DIVERSITY OF MOSQUITOES COLLECTED FROM THE SOUTHERN AREAS OF KHYBER PAKHTUNKHWA - PAKISTAN

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ABSTRACT:

OBJECTIVES:
The objective of the study was to assess the diversity of mosquitoes in various towns of the southern belt of KPK.

METHODOLOGY:
This was a descriptive study that was conducted in numerous towns of Western belt of Khyber Pakhtunkhwa. The study areas were Darra Adam Khel, District Kohat, District Karak, District Bannu, District D.I. Khan and newly merged districts Mir Ali and Miranshah. From each study site, the samples were collected randomly. The sample collection was done through survey and area visits whereas; the identification process was done in a parasitology laboratory of Hayatabad-Peshawar. Sampling was done from June 2016 to May 2017.

RESULTS:
A total of 2150 adult mosquitoes were gathered and collected from 42 different locations of the southern belt of KPK and were identified. Based on their identification, 5 genera of the mosquitoes were recognized which were Culex, Anopheles, Psorophora, Aedes and Uranotenia. Culex was found to be most dominant in all the visited areas with a percentage of 12.65 in Miranshah followed by 11.81 in Mirali, 7.16 in Karak, 6.88 in Darra Adam Khel, 6.69 in D.I.Khan, 6.41 in Kohat and 5.11 in Bannu respectively. The results of our findings also revealed the presence of Culex genera in all the habitats and remained the dominant genera among the others followed by Anopheles and Psorophora. Aedes was found in the habitat of plants and grasses etc. whereas Uranotenia was found in marsh/swampy areas as well as in plants/grasses habitat only.

CONCLUSION:
The outcomes reveal that a climate shifting and extensive urbanization process is enforcing the diversity of mosquitoes’ fauna in the southern belt of KPK.

KEYWORDS: Diversity, Fauna, Mosquitoes, Anopheles, Culex, Aedes

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INTRODUCTION:
Numerous deadly diseases including Dengue, Malaria, Chikungunya and Zika virus are caused by few medically important arthropods known as mosquitos. The annual death ratio due to lethality of this vector is around one million¹. There are some 3541 reported species of mosquitoes around the globe. These species belong to 42 genera². As far as Pakistan is concerned, the reported species are 134, which fit in Culicinae and Anopheline sub-families³. The well-known human malaria vector has its place in genus Anopheles whereas the vector for bird malaria belongs to genus Culex. The role of mosquitoes in the food chain of numerous eco-systems is well established⁴. Vector borne diseases are threatening the lives of people in developing countries like Pakistan. Here in Pakistan, the climatic changes are encouraging the
chances of mosquito borne diseases\(^5\). In Lahore, there were 11283 reported cases of Dengue fever in year 2012\(^6\). More or less 2179 cases of Dengue fever were found positive in a recent outburst in KPK. The South Asian countries’ climatic change has worsened the prevalence of mosquito borne diseases in the area\(^2,3\). Furthermore, numerous other factors including rainfall, temperature, humidity etc. are in favor of an increase in population of mosquitoes with an enhanced survival\(^9,10\). Various studies have been conducted throughout Pakistan to report the diversity of mosquitoes in different areas\(^11-13\). In the year 2013, a study was conducted in Lahore on bio-diversity of mosquitoes with its climatic scenario. The reported species were Anopheles stephensi, Aedes aegypti, Culex quinquefasciatus and Anopheles subpictus. The mosquito fauna of Pakistan was powered by the addition of three Culex and one Anopheles species during 1934 to 1971\(^16\). In the year 1977, 30 species were reported from Changa Manga, Lahore\(^17\). In 1993, numerous mosquitoes’ species were reported from Peshawar and its neighboring areas with their relative abundance\(^18\). The breeding of the mosquitoes can be seen in vicinities where water remains stagnant in pools or ditches etc., near holes of the trees, pools containing leafy debris, forest area and blocked gutters etc. Adult mosquitoes remain throughout their life near larval habitation. Plant nectar and juices remain the major source for their feeding. The female mosquitoes also nourish on juices but they need a blood diet for developing their eggs. The study was aimed to assess the diversity of numerous species of mosquitoes in varying habitats of southern KPK districts.

**METHODOLOGY:**

This was a descriptive study that was conducted in numerous towns of Western belt of Khyber Pakhtunkhwa to compare population dynamics and to establish a baseline data on diversity of mosquitoes’ genera. The localities included were Darra Adam Khel, District Kohat, District Karak, District Banu, District D.I.Khan and newly merged districts Mir Ali and Miranshah. From each study site, the samples were collected randomly. The sample collection was done through survey and area visits whereas; the identification process was done in a parasitology laboratory of Hayatabad-Peshawar. Sampling was done from June 2016 to May 2017. Special codes were allotted for various mosquitoes and tagged. A total of 42 localities (6 from each area) were assessed in which, some localities were of habitats. These habitats were: toilets, muddy or wet areas, plants, trenches, animal hutch and houses etc. Various habitats were observed with naked eye for the determination of prevalence of mosquitoes. The mosquitoes were collected from the specified localities at the ground level earlier in morning and evening time. Sweepers and battery-operated aspirators were used for the collection of adult mosquitoes. The collected samples were shifted to plastic jars that were covered with nets for further identification process in the parasitology lab of Hayatabad-Peshawar. The samples were specified and tagged properly and their respective geographical locations were mentioned. To kill the mosquitoes, they were sucked from the plastic jars with the help of an aspirator and were then shifted to conical jars that were kept airtight and contained a cotton pad immersed in chloroform. All the adult mosquitoes were killed within 2 to 3 minutes. The mosquitoes were preserved in test tubes that contained a tiny quantity of silica gel for preservation. The identification of the mosquitoes was done using “Standard Taxonomic Keys”\(^4,5\).

The ethics committee of the Bacha Khan University, Charsadda approved the study.

By using the following formula, relative abundance was determined.

\[
\text{Relative abundance} = \frac{n}{\text{total no of species}} \times 100
\]

In the above formula, \(n\) is the number of mosquitoes divided by the total number of mosquitoes gathered from the localities multiplied by 100. A value of more than 10% of relative abundance was considered as dominant species whereas from 3 to 10% were termed as sub-dominant species.

**RESULTS:**

A total of 2150 adult mosquitoes were gathered and collected from 42 different sites of the southern belt of KPK both from urban and rural areas of the selected districts and were identified.

![Figure 1: Western Belt of KPK and Sampling Areas Shown with Arrows](image)
Table 1: Geo-Coordinates of Sampling Areas

| No | Sampling Area       | Geographical Region | Geo-Coordinates                |
|----|---------------------|---------------------|--------------------------------|
| 1  | Darra Adam Khel     |                     | 33° 6' 54.9664" N 71° 5' 43.9260" E |
| 2  | Kohat               |                     | 33° 19' 60.00" N 71° 09' 60.00" E |
| 3  | Karak               |                     | 33° 6' 54.9664" N 71° 5' 43.9260" E |
| 4  | Bannu               |                     | 32° 59' 9.9996" N 70° 36' 14.9904" E |
| 5  | D.I.Khan            |                     | 31° 49' 53.3352" N 70.2776° E |
| 6  | Mirali              |                     | 32.9700° N 70.2776° E |
| 7  | Miranshah           |                     | 33.00250° N 70.06889° E |

Table 2: Prevalence of Different Genera of Mosquitoes' in Southern Belt of KPK

| Genus  | Darra | %    | Kohat | %    | Karak | %    | Bannu | %    | D.I Khan | %    | Mirali | %    | Miran Shah | %    | Total |
|--------|-------|------|-------|------|-------|------|-------|------|----------|------|--------|------|-------------|------|-------|
| Culex  | 148   | 6.88 | 138   | 6.41 | 154   | 7.16 | 110   | 5.11 | 144      | 6.69 | 254    | 11.81| 272       | 12.65| 1220  |
| Anopheles | 21   | 0.97 | 27    | 1.25 | 40    | 1.86 | 68    | 0.03 | 82       | 3.81 | 74     | 3.44 | 78         | 3.62 | 390   |
| Aedes   | 4     | 0.18 | 19    | 0.88 | 44    | 2.04 | 76    | 3.53 | 98       | 4.55 | 101    | 4.69 | 121        | 5.62 | 463   |
| Psorophora | 4   | 0.18 | 8     | 0.37 | 12    | 0.55 | 4     | 0.18 | 14       | 0.65 | 6      | 0.27 | 4          | 0.18 | 52    |
| Uranotenia | 2   | 0.09 | 0     | 0    | 4     | 0.18 | 1     | 0.04 | 5        | 0.23 | 5      | 0.23 | 8          | 0.37 | 25    |
| Total   | 636   | 556  | 448   | 154  | 88    | 268  | 2150  |      |          |      |        |      |            |      |       |

Table 3: Different Genera of Mosquitoes' Collected in Numerous Habitats in Southern Belt of KPK

| Genus         | Marsh/Swamp areas | Toilets | Plants/Grasses | Animal Hutch (U) | Animal Hutch (R) | Trenches | House/Containers | Total |
|---------------|-------------------|---------|----------------|------------------|------------------|----------|------------------|-------|
| Culex         | (n=)              | %       | (n=)           | %                | (n=)            | %        | (n=)            | %     |
| 194           | 30.5              | 424     | 76.25          | 180              | 40.17           | 154      | 100             | 180   |
| Anopheles     | 204               | 32.07   | 54             | 44               | 9.82            | 0        | 0               | 0     |
| Aedes         | 218               | 34.27   | 78             | 14.02            | 167             | 37.27    | 0               | 0     |
| Psorophora    | 0                 | 0       | 0              | 25               | 5.58            | 0        | 0               | 0     |
| Uranotenia    | 20                | 3.14    | 0              | 32               | 7.14            | 0        | 0               | 0     |
| Total         | 636               | 556     | 448            | 154              | 88              | 268      | 2150            |       |

Table 4: Details of the Species Collected from Different Genera

| No | Genera | Species                |
|----|--------|------------------------|
| 1  | Culex  | Cx. pseudovishnui (U)  |
| 2  |        | Cx. pseudovishnui (R)  |
| 3  |        | Cx. quinquefaciatus (U)|
| 4  |        | Cx. quinquefaciatus (R)|
| 5  |        | Cx. tritaeniorhynchus (U)|
| 6  |        | Cx. tritaeniorhynchus (R)|
| 7  | Anopheles | An. subpictus (U) |
| 8  |        | An. subpictus (R)      |
| 9  |        | An. stephensi (U)      |
| 10 |        | An. stephensi (R)      |
| 11 | Aedes  | Ae. aegypti (U)        |
| 12 |        | Ae. aegypti (R)        |
| 13 |        | Ae. albopictus (U)     |
| 14 |        | Ae. albopictus (R)     |

DISCUSSION:

In this study, overall, 14 species belonging to 3 different genera (Culex, 6 species, Aedes, 4 and Anopheles, 4 species) were identified that were collected from both urban and rural areas of the study scenario. The other collected species remained unidentified. The identified species were: Cx. pseudovishnui (U), Cx. pseudovishnui (R), Cx. quinquefaciatus (U), Cx. quinquefaciatus (R), Cx. tritaeniorhynchus (U), Cx. tritaeniorhynchus (R), An. subpictus (U), An. subpictus (R), An. stephensi (U), An. stephensi (R), Ae. aegypti (U), Ae. aegypti (R), Ae. albopictus (U) and Ae. albopictus (R) respectively. In a study that was conducted in Murree-Pakistan, 13 species were mainly identified that were collected from the various localities of the Murree. The most abundant species were Aedes aegypti, Armigerus obturans
and Cx. fuscitarsis respectively followed by Cx. nilgiricus and Cx. vagans. The all reported species in Murree were the same as reported by Barruad (1934) excepting one type that was Cx. raptor. The aforementioned study revealed all the collected species majorly in the month of July to November whereas April was the month in which, minimum number of species were collected. The months of January, February, March and December were the ones in which no sampling was done because of rain and snowfall. The mosquitoes’ fauna of various districts of Pakistan is reported in various research studies. In a study conducted in Lahore, 29 species of mosquitoes were reported. In another study in the same city, the mosquitoes’ fauna was reported to be 31 from seven different genera. The outcomes of a study done in Faisalabad displayed 36.8% existence of Culex genera and 32.1% existence of Anopheles genera. In a report that was published from Thailand, revealed the dominant existence of Culex species in all the collected mosquitoes’ species. In a Tanzanian report, it was stated that the Anophelinae species were the dominant among all from traditional flooding rice irrigation ecosystem. A local study that was conducted in Peshawar revealed the presence of Culex and Anophelinae species in polluted water trenches in Peshawar. The study revealed the presence of Culex species more or less 99% from the samples. The outcomes of a Pakistani study that was conducted in Faisalabad shows 26.3% population of Aedes species in district Faisalabad and its nearby territories. Mukhtar et al. explored the breeding of mosquitoes in wastewater irrigation where he found three genera Aedes, Culex and Anopheles. Ali and Rashid showed the presence of Cx. quinquefasciatus and An. stephensi in polluted water of Palosai stream in district Peshawar. The Aedes species were missing in this water. This research showed that the Aedes species are inhabitants of temporary habitats only. In a study that was conducted in Jhelum-Pakistan, a total of 21 mosquito species containing 365 samples were documented. The species observed were: Culex (9), Anopheles (6), Lutzia (2), Aedes (2), and Armigeres (2). Anopheles stephensi was gathered from 3 habitats, comprising animal shelters, houses and water streams etc. Higher richness/abundance was observed in animal shelters and houses, whereas lower richness/abundance was observed in water streams. Anopheles maculatus was gathered from 2 habitats, comprising animal shelters and water streams. Higher richness/abundance was observed in animal hutches, whereas lower richness/abundance was perceived in water streams. Anopheles annularis collection was done from 3 habitats, containing animal shelters, houses and water streams. Higher richness/abundances were observed from animal shelters and housing areas, whereas the lower richness/abundance was perceived from water rivulet. Anopheles culicifacies and Anopheles tessellatus shared the equal habitats, comprising animal shelters, housing areas and watercourses. Higher richness/abundances of Anopheles culicifacies were observed from animal huts and rivers, whereas lower abundance was observed from housing colonies. Anopheles tessellatus samples were perceived in the same abundances from these habitats. Anopheles theobaldi was gathered from 2 habitats, i.e. animal shacks and inhabited areas. The abundances found from these habitats were equal. Our study revealed that Culex was found to be most dominant in all the visited areas with a percentage of 12.65 in Miranshah followed by 11.81 in Mirali, 7.16 in Karak, 6.88 in Darra Adam Khel, 6.69 in D.I.Khan, 6.41 in Kohat and 5.11 in Bannu respectively. The results of our findings also revealed the presence of Culex genera in all the habitats and remained the dominant genera among the others followed by Anopheles and Psorophora. Aedes was found in the habitat of plants and grasses etc. whereas Uranotenia was found in marsh/swampy areas as well as in plants/grasses habitat only. Our results revealed the presence of Culex genera in ditches (100%), in animals’ hutches (100%), in toilets (76.25%), in plants and grasses etc. (40.17%) and in marshy and swampy places (30.50%). The results show the plants and grasses (5.58%) as a positive habitat for Aedes genera while marshy/swampy places (3.14%) and plants/grasses (7.14%) as positive habitat for genera Uranotenia. The positive habitats for genera Psorophora were marshy/swampy places (34.27%), toilets (14.02%) and plants/grasses (37.27%) respectively.

**CONCLUSION:**

The outcomes of this study reveal that climate shifting and extensive urbanization process is enforcing the diversity of mosquitoes’ fauna in the southern belt of KPK. The results will be a helping hand in controlling and managing their effective control in the study areas. More studies with larger samples are recommended to cope with the deadly effects of various species of the mosquitoes in the area.

**LIMITATIONS:**

Our study is limited to rural and urban areas of a
few districts of KPK. More areas of the western belt are to be needed to explore for the assessment of mosquitoes profile and diversity by using a larger number of samples in future.

CONFLICT OF INTEREST: None

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