Minor Project Report

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Title: Mosquito diversity of Baddi (Himachal Pradesh) and surrounding areas: Impact of developmental changes and demographic factors.

Mosquitoes are cosmopolitan, as these tiny insects have great adaptability to exploit and inhabit new areas. These insects have viped out many ancient human civilizations by spreading Malaria. Mosquitoes are also responsible for the transmission of Yellow fever, Dengue, Dengue hemorrhagic fever, Filariasis, Chikungunyia and several viral Encephalitides. The tropical and subtropical countries are facing the major challenge of reducing the spread of these diseases and preventing people from getting infected. Rapid development, ever-increasing human population, use of insecticides, industrialization, and migration of laborers are some of the factors responsible for making our country more vulnerable to disruptions and diseases.

The land of Baddi area used to be barren prior to the industrial development. Within two decades this area has become Asia's largest pharmaceutical hub and the biggest industrial centre in North India. Baddi is now known as the most developed region of Solan district. It is fastest growing town as well as a leading industrial centre. It is a part of industrial corridor stretching from Barotiwala to Nalagarh along the western border of Solan district. Today there are around 400 medical and pharmaceutical companies operating in Baddi, apart from 200 packaging units. This rapid industrialization and indiscriminate urbanization coupled with accelerated construction activity and aggregation of migrant labor has created mosquitogenic conditions in this area. The improper disposal of waste water has reshaped the ecology of mosquitoes that necessitated the exploring of the actual breeding spectrum of these insects, which can be further utilized in vector control programmes for containment of malaria and other mosquito borne diseases. There was need of evaluating and interpreting the interrelationship of economic & industrial development and vector borne diseases. Keeping in view the development related changes and demographic factors, intensive collection cum survey tours were conducted to study the exact status of mosquito fauna in Baddi. Earlier 44 species of mosquitoes were reported from Sirmour,Simla, Kinnaur, Mandi, Kullu, Kangra, Chamba, Lahul & Spiti areas of Himachal Pradesh (Rao et al. ,1973) and 15 species belonging to 5 mosquito genera from Dalhousi, Mandi, Chamba, Kullu, Kangra, Palampur and Chail areas were reported by kaur (2001). This area was

never explored for its mosquito diversity. It has been observed that the ecological changes associated with the developmental activities have negative health impact due to creation of new mosquito breeding sites. Mosquito eggs, larvae and pupae exist in the aquatic habitat, whereas the adults dwell on land. The spraying of insecticides to kill the adults pollutes the environment and develops insecticide resistance amongst mosquito species; hence focus should be on alternative methods to reduce the mosquito population at early stages. By identifying the preferable physico-chemical characteristics of the breeding water, the measures can be taken to reduce the suitability of habitat for mosquito survival. The later requires the extensive knowledge about habitat preferences of specific vector species; hence the impact of various water quality parameters like temperature, pH, Conductivity, Salinity, total dissolved solids, Dissolved oxygen and B.O.D. on the mosquito diversity has been studied.

OBJECTIVESES OF THE PROJECT: 1.To conduct entomological surveys in the industrial area of Baddi and its vicinity, so that the exact status of vector and non vector mosquito species can be assessed.

2. To study the role of important physicochemical factors (ph, temperature, humidity, conductivity, salinity, Total dissolved solids, Dissolved oxygen and B.O.D.) in the selection of breeding site by different species.

Results: In order to achieve the desired objectives of the present minor project, intensive and extensive collection cum survey tours were conducted in various localities of Baddi and its surrounding Katha, Barotiwala, Tipra, Jharmajri, Mandhala, Kishanpur, Dammuwala, Lakkarpur, Soorajmajra, Malkumajra, Maranwala, Nalagarh, Karuni, Sanerh, Sattiwala, Sainsiwal, Bhudd and Nawan Nagar from April 2017 to November 2018. Immature stages of mosquitoes were collected with the help of soup ladle from various types of water bodies (domestic, peridomestic, artificial and natural) in the sampling bottles. The bottles were properly labeled describing the name of breeding site, place, date and time. These immature stages of mosquitoes were brought to the laboratory to rear them into the adults. These specimens were preserved in the insect cabinets after mounting them on paper triangle. The various morphological traits were examined to identify them up to species level with the help of relevant keys given by Barraud (1934), Wattal & Kalra(1961) Srivanakarn (1976), Reuben *et al*. (1994), Reinert *et al*. (2004) and Tyagi *et al* (2014). 30 entomological surveys carried out in different localities of Baddi and surrounding areas resulted in to the collection of 42 mosquito species belonging to 11 genera.

