

## INTERCROPPING IMPACT AGAINST THE DIVERSITY OF MESOSTIGMATID MITES IN CITRUS SOILS OF PUNJAB, PAKISTAN

Abdul Ghaffar<sup>1</sup>, Muhammad Hamid Bashir<sup>1,\*</sup>, Bilal Saeed Khan<sup>1</sup> and Nazir Javed<sup>2</sup>

<sup>1</sup>Department of Entomology University of Agriculture, Faisalabad, Pakistan; <sup>2</sup>Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan.

\*Correspondence author's e-mail: ghaffar323@gmail.com

The effect of intercropping on the diversity of mesostigmatid soil mites was investigated in three types of citrus orchards of four selected districts in replicated field experiment. Maximum Shannon diversity ( $H' = 2.11$ ) was recorded from citrus + barseem intercropped orchards while minimum ( $H' = 2.03$ ) from citrus orchards without intercropping. Maximum mean abundance and richness of Mesostigmata ( $9.22 \pm 0.57$ ,  $S = 4.40 \pm 0.18$ ) was reported from citrus orchards without intercropping while minimum values for abundance and richness ( $7.46 \pm 0.42$ ,  $S = 4.06 \pm 0.17$ ) from citrus + wheat intercropped orchards. Community structure of mites showed that Ameroseiidae, Ascidiidae, Laelapidae, Pachylaelapidae was found maximum from citrus orchards without intercropping Melicheridae and Phytoseiidae from citrus + wheat intercropped while, Parasitidae, Sejidae and Uropodidae was recorded most abundant from citrus + barseem intercropped orchards.

**Keywords:** Inhabiting mites, arthropods pests, ground cover, citrus orchards, Shannon diversity.

### INTRODUCTION

Mesostigmata comprises of important and large component of soil inhabiting mites. Being predators their direct contribution to soil is still unclear but indirectly these mites have key role in managing the population of soil dwelling arthropods and nematodes (Krantz, 1983; Gerson *et al.*, 2003; Arjomandi *et al.*, 2013).

Intercropping as one form of polyculture that being used by indigenous peoples throughout the world (Altieri, 1991). Ground cover plants are identified as a potential source of natural enemies of arthropods pests in citrus (Liang and Huang, 1994). However, the composition, structure and management of the ground cover plants have a significant influence against the diversity of soil arthropod and their total and relative densities within soils (Kogan, 1981; Bugg and Waddington, 1994).

Mesostigmatid mites can inhabit a broad range of habitats. Yet, they are known to show a negative association with the physical and chemical disturbances and low levels of organic matter that normally occur in agricultural soils (Petersen and Luxton, 1982; El Titi, 1984; Siepel and Van de Bund, 1988). After completion of land preparation activities, organic matter is expected to increase gradually, and diversity of soil mite is improved. Fields with less tillage practices along with application of organic matter show relatively high diversity of Mesostigmata group (Twardowski, 2006). According to different studies it is showed that intensive agricultural practices have great impact on the abundance and diversity of soil arthropods (Tsiafouli *et al.*, 2015). It is observed that below ground diversity is highly effected due to high use of

inputs. Conversion of natural ecosystem to agro-ecosystem is believed to be cause of lowering of soil carbon that ultimately modify soil inhabiting arthropods. Consequently, the agricultural production is affected due to variations in biodiversity of underground arthropods so agricultural practices like tillage, use of agrochemicals, drainage, irrigation, burning etc. have drastic impact on diversity of soil arthropods including Mesostigmata (El-Banhawy *et al.*, 1997, 1998; Berch *et al.*, 2007). Continuous cultivation provides short period for Mesostigmata to recover themselves after cultivation (Murphy and Jalil, 1964; Webb, 1977; Stamou and Asikids, 1992). Un-cultivated soils are the major source of accumulation for soil mesofauna and serve as sanctuaries resulting in more colonization of Mesostigmata than disturbed one (Behan-Pelletier, 1999).

Soil arthropods community is highly affected due to human activities and constant stress (Lowrance *et al.*, 1984). Human intervention like hoeing, ploughing mostly destroy the habitat and impose effect on soil inhabiting arthropods (Altieri, 1991). However, information about the response of microarthropods, known to be very sensitive to changes in soil (Andre *et al.*, 1997; Giller *et al.*, 1997). Species richness, abundance, and community composition of arthropods respond to human habitat modification, and these effects were in part reported to be mediated by environmental habitat variables (Noti *et al.*, 2003; Ducarme *et al.*, 2004; Dexter, 2004; Lipiec *et al.*, 2006; Sinclair and Stevens, 2006; Morris *et al.*, 2007; Bokhorst, 2008; Greenwood *et al.*, 2011). Tree plantation comprised of monoculture plant pose great impact on the biodiversity due to structure, age and vertical movement of tree root system and effect of above factors on

the surrounding microhabitat under canopy (Zerbe and Wirth, 2006; Knight *et al.*, 2008; Mueller *et al.*, 2012) also affect and modify the micro climate under canopy and these factors affect the soil dwelling arthropods diversity due to presence of coarse woody debris, litter layers. Vascular plants require such type of microhabitat (Zerbe *et al.*, 2007; Juutilainen *et al.*, 2014; Jagodzinski *et al.*, 2018; Wierzcholska *et al.*, 2018). The aim of current study will be to quantify the effect of intercropping on diversity of Mesostigmata and functioning of crop-soil interaction within citrus orchards. Till now, no work has been done from Pakistan on this topic of study.

## MATERIALS AND METHODS

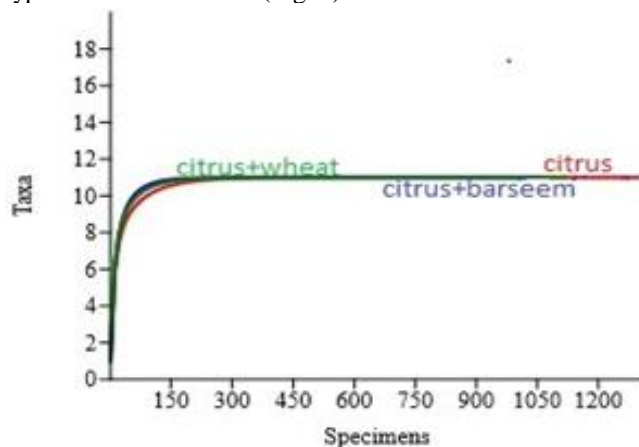
Diversity of soil inhabiting Mesostigmata was studied in three types of citrus orchards which were selected at Faisalabad, Sargodha, Toba Tek Singh and Layyah for this purpose. These orchards were selected by taking almost uniform plant age and agronomic practices in the selected areas. Soil samples from each citrus orchard were taken at monthly interval with the help of soil sampler of diameter 10.5 cm and height 12 cm. The samples were placed in zip lock polyethene bags, kept in cool place and brought to Acarology Laboratory, University of Agriculture, Faisalabad, at the earliest to avoid desiccation of soil samples. Soil mites were isolated with the help of modified Berlese Tullgren funnels apparatus Glass vials having 75% alcohol along with few drops of glycerine were used for preservation of soil mites. Vials were labelled according to date, place and type of orchards separately. Collected mite specimen of Mesostigmata were sorted from rest of soil fauna under stereoscope and mounted permanently on glass slides by using Hoyer's medium. Specimens were studied under high power phase contrast microscope and identified up to family level by using published taxonomic key of soil mites by Evans and Till (1979).

**Statistical analysis:** The individual base rarefaction curved were calculated with help of computer software 'PAST' (Hammer *et al.*, 2001). Abundance and family richness for each types of orchard were also computed. Shannon-Wiener diversity index was calculated and diversity of soil mites per sample was found. Biodiversity analysis was done by using Shannon diversity index (Shannon, 1948) for estimation of abundance and richness of Mesostigmata mites from each type of citrus orchard. Chao 1 diversity index for mesostigmatid soil mites was also calculated to evaluate the richness of soil mites and to compute the no of missing families due to sampling methods (Colwell, 2012). Fisher alpha diversity for each orchard was also calculated. The values were compared using T test at significance level  $\alpha=0.05$  with the help of 'R' software.

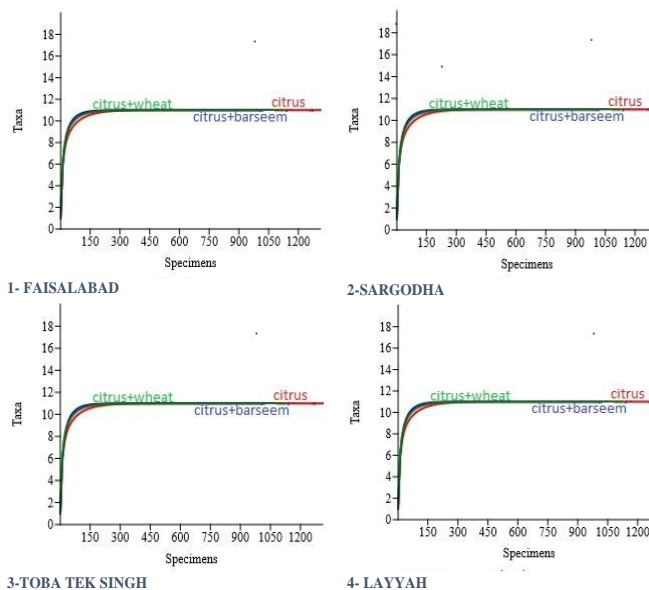
## RESULTS

Soil sampling was done from all selected orchards for one

year and 11250 soil inhabiting mites were collected. Out of this fauna mesostigmatid soil mites were 3431 belonging to 11 families. Individual based cumulative rarefaction curve indicated that sampling was enough as standard rarefaction curve was obtained. Maximum taxa of mesostigmatid soil mites was recorded due to soil sampling of citrus orchards. Similarly, rarefaction curves as a result of soil sampling explained the sufficiency of collected samples in all three types of citrus orchards (Fig. 1)



**Figure 1. Rarefaction curve for cumulative mesostigmatid taxa of citrus orchards.**



**Figure 2. District wise Rarefaction curve for mesostigmatid taxa for three types of citrus orchard.**

Shannon diversity index values showed slight variations among different types of citrus orchards and maximum Shannon diversity index value ( $H' = 2.11$ ) was reported for citrus + Barseem orchards while minimum value ( $H' = 2.03$ ) for citrus orchards without intercropping. Fisher's Alpha

diversity also varied slightly and maximum value (1.71) was reported for citrus + wheat orchards and minimum (1.64) for citrus orchards without intercropping (Table 1). Chao 1 diversity index showed similar results and no variation as all types of citrus orchards showed same number (11) of families (Table 2).

**Table 1. Overall Shannon diversity and Fisher's alpha index of Mesostigmata from citrus orchards.**

Orchards	Shannon diversity	Fisher's alpha
Citrus	2.03	1.64
Citrus + Wheat	2.09	1.71
Citrus + Barseem	2.11	1.69

**Table 2. Chao1 diversity index of Mesostigmata from citrus orchards.**

Orchards	S.obs	S.chao1	se.chao1
Citrus	11	11	0
Citrus + Wheat	11	11	0
Citrus + Barseem	11	11	0

Data regarding abundance of mesostigmatid mites from different types of citrus orchards showed highly significant variations in all types of citrus orchards ( $F_{\text{value}}=9.04$ ;  $P\leq 0.000$ ) and maximum abundance ( $9.22\pm 0.57$ ) was for citrus orchards (without intercropping) and minimum ( $7.46\pm 0.42$ ) was reported for citrus + wheat intercropped orchards while citrus + barseem intercropped orchard showed  $7.86\pm 0.48$ . Highly significant variations ( $F_{\text{value}}=2.09$ ;  $P\leq 0.003$ ) were recorded due to interaction of citrus orchards with months for abundance of mesostigmatid mites. Maximum abundance ( $15.75\pm 1.88$ ) was reported from citrus orchards without intercropping during April while minimum ( $3.5\pm 0.87$ ) was observed during October. Citrus + wheat intercropped orchards showed the maximum abundance ( $12.42\pm 1.73$ )

during March and minimum ( $4.25\pm 0.76$ ) was observed during December. Maximum abundance ( $12.08\pm 1.35$ ) was recorded from citrus+barseem orchards during April and minimum ( $4.25\pm 0.72$ ) during November (Table 3).

Data regarding interaction of district and orchard types also exhibited great variation for abundance of mesostigmatid mites. Maximum abundance ( $11.08\pm 1.16$ ) for citrus orchards without intercropping was reported from Layyah and minimum ( $7.97\pm 1.04$ ) from Toba Tek Singh district. Citrus + wheat intercropped orchards showed maximum abundance ( $8.58\pm 0.76$ ) from Layyah and minimum ( $6.64\pm 1.12$ ) from Sargodha district while citrus+barseem intercropped orchards also showed maximum ( $9.33\pm 0.75$ ) abundance from Layyah and minimum ( $6.64\pm 0.73$ ) from Sargodha district (Table 4).

**Table 4. Abundance of mesostigmatid mites (District x Orchard type).**

Districts	Citrus	Citrus + Wheat	Citrus + Barseem
Faisalabad	$8.53\pm 1.02a$	$7.22\pm 0.87A$	$8.33\pm 1.06a$
Layyah	$11.08\pm 1.16a$	$8.58\pm 0.76B$	$9.33\pm 1.12b$
Sargodha	$9.31\pm 1.26a$	$6.64\pm 0.75B$	$6.64\pm 0.73b$
Toba Tek Singh	$7.97\pm 1.04a$	$7.39\pm 0.93A$	$7.14\pm 0.83a$

Means sharing similar letters are non-significant ( $P\geq 0.05$ ); Small letters in each column represent differences at each orchard type within each district.

Data of richness of mesostigmatid mites from different orchards also showed highly significant differences ( $F_{\text{value}}=3.66$ ;  $P\leq 0.027$ ) and maximum richness ( $4.40\pm 0.18$ ) of Mesostigmata was for citrus orchards (without intercropping) and minimum ( $4.06\pm 0.17$ ) was reported for citrus + wheat intercropped orchards while citrus + barseem intercropped orchard showed  $4.13\pm 0.17$ . Highly significant variations ( $F_{\text{value}}=1.725$ ;  $P\leq 0.0244$ ) were recorded for richness due to interaction of types of orchards and months for mesostigmatid

**Table 3. Abundance of mesostigmatid mites (Month X Orchard Type).**

Months	Citrus	Citrus + Wheat	Citrus + Barseem	Average
January	$13.08\pm 2.43ab$	$6.83\pm 1.10cde$	$8.08\pm 1.52bcd$	$9.33\pm 1.09A$
February	$8.42\pm 1.79c$	$7.83\pm 1.24bc$	$10.50\pm 2.66abc$	$8.92\pm 1.13BC$
March	$13.50\pm 1.91ab$	$12.42\pm 1.73a$	$10.83\pm 2.33ab$	$12.25\pm 1.14A$
April	$15.75\pm 1.88a$	$10.92\pm 1.70a$	$12.08\pm 1.35a$	$12.92\pm 0.99A$
May	$8.08\pm 2.26c$	$7.25\pm 1.58bcd$	$7.25\pm 1.46de$	$7.53\pm 1.01C$
June	$14.83\pm 1.49a$	$10.08\pm 1.32ab$	$9.08\pm 1.80bcd$	$11.33\pm 0.97A$
July	$9.33\pm 1.76c$	$7.92\pm 1.59bc$	$8.17\pm 1.91bcd$	$8.47\pm 0.99BC$
August	$5.00\pm 0.81d$	$5.25\pm 1.18cde$	$6.83\pm 0.85def$	$5.69\pm 0.55D$
September	$10.92\pm 1.08bc$	$7.92\pm 0.65bc$	$7.58\pm 1.05cde$	$8.81\pm 0.59BC$
October	$3.50\pm 0.87d$	$4.50\pm 1.05de$	$4.67\pm 0.85ef$	$4.22\pm 0.53D$
November	$3.67\pm 0.87d$	$4.33\pm 0.93de$	$4.25\pm 0.72f$	$4.08\pm 0.48D$
December	$4.58\pm 0.54d$	$4.25\pm 0.76E$	$5.00\pm 0.82ef$	$4.61\pm 0.40D$
Overall Mean	$9.22\pm 0.57A$	$7.46\pm 0.42B$	$7.86\pm 0.48B$	

Means sharing similar letters are non-significant ( $P\geq 0.05$ ); Small letters in each column represent differences between months at each Locality while capital letters in the last column represent monthwise difference in all localities, capital letters in last row, represent overall difference in each locality.

**Table 5. Richness of mesostigmatid mites (Month X Orchard type).**

Month	Citrus	Citrus + Wheat	Citrus + Barseem	Average
January	5.17±0.46cd	4.33±0.51cde	4.17±0.53bc	
February	4.17±0.61def	4.33±0.56cde	4.92±0.54ab	4.56±0.29B
March	5.50±0.45bc	5.92±0.38a	4.83±0.66ab	4.47±0.32B
April	6.50±0.53ab	5.50±0.62ab	5.83±0.51a	5.42±0.30A
May	4.25±0.64de	4.25±0.71cde	4.08±0.61bc	5.94±0.32A
June	6.67±0.54a	5.17±0.49abc	4.42±0.69b	4.19±0.37BC
July	3.67±0.48ef	4.00±0.56de	3.33±0.56cd	5.42±0.36A
August	3.58±0.50ef	3.33±0.71ef	4.17±0.42bc	3.67±0.30CD
September	5.08±0.36cd	4.50±0.29bcd	4.42±0.47b	3.69±0.32CD
October	2.50±0.50g	2.42±0.47f	2.67±0.45d	4.67±0.22B
November	2.50±0.53g	2.92±0.54f	2.67±0.36d	2.53±0.27E
December	3.17±0.24fg	2.92±0.47f	3.17±0.46cd	2.69±0.27E
Overall Means	4.40±0.18A	4.13±0.17AB	4.06±0.17B	

Means sharing similar letters are non-significant ( $P \geq 0.05$ ); Small letters in each column represent differences between months at each Locality while capital letters in the last column represent monthwise difference in all localities, capital letters in last row, represent overall difference in each locality.

mites. Maximum richness ( $S=5.94 \pm 0.32$ ) during April and minimum richness ( $S=2.53 \pm 0.27$ ) was reported during October. Citrus orchards without intercropping showed maximum richness ( $S=6.67 \pm 0.54$ ) during June while minimum ( $S=2.50 \pm 0.50$ ) was observed during October. Citrus + wheat intercropped orchards showed the maximum richness ( $S=5.92 \pm 0.38$ ) during March and minimum ( $S=2.42 \pm 0.47$ ) was observed during October. Maximum richness ( $S=5.83 \pm 0.51$ ) was recorded from citrus+barseem orchards during April and minimum ( $S=2.67 \pm 0.36$ ) during November (Table 5).

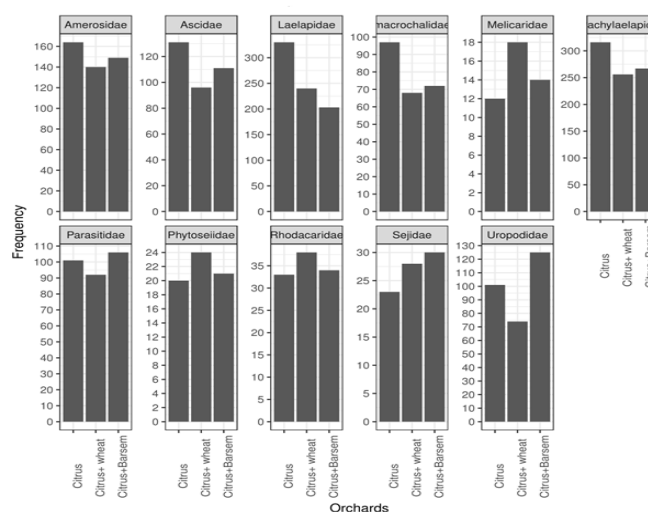
Data regarding interaction of district and orchard types also exhibited great variation for richness of mesostigmatid mites. Maximum richness ( $S=5.08 \pm 0.29$ ) for citrus orchards without intercropping was reported from Layyah and minimum ( $S=4.08 \pm 0.37$ ) from Toba Tek Singh district, Citrus + wheat intercropped orchards showed maximum richness ( $4.72 \pm 0.27$ ) from Layyah and minimum ( $S=3.69 \pm 0.35$ ) from Sargodha district while citrus+barseem intercropped orchards also showed maximum ( $S=4.44 \pm 0.41$ ) richness from Faisalabad and minim ( $S=3.72 \pm 0.29$ ) from Sargodha district (Table 6).

**Table 6. Richness of mesostigmatid mites (District X Orchard type) Citrus Citrus + Wheat Citrus + Barseem.**

Districts	Citrus	Citrus + Wheat	Citrus + Barseem
Faisalabad	4.19±0.33a	4.03±0.38a	4.44±0.41a
Layyah	5.08±0.29a	4.72±0.27ab	4.28±0.33a
Sargodha	4.22±0.42a	3.69±0.35a	3.72±0.29a
Toba Tek Singh	4.08±0.37a	4.08±0.36a	3.78±0.28a

Means sharing similar letters are non-significant ( $P \geq 0.05$ ); Small letters in each column represent differences at each orchard type within each district.

Community structure for different types of citrus orchards showed variation for incidence of different families of mesostigmata as Ameroseiidae was recorded maximum from citrus (without intercropping) orchards Melicheridae was found maximum from citrus + wheat intercropped orchards and minimum from citrus (without intercropping) orchards. Uropodidae and Parasitidae was found maximum from citrus + barseem intercropped orchards and minimum from citrus + wheat intercropped orchards while Phytoseiidae and Rhodacaridae was recorded maximum from citrus + wheat intercropped citrus orchards and minimum from citrus (without intercropping) orchards. Similarly, Sejidae was observed maximum from citrus + barseem intercropped orchards and minimum was reported from citrus (without intercropping (Fig. 3).



**Figure 3. Community structure of mesostigmatid mites from different types of citrus orchards of Punjab, Pakistan.**

## DISCUSSION

We found a total of 3431 mesostigmatid mