynx length to breadth ratio=2.10-2.42 pharynx anterior part narrow and posterior side walls show a clear constriction, pharyngeal teeth are uniform blunt broad leaf like directed obliquely down, to the center and 4 or 5 segments in the spermatheca, with broad apical segment are the important diagnostic features for identification of ..... *Ph. sergenti*

A3 relatively shorter 0.11-0.14 in length, 0.51x length of labrum, labrum shorter 0.24-0.25 in length, pharynx gradually widen posteriorly, pharynx length to breadth ratio=1.31-1.42, spermathecal segments 7-9, apical segments not expanded and not larger than other segments..... *Ph. alexandri*

**Male:** A3 comparatively larger in length (0.25-0.31), labrum longer (0.20-0.25) pharynx length to breadth ratio= 3.14-3.39, shorter style, basal process (long, 0.044-0.060) is also characteristics, with asymmetrical thin head directed obliquely down, a little differentiation in between head and neck of basal process, genital pump long (0.16-0.193) with broad funnel (0.024), genital filament/ pump ratio=1.036-1.125..... *Ph. sergenti*  
A3 shorter (0.12-0.14), labrum shorter (0.14-0.16), pharynx length to breadth ratio=3.15-4.93, basal process of coxite relatively shorter (0.025-0.04) with thick mushroom shaped head and fan shaped long hairs, a marked differentiation in between head and neck, long slender style, terminal spines of the style stand on tubercles of unequal length, short genital pump (0.11-0.12) with less broad funnel (0.016), genital filament/pump ratio=1.36-1.58..... *Ph. alexandri*.

## DISCUSSION

A global discussion about the variation between Pakistani specimens with published data from other countries and especially intrapopulation variation (Table 1, 2) is given as under:-

Table 1: Comparison of taxonomic characters (in mm) of *Phlebotomus sergenti* Parrot

♀ Taxonomic characters	Balochistan (SW Pakistan) (Present study)	Transcaucasia and Central Asia (Perfiliev, 1968)	Saudi Arabia (Lewis and Butti ker, 1980)	Syria & Iraq (Adler and Theodor, 1929) (In micron)	Countries of the east mediterranean region (Rasht and Nadi m 1992)	Africa and Portugal (Kirk and Lewis, 1951)
Wing Length	2.28 (2.0-2.42)	2.5-2.7		-	-	2.5
Breadth	0.686 (0.588-0.753)	0.80-0.87	2.31 (2.08-2.46)			0.8
δ	0.12 (0.02-0.16)					+0.08
Alar index	1.15-1.29					1.0
A3 length	0.282(0.263-0.30)	0.27-0.31	-	A3>A4+5	240-330	0.27-0.30
A3/A4+5	A3>A4+5	A3>A4+5, A3=A4+5		but in one specimen A3<A4+5		
A3/labrum	0.989					
Ascoïd 4/A 4	0.532	Half of the length of the segment.	-	2/3-15	-	2/3-15
Ascoïd formula	2/3-15		-	-	260-450	0.32-0.44
Labrum	0.285 (0.265-0.31)	0.32-0.45	-	-	-	0.32-0.44
Palp formula	1,4,2,3,5; 1,2-4,3,5; 1,2,4,3,5	1,4,2,3,5; 1,2-4,3,5, other formula may also occur.	-	1,2,4,3,5; 1,4,2,3,5; 1,2-4,3,5.	-	1,4,2,3,5; often variable
Pharynx	Pharynx is 3.4 times as wide posteriorly as anteriorly, several broad and long leaf like spicules directing obliquely towards center whereas basal part consists of transverse ridges edged with teeth, armature occupies 3.5x length of pharynx.	Pharyngeal armature occupies about a quarter of the length of pharynx, spines well marked in form of large, broad, uniform scales with rounded or slightly pointed tip, scales standing more densely in middle of armature, a few rows of punctuate lines at the base of the area of spines, base of the pharynx less than 3 times as wide as the apex.	Pharyngeal armatur is about a quarter of the length of the pharynx, teeth not very numerous and rather large.	Pharyngeal armature in optical section appear as long curved scales, anteriorly they appear as long curved teeth with their axis longitudinal or slightly oblique.	-	Pharynx is about 2.5 times as wide posteriorly as anteriorly and armed with very characteristic broad scales li ke teeth.
Spermatheca	4-5 segmented, apical segment large and globular, the other segments narrow towards the duct.	With only 3 segments, apical segment large and rounded, the other segments narrow towards the duct.	Spermatheca segmented.	5-6 segmented, super segment is rounded and larger than other which diminish in diameter before backwards.	-	4-6 segments, superior segment larger than others which diminish in size from about down wards, ducts enter vulva separately.
♂Taxonomic characters						
Wing length	2.041 (1.84-2.17)	1.8-2.2	2.20 (2.05-2.37)	-	-	1.9
Breadth	0.587 (0.511-0.649)	0.52-0.55				0.53
δ	0.099 (0.062-0.147)					+0.07
Alar index	1.052-1.075					1.23
A3	0.285 (0.253-0.311)	0.22-0.36	0.29 (0.28-0.31)	Antennae as in the females	220-340	0.25
A3 / A4+5	A3>A4+5	A3>A4+5				1.0-1.4
A3 / labrum	1.239					A3=A4+5
Ascoïd 4/ A4	0.384	Ascoïd 4 about the third of the segment	-	-	-	0.3
Labrum	0.23 (0.20-0.25)	0.24-0.32	0.25	-	220-320	-
Palp formula	1,2,4,3,5, or 1,2-4,3,5, 1,4,2,3,5,	1,4,2,3,5, or 1,2,4,3,5,	-			
Pharynx	1.4 times as wide posteriorly as anteriorly	-	-	Palps as in the females		1,4-2,3,5, About twice as wide posteriorly as anteriorly
Coxite	0.25 (0.22-0.29)			Armature less developed		
Head of basal process	0.018 (0.016-0.02)	0.23-0.25				
Neck	0.016 (0.04-0.017)	0.016	-	-	200-260	0.25
Style						
Length	0.095 (0.09-0.10)	0.11				
Breadth	0.046 (0.04-0.05)	0.04	-	-	80-110	0.11
Paramere	0.12 (0.091-0.152)	0.18-0.20	-	-	-	0.15
Aedeagus	0.063 (0.053-0.073)	0.08	-	-	-	0.06
Surstyle	0.255 (0.24-0.277)	0.26-0.28	-	-	-	0.13



Table 2: Comparison of taxonomic characters (in mm) of *Phlebotomus alexandri* Sinton

	Pakistan Northern Balochistan (Pakistan) (Present study)	Ex. USSR, Central areas, Sindh and NWFP (Lewis, 1967)	Asian Countries (Perfiliev, 1968)	Countries of the Eastern Mediterranean region (Rashti, and Nadim, 1992) (in micron)	Saudi Arabia (Lewis and Buttiker, 1980)	Anglo Egyptian, Sudan (Kirk and Iran, Lewis, 1951)	(Nadim and Javadian) 1976	Afghanistan (Artemiev, 1978)
♀ Taxonomic characters								
Wing length	1.77 (1.60-1.84)		1.8-2.0		1.80(1.5-2.2)	1.85-2.0		
Breadth	0.536 (0.48-0.6)	-	0.48-0.53	-		0.48-0.51	-	-
Head length	0.48 (0.46-0.49), 0.271xlength of wing	-	-	-	0.39(0.35-0.43), 0.22 (0.20-0.24) xlength of wing	-	-	-
Eye length	0.256 (0.24-0.264), x0.5xbreadth of head				0.20(0.18-0.23), 0.51 (0.46-0.57) x length of head			
Labrum length	0.245 (0.24-0.25), 0.138x length of wing, 0.51xlength of head	-	0.22-0.26	220-250	0.23(0.21-0.27), 0.13 (0.12-0.14) xlength of wing, 0.60(0.56-0.63) length of head	0.22-0.26	-	-
A3 length	0.125 (0.11-0.14),	0.12-0.16 mm,	0.13-0.17	100-170	0.14-0.16,	0.14-0.016	Short, 0.5-	A3 shorter
A3/labrum	0.51 x length of labrum	0.5-0.6xlength of labrum			0.6 x length of labrum	0.58-0.62	0.6xlength of labrum	Than A4+5
Ascoid on A3	short and stout							
A4 length	0.068							
A5 length	0.067							
A3/A4+5	almost equal in length (A3=A4+5)		Segment 4 usually as long as 5			A3 < A + 5 0.5		
Ascoid 4/A4	0.41							
Pharynx	Pharyngeal armature - occupies 0.211x length of pharynx, pharynx with hind width with 1.91 times fore width	-	-	Pharynx with hind width 2-2.5 times fore width, pharyngeal armature occupies 0.25x length of pharynx	-	-	-	-
Spermatheca	7-9 segmented, apical segment not larger than the other segments, hind segments similar in breadth as fore segments.	-	6-8 segmented, apical segment not larger than the other segments	-	-	-	-	-
<i>♂ P. alexandri</i>								
Wing length	1.56 (1.36-1.76)		1.3-2.0		1.8(1.5-2.2)	2.0		
Breadth	0.42 (0.36-0.48)		0.35-0.55			0.56		
Labrum length	0.15 (0.14-0.16)		0.20-0.30	150-300		-	-	-
A3 length	0.15 (0.14-0.16),	0.154 (northern areas) 0.12-0.16	0.13-0.16	100-150	0.12-0.16,	A3/L =0.7-1.1	0.12-0.16,	Short
A3/labrum	0.866	0.12-0.16	A3<A4+5		0.7-0.9	A3≤A4+5	0.7, length of labrum	
A3/A4+5	A3<A4+5	(West Pakistan), 0.66xlength of labrum (northern area), 0.7-0.9x length of labrum (West Pakistan)						
Ascoid 4/A4	0.375	-	Slightly shorter than half length of segment	-	-	-	-	-
Coxite length	0.20 (0.18-0.22)	-	0.19-0.23	160-260	-	0.21	-	-
Breadth of head of the basal process, Breadth of neck of the basal process	0.023 (0.022-0.025) 0.014 (0.014 -0.015)		0.024-0.025 0.016-0.019					

Table 2: Continue

♂ <i>P. alexandri</i> taxonomic characters	Pakistan Northern Balochistan (Pakistan) (Present study)	Ex. USSR, Central areas, Sindh and NWFP (Lewis, 1967)	Asian Countries (Perfiliev, 1968)	Countries of the Eastern Mediterranean region (Rashti, and Nadim, 1992) (in micron)	Saudi Arabia (Lewis and Buttiker, 1980)	Anglo Egyptian, Sudan (Kirk and Iran, Lewis, 1951)	(Nadim and Javadian) 1976	Afghanistan (Artemiev, 1978)
Style length	0.118 (0.11-0.124)	-	0.14-0.15	90-150	-	0.15	-	-
Subterminal spine	0.818 of style length	-	0.77-0.88 of style length	-	-	-	-	-
Paramere	0.114	-	-	-	-	0.2	-	-
Aedeagus	0.072 (0.074-0.076)	-	-	-	-	0.09	-	-
Sperm pump	0.121 (0.11-0.13)	-	-	-	0.12	-	-	-
Surstyle	0.195 (0.18-0.21), slightly shorter than coxite	-	-	-	-	0.24-0.26, longer than coxite	-	-

**Ph. sergenti:** (Table 1) The male flies from Pakistan showed a slightly greater wing length and breadth, delta, A3, A3>A4+5 as compared with those from Africa and Portugal<sup>[35]</sup>, a greater ascoid 4/A4 as compared with those from Transcaucasia and Central Asia<sup>[25]</sup> and Africa and Portugal<sup>[35]</sup>, a greater wing breadth as compared with those from Transcaucasia and Central Asia, a greater coxite as compared with those from East Mediterranean Region countries<sup>[36]</sup>, Africa and Portugal and Transcaucasia, Central Asia, a greater breadth of head and neck of basal process as compared with those from Transcaucasia and Central Asia, a greater aedeagus and surstyle as compared with those from Africa and Portugal. Further, male flies did show a slightly shorter A3 as compared with those from Transcaucasia, Central Asia, Saudi Arabia<sup>[34]</sup> and countries of East Mediterranean Region, a shorter A3/labrum as compared with those from Africa and Portugal, a shorter labrum as compared with those from Saudi Arabia, Africa, Portugal, Transcaucasia and Central Asia, a shorter style as compared with those from countries of East Mediterranean Region, Transcaucasia, Central Asia, Africa and Portugal, a shorter paramere, aedeagus and surstyle as compared with those from Transcaucasia and Central Asia. Similarly, female flies from Pakistan were showing a slightly shorter length and breadth of wing as compared with those from Africa, Portugal, Transcaucasia, Central Asia and Saudi Arabia, a shorter A3 as compared with those from countries of East Mediterranean Region, Transcaucasia and Central Asia and a shorter labrum as compared with those from Africa, Portugal, Transcaucasia, Central Asia and countries of the East Mediterranean Region. Similarly, female flies of Pakistan were measured to have a slightly greater delta, alar index and a ratio of hind width to forewidth of pharynx as compared with those from Africa and Portugal, a greater ascoid 4/A4 as compared with those from Transcaucasia, Central Asia, Africa and Portugal, a greater pharyngeal armature/length of pharynx as compared with those from Transcaucasia, Central Asia

and Saudi Arabia. Female specimens of the present study were found having fewer number of spermathecal segments (4-5) as compared with those from Africa, Portugal (4-6 segments), Syria and Iraq (5-6 segments)<sup>[37]</sup> but have a larger number of segments as compared with those from Transcaucasia and Central Asia (3 segments). Further, pharyngeal armature was found 3.5xlength of the pharynx as compared with those from Transcaucasia, Central Asia and Saudi Arabia where pharyngeal armature occupy about a quarter of the length of pharynx. Pharynx was measured 3.4 times as wide posteriorly as anteriorly in Pakistani forms whereas pharynx of 2.5 times as wide posteriorly as anteriorly was reported from Africa and Portugal. Similarly, A3 in female specimens from Pakistan was measured slightly larger as compared with those from Afghanistan (240-300 µm)<sup>[22]</sup> while A3 in specimens of both the countries was measured greater than A4+5. Spermatheca of both the countries were observed to have a similar number of segments (4-5). However, female specimens from Pakistan were showing a striking resemblance with the published data from Egypt<sup>[38]</sup> in characters like a shorter A3 than labrum and 3rd and 4th antennal segments with ascoids almost reaching the tip of the segments. Nadim and Javadian<sup>[39]</sup> reported 4-6 segmented spermathecae in Iranian forms whereas Pakistani forms were found having 4-5 segmented spermatheca. However, Pakistani forms were observed in full accord with those from Transcaucasia and central Asia in diagnostic characters like ascoid formula, palpal formula and A3>A4+5. Further, Pakistani forms did show similarity with those from Africa and Portugal in A3, a similar A3>A4+5 as compared with those from Syria, Iraq, Africa, Portugal, Transcaucasia and central Asia. Like wise, ascoid formula and palpal formula of Pakistani forms did show a resemblance with those from Transcaucasia, Central Asia, Africa, Portugal, Syria and Iraq.

**Ph. alexandri:** Results of the present study are compared with the published data of *Ph.alexandri* from Asian and

African countries (Table 2). *Ph. alexandri* ♂ Balochistan were observed with a slightly shorter wing as compared with published data of this species from Saudi Arabia<sup>[34]</sup>, Anglo-Egyptian Sudan<sup>[35]</sup>, a shorter labrum as compared with specimens of Central Asian countries<sup>[25]</sup> and East Mediterranean Region countries<sup>[36]</sup>, a shorter A3/labrum as compared with that of Saudi Arabia, Anglo-Egyptian Sudan, a shorter ascoid4/A4 as compared with that of Central Asian countries, a shorter coxite as compared with that of Central Asian countries, East Mediterranean Region countries, a shorter head and neck of basal process of coxite as compared with that of Central Asian countries, a shorter style as compared with that of Central Asian countries and a shorter aedeagus as compared with that of Anglo-Egyptian Sudan. Specimens from SW Pakistan were found having a slightly larger wing as compared with that of Central Asian Countries, a larger antenna 3 as compared with that of northern areas of Pakistan, Iran<sup>[39]</sup> and Saudi Arabia and a larger A3/labrum as compared with that of northern areas of Pakistan and Iran. A3<A4+5 of Pakistan specimens were found similar as that from Central Asian countries and Anglo-Egyptian Sudan. *Ph. alexandri* ♀ from SW Pakistan were found having a slightly shorter wing as compared with that of Central Asian countries, Saudi Arabia and Anglo-Egyptian Sudan, a shorter labrum as compared with that of Saudi Arabia and Central Asian countries and a shorter A3 as compared with that of northern areas of Pakistan<sup>[4]</sup>, Central Asian countries, East Mediterranean Region countries and Saudi Arabia.

Female specimens of the present study were observed to be in full accord in A3<A4+5 and ascoid formula 2/3-15 with the forms of Afghanistan<sup>[22]</sup>, Central Asian countries and Saudi Arabia.

The results of the present study show that *Ph. alexandri* is a rare species (82/2013, 4.1%) and is found distributed only in south- central Balochistan. It has a discontinuous distribution and it prefers hot and dry areas. Though it has a very wide distribution in Asia and Africa and thus, usually termed "Mediterranean" species in the widest sense<sup>[33]</sup>, it has also been recorded from Palestine<sup>[13]</sup> and from north Iran . Theodor and Mesghali<sup>[33]</sup> reported this species from central desert of Kerman, Shiraz and Pak-Iran border cities of Zahedan and Iran Shaher, which are close to the Pakistani Balochistan Province. However, during the present study, the author could not record it in the collection of sandflies from Pak-Iran border areas, extreme south of Makran region, extreme western Pak-Afghan border nor it was observed in north and western areas of the province where cutaneous leishmaniasis is quite prevalent. Similarly, *Ph. sergenti* is also found to be an uncommon species

(7%, N=2013). It has localized but discontinuous distribution especially in the province and becomes rare towards south as the author could not report it from extreme south. The overall prevalence of this species shows that it prefers relatively semi colder areas and its presence partly coincides with that of the prevalence of cutaneous leishmaniasis in the region.

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