List of Species reported along with site of collection :

Sr. No.	Name of mosquito Species	Site of collection
1	*Culex (Culex) quinquefasciatus* Say, 1823	Baddi, Nawan nagar, Barotiwala, Mandhala, Tipra, Sattiwala, Lakkarpul, Katha, Malkumajra, Bhudd

2	*Culex (Culex) vagans* Wiedemann, 1828	Baddi, Nawan nagar, Barotiwala, Dammuwala, Sattiwala, Tipra, Lakkarpul, Karuni, Kishanpur
3	*Culex (Culex) univittatus* Theobald, 1901	Barotiwala, Tipra, Baddi
4	*Culex (Culex) fuscocephala* Theobald, 1907	Tipra, Nawan nagar, Sattiwala, Lakkarpul, Kishanpur, Khud Kurahwala
5	*Culex (Culex) theileri* Theobald, 1903	Malkumajra, Tipra, Katha
6	*Culex (Culex) pseudovishnui* Colless, 1957	Nawan nagar, Tipra, Katha
7	*Culex (Culex) vishnui* Theobald, 1901	Nawan nagar, Katha, Nalagarh, Tipra, Mandhala, Lakkarpul, Sainsiwal, Baddi
8	*Culex (Culex) tritaeniorhynchus* Giles, 1901	Nawan nagar, Katha, Nalagarh, Tipra, Dammuwala, Lakkarpul, Mandhala, Baddi
9	*Culex (Culex) gelidus* Theobald, 1901	Nawan nagar, Katha, Karuni, Tipra, Sainsiwal
10	*Culex (Culex) whitmorei* (Giles, 1904)	Tipra, Nawan nagar, Baddi, Katha
11	*Culex (Culex) edwardsi* Barraud, 1923	Nawan nagar, Barotiwala, Katha, Sanerh, Tipra, Karuni, Sattiwala, Dammuwala, Lakkarpul, Soorajmajra, Bhudd, Sainsiwal, Charanea, Baddi
12	*Culex (Culex) barraudi* Edwards, 1922	Sattiwala, Nawan nagar, Mandhala, Sainsiwal, Lakkarpul, Baddi, Karuni, Dammuwala, Tipra
13	*Culex (Culex) sitiens* Wiedemann, 1828	Tipra, Nawan nagar, Mandhala, Baddi,
14	*Culex (Culex) mimulus* Edwards, 1915	Khud Kurhawala
15	*Culex (Oculeomyia) bitaeniorhynchus* Giles,1901	Soorajmajra, Sanerh, Nawan nagar, Tipra, Malkumajra, Dammuwala, Mandhala, Karuni, Sainsiwal
16	*Culex (Oculeomyia) infula* Theobald, 1901	Karuni, Khud kurhawala
17	*Culex (Eumelanomyia) pulvialis* Barraud, 1924	Tipra, Karuni
18	*Culex (Eumelanomyia) malayi* (Leicester, 1908)	Jharmajri
19	*Rhinoskusea longirostris* (Leicester, 1908)	Tipra
20	*Fredwardsius vittatus* (Bigot, 1861)	Sanerh, Mandhala, Karuni, Nawan nagar, Sattiwala, Sainsiwal, Malkumajra
21	*Mimomyia (Ingramia) fusca* (Leicester, 1908)	Tipra
22	*Mimomyia (Ingramia) chamberlaini* Ludlow, 1904	Tipra
23	*Stegomyia albopicta* (Skuse, 1895)	Barotiwala, Mandhala, Dammuwala, Malkumajra
24	*Stegomyia (Stegomyia) aegypti* (Linnaeus, 1762)	Baddi, Kishanpur
25	*Collessius (Alloeomyia) pseudotaeniatus* (Giles, 1901)	Dammuwala
26	*Lutzia (Metalutzia) fuscana* (Wiedemann, 1820)	Baddi, Sainsiwal, Tipra, Bhudd, Nawan nagar
27	*Lutzia (Metalutzia) raptor* Edwards	Malkumajra
28	*Armigeres (Armigeres) kuchingensis* Edwards, 1915	Barotiwala
29	*Aedimorphus culicinus* (Edwards, 1922)	Sainsiwal
30	*Mansonia (Mansonioides) annulifera* (Theobald, 1901)	Sainsiwal

31	*Anopheles (Anopheles) barbirostris* Van der Wulp, 1884	Nawan nagar
32	*Anopheles (Anopheles) peditaeniatus* (Leicester, 1908)	Baddi
33	*Anopheles (Cellia) annularis* Van der Wulp, 1884	Mandhala
34	*Anopheles (Cellia) culicifacies* Giles, 1901	Nawan nagar, Maranwala, Tipra, Sainsiwal, Karuni, Dammuwala, Lakkarpul, Khud kurhawala, Mandhala
35	*Anopheles (Cellia) fluviatilis* James, 1902	Nawan nagar, Sanerh
36	*Anopheles (Cellia) maculates* Theobald, 1901	Nawan nagar, Khud kurhawala
37	*Anopheles (Cellia) pseudojamesi* Stickland & Chowdhury, 1927	Nawan nagar
38	*Anopheles (Cellia) psudowillmori* (Theobald, 1910)	Khud kurhawala, Nawan nagar
39	*Anopheles (Cellia) stephensi* Liston, 1901	Nawan nagar
40	*Anopheles (Cellia) subpictus* Grassi, 1899	Tipra, Sattiwala, Mandhala, Nawan nagar, Katha, Khud kurhawala, Sainsiwal
41	*Anopheles (Cellia) vagus* Donitz, 1902	Tipra
42	*Anopheles (Cellia) willmori* (James, 1903)	Khud kurhawala

In the present collections the genus Culex emerged as the largest taxon with 18 species, followed by *Anopheles* with 12 species under it. The genera *Mimomyia*, *Lutzia* and *Stegomyia* are represented by 2 species each. Only one species referable to each of genera *Rhinoskusea*, *Fredwardsius*, *Armigeres*, *Aedimorphus, Mansonia* and *Collessius* could be collected from the area under study(Fig.1).

Nawan nagar and Tipra villages (2.5 km. from Baddi) togetherly comprise 28 percent of the mosquito diversity. 12 species could be collected from Baddi town. Mandhala is a small village 13 km away from Baddi. This village has a beautiful pond with naturally grown lotus plants in it, embankments of this pond has small grasses with small aquatic insects including mosquito larvae near to it. The collected larvae emerged into 11 mosquito species. In the village of Sainsiwal, lots of large ditches were dug by the slum residents to store rain water for various domestic purposes, which were niches of 10 mosquito species. The same number of species is reported from Karuni village (near Nalagarh). The areas like Katha, Khud Kurahwala, Lakkarpul and Dammuwala were surveyed for 8 mosquito species each. The pond of village Sattiwala reccieves industrial waste directly from nearby industry that serves as habitat of 7 *culex* species.6 mosquito species were reported each from Barotiwala and Malkumajra (Fig. 2).

Maximum number of mosquito species (26) were collected in the month of October, followed by July (21 species), November (16 species), June & September (13 species), August (12

species), May (11 species), April (8 species) and March with minimum no. of 5 mosquito species (Fig.3). No mosquito larva could be collected in the months of December to February. The most favorable temperature for mosquito larval endurance is 25-30°C, as 25 species were collected at this range followed by the 30.1-35°C with 18 species. As the temperature decreases the survival of most of the mosquito species is affected (Fig. 7). Ph 7-8 seems to be most suitable for the continued existence of mosquito species, the deviation from this ph range have negative impact on the mosquito diversity (Fig. 4).

Fig. 1. Composition of mosquito species under different genera

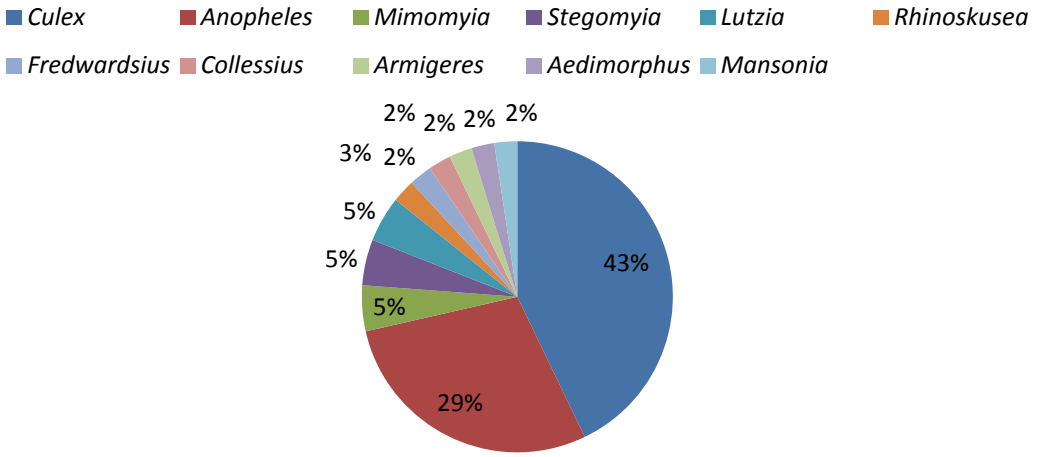
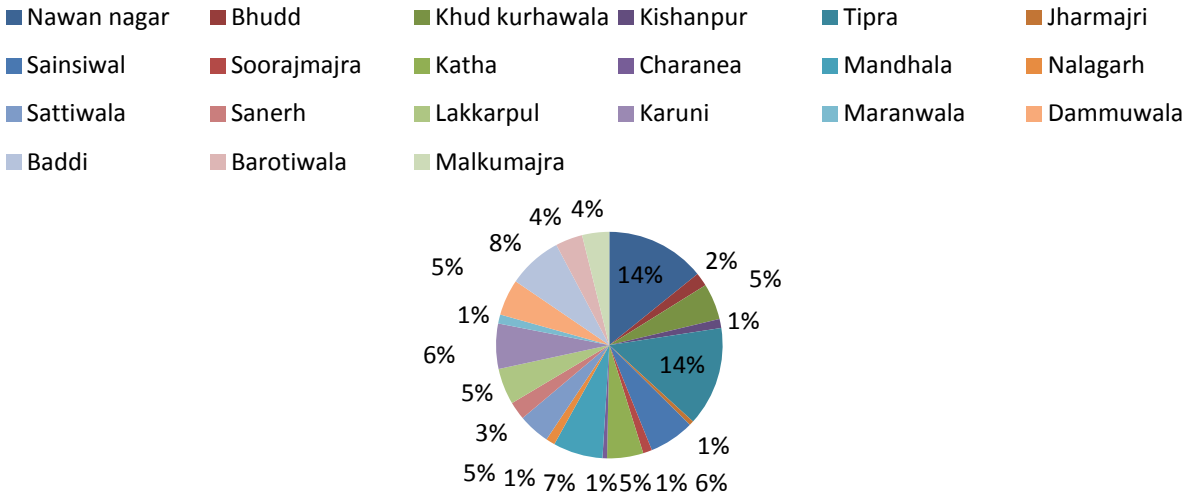
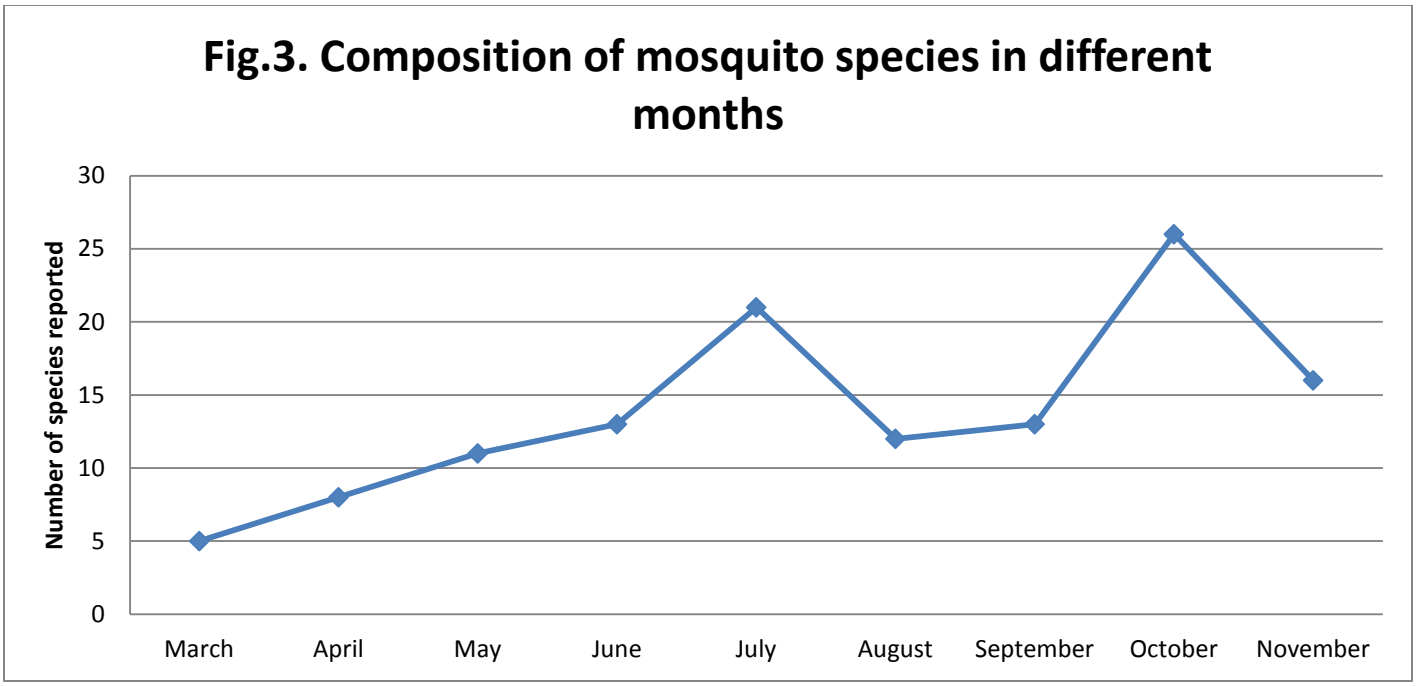


Fig. 2. Composition of mosquito species in different places





The water samples collected from the various mosquito habitats were brought to the laboratory. The physico-chemical parameters of water were measured with standard methods such as pH (pH meter); Conductivity, Salinity and total dissolved solids (uP conductivity/TDS/Salinity & temperature meter); Turbidity (Turbidity meter); Dissolved oxygen (D.O.Meter) and B.O.D. The environmental data like temperature, humidity and time of collection was noted in a field diary. Subsequent to the analysis of physicochemical parameters, it has been observed that most of the mosquito species have wide range of tolerance to the majority of the physicochemical characteristics of aquatic habitats. Culicine larvae are known for their ability to survive in the stagnant water bodies with very low or nil dissolved oxygen, the same has been reported in the present studies. The wide range of turbidity i.e. 0 to 845 shows that the survival of the mosquito species is hardly affected with the value of this parameter. The lowest recorded value of ph is 3.86(swampy habitat at Sanerh) while the highest is 9.28(industrial waste disposal site at Katha). During this period the average air humidity was 74.11% (for nearly all species). It has also been observed that young forms of some *Culex* species like *quinquefasciatus, edwardsi* and *vagans* crop up at the all ranges of conductivity, salinity and T.D.S. whereas *bitaeniorhynchus* and *Lutzia fuscanus* shows wide range of tolerance to T.D.S. only. The survival of larvae of *Culex vishnui* and *C. tritaeniorhynchus* are hardly affected with the value of conductivity. The majority of mosquito species (31) thrive at conductivity range of 300-600 µS/cm (Fig. 5),the salinity range of 200-400ppm and T.D.S. range of 70- 200ppm(Fig.6)

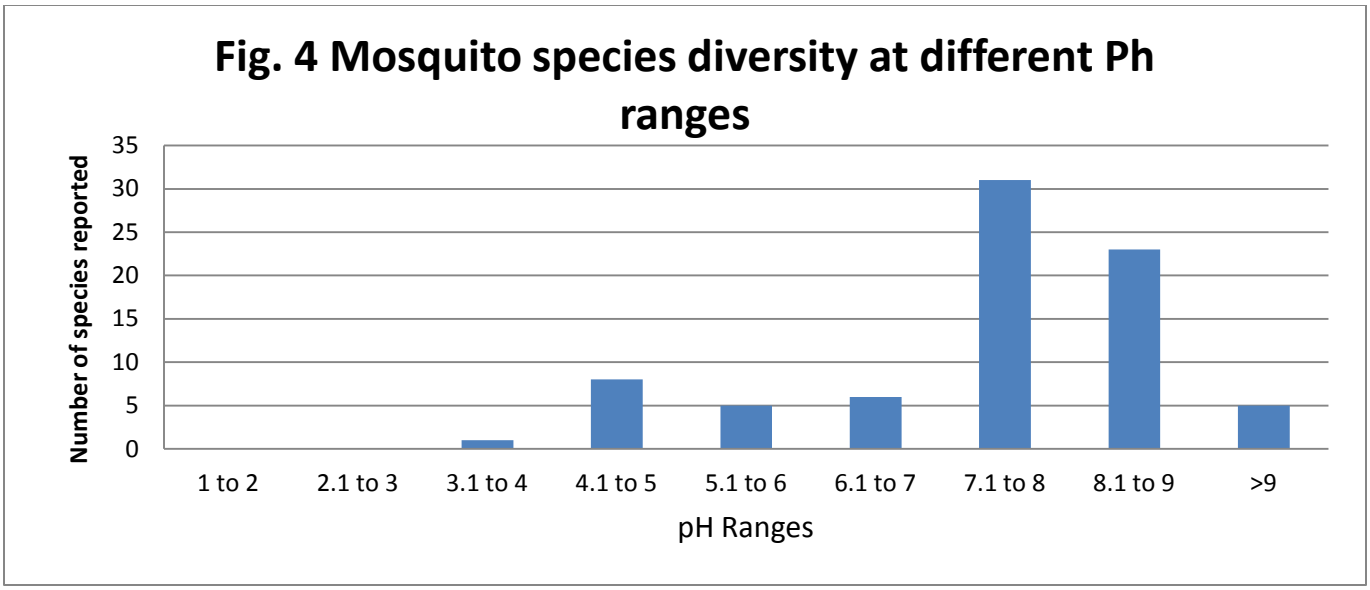


Fig. 4 Mosquito species diversity at different Ph ranges
Number of species reported
35
30
25
20
15
10
5
0
1 to 2
2.1 to 3
3.1 to 4
4.1 to 5
5.1 to 6
6.1 to 7
7.1 to 8
8.1 to 9
>9
pH Ranges

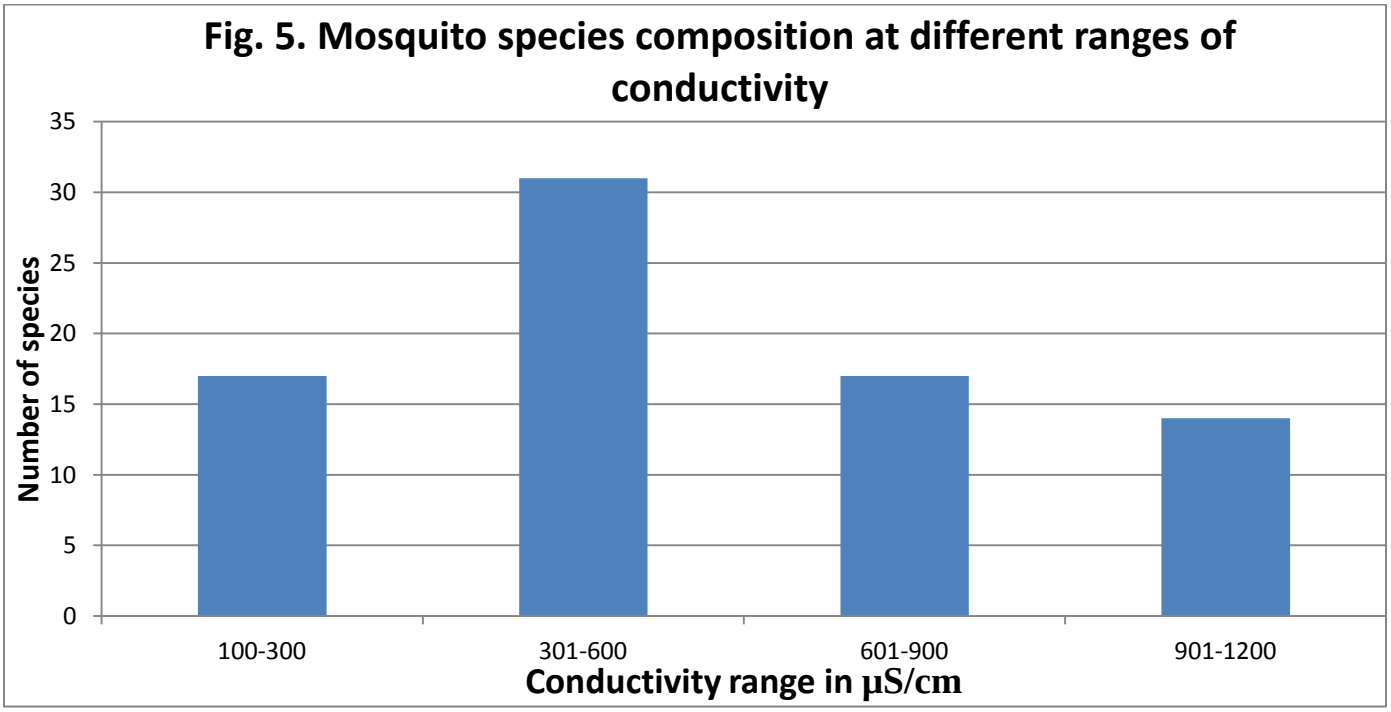
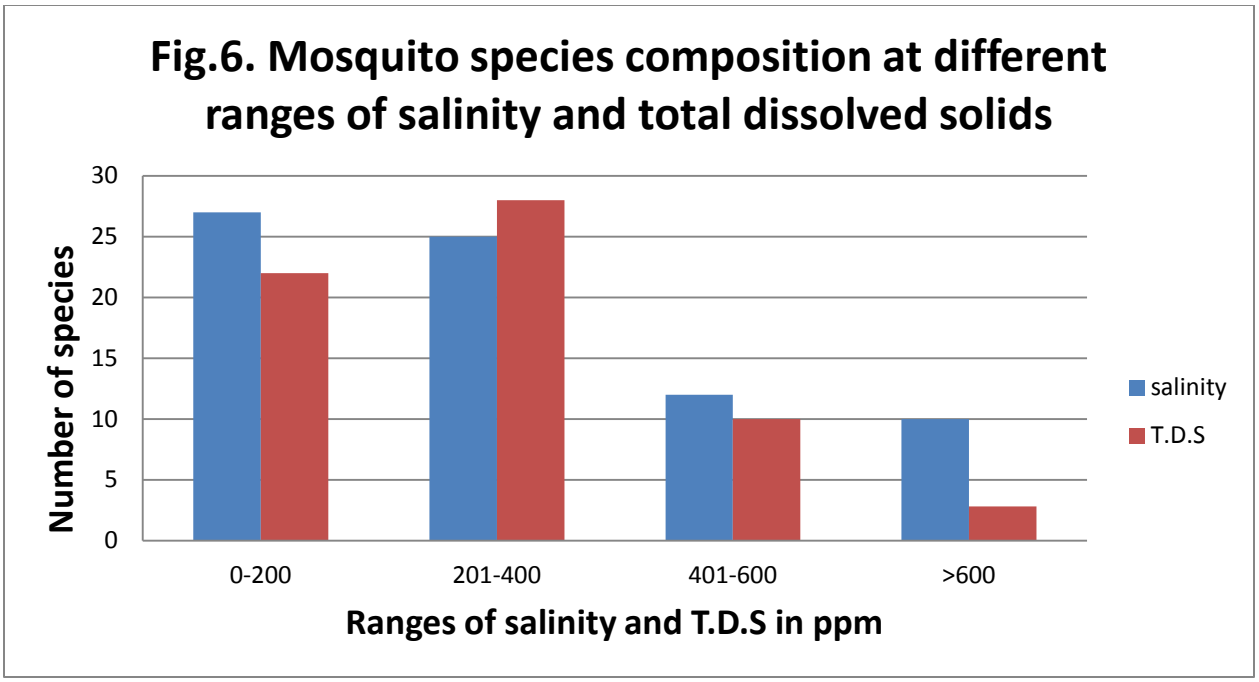
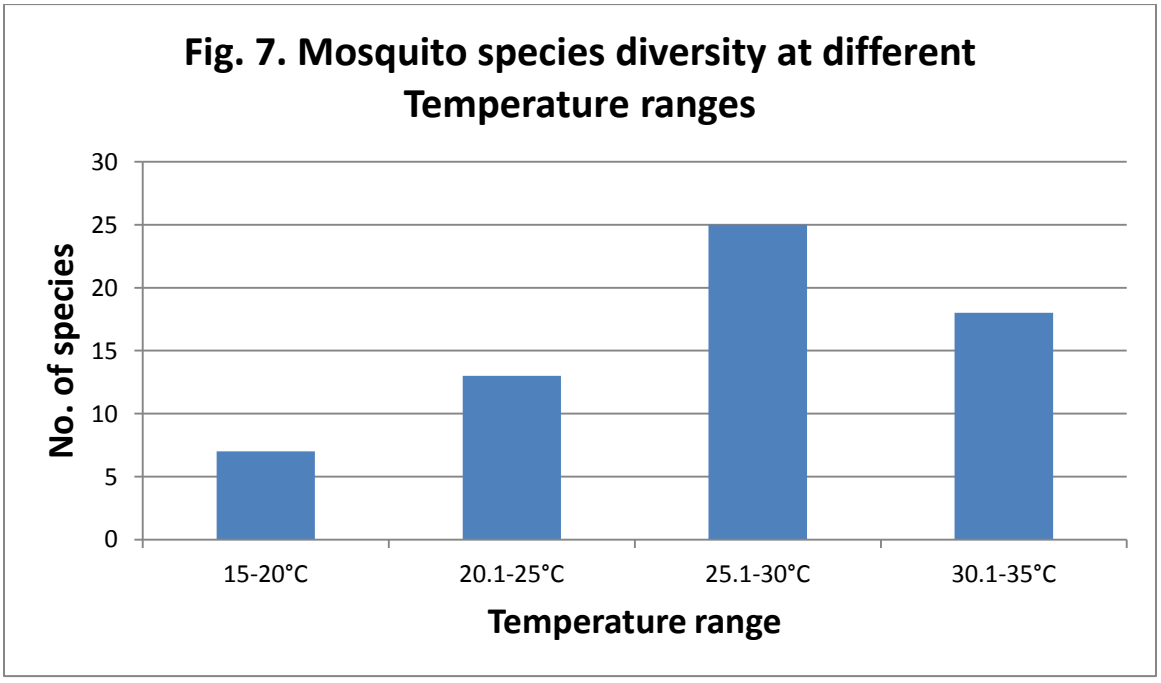


Fig. 5. Mosquito species composition at different ranges of conductivity
Number of species
35
30
25
20
15
10
5
0
100-300
301-600
601-900
901-1200
Conductivity range in µS/cm





The amount of dissolved oxygen was found very low in most of the habitats except the flowing water in sirsa tributary at Nalagarh (8.00mg/L) and Khud Kurahwala (5 mg/L). The D.O. amount in all the other mosquito larval sites was observed to vary in the range of 0.00mg/L to 1.7 mg/L. The B.O.D. values

showed a wide variation from 2 mg/L (in flowing water) to 144 mg/L (in stagnant water at lakkarpul). The larvae of 3 species of *Anopheles* were found co-existing with larvae of *Culex mimulus* in the slow moving water at Khud Kurahwala with B.O.D. of 3 mg/L. The average recorded range of B.O.D is 2mg/L to 27mg/L with exceptionally high values at the sites of blocked sewage drains. The later sites with B.O.D. amount of more than 100mg/L were the favourite breeding places of *Cx. Quinquefasciatus, vagans* and *theileri* etc.

Conclusion of the study:

The earlier studies on culicidae diversity of Himachal Pradesh dates back to 1933(Christophers), 1934(Barraud) and 1973(Rao et. al.). There are no records of the mosquito faunal studies from Solan district. Industrialization, urbanization and migration of labor in and around Baddi have resulted into the creation of conducive environment for mosquito survival. As a result numerous culicidae species have invaded this area to amplify the mosquito fauna. The reporting of 18 new records from this area of Himachal Pradesh is the result of invasion by mosquito species due to the establishment of new-fangled breeding places.

The prevalence of the vectors of zika, dengue, dengue hemorrhagic fever and chikungunya i.e. *Stegomyia aegypti* (Linnaeus,1762) , *Stegomyia albopicta* (Skuse,1895) and *Frewardsius vittatus* (Bigot, 1861)(Potential vector) is a threatening signal towards the prospect of these diseases. The later mosquito species show resistance to all the unfavorable environmental conditions at egg stage.

The vectors of malaria i.e various Anopheline species indicate the resurgence of this deadly disease in future. The species referable to genus *Culex i.e. vishnui, pseudovishnui, tritaeniorhynchus* may lead to the transmission of Japenese encephalitis, once the virus gets introduced from the neighbouring states.

Subsequent to the analysis of physicochemical parameters, it has been observed that most of the mosquito species have wide range of tolerance to the majority of the physicochemical characteristics of aquatic habitats.The larvae of culicine mosquitoes are known for their ability to survive in the stagnant water bodies with very low or nil dissolved oxygen, the same has been reported in the present studies. The rain puddles serve as the most favorable habitats for *F.vittatus,*whereas the domestic containers and tires being the most preferable habitats for *S.albopicta* and *aegypti. Anopheles* larvae were found in high density in the waters with good dissolved oxygen and low B.O.D. but *A. subpictus* was found in swamps sharing its habitat with *Cx. Bitaeniorhynchus.*

Nawan nagar marshy area

Nawan nagar pond

Dammowala pond

Mandhala pond

Baddi blocked drain

Sirsa River near baddi

Standing rain water Nawan nagar

Soorajmajra pond

Bhudd sliced water tank

Sattiwala pond

Sattiwala pond with high larval denisty

Katha industrial waste


Soorajmajra pond


Nalagarh


Baddi sewage disposal


Mandhala pond


Tipra pond


Lakkarpul


Karuni standing rain water


Malkumajra water in buckets


Baddi


Sainsiwal


Nawan nagar

Barotiwala

Nawan nagar cattle shed

Nawan nagar below the bridge

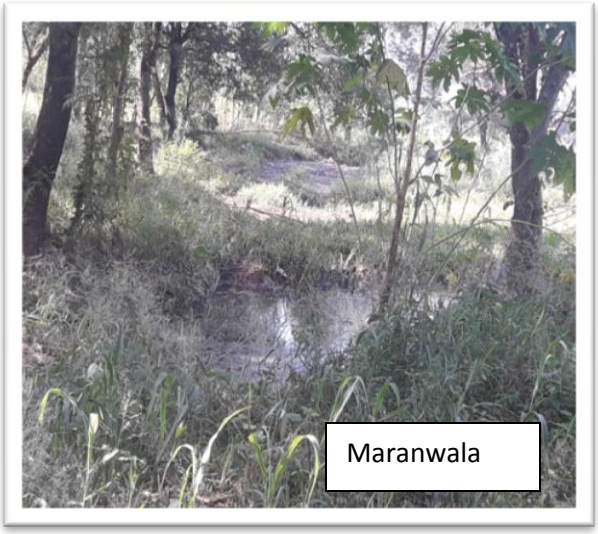
Nawan nagar agriculture run off

Tipra domestic waste disposal

Baddi industrial waste

Lakkarpul


Nawan nagar pothole


Maranwala


Khud Kurahwala


Sanerh

Refrences:

Barraud PJ (1934). The fauna of British India including Sri Lanka and Burma. Diptera, family Culicidae, Tribe Megarhinini and Culicini. Vol. 5, Taylor and Francis, London: 1-463.

Christophers SR (1933). The fauna of British India including Sri Lanka and Burma. Diptera, family Culicidae, Tribe Anophelini. Vol. 4, Taylor and Francis, London: 1-271.

Kaur J. (2001). Taxonomic studies on family Culicidae (Diptera) from North-West India with particular reference to the structure of external genitalia. Ph.D Thesis, Punjabi University, Patiala.

Rao TR, Dhanda V, Bhatt HR and Kulkarni SM (1973). Ind J Med Res. A survey of haematophagous arthropods in Western Himalayas, Sikkim, hill district of West Bengal. A general account.61: 1421-1437.

Reinert JF, Harbach RE and Kitching I (2004). Zoological journal of Linnean Society. Phylogeny and classification of Aedinii (Diptera: Culicidae), based on morphological characters of all life stages. 142: 289-368.

Reuben R, Tewari SC, Hiriyan J and Akiyama J (1994). Mosquito Systematics .Illustrated keys to species of *Culex (Culex)* associated with Japanese Encephalitis in Southeast Asia (Diptera: Culicidae). 26(2): 75-96.

Srivanakarn S (1976). Contributions of American Entomological Institute. A revision of subgenus *Culex* in the Oriental region (Diptera: Culicidae). 9(1): 1-109.

Tyagi BK, Munirathinam A and Venkatesh A (2015). International Journal of Mosquito Research. A catalogue of Indian mosquitoes. 2(2): 50-97.

Wattal BL and Kalra NL (1961).Bull Nat Soc Ind Mal Mosq Dis. Regionwise pictorial keys to the female Indian *Anopheles* 9: 85-138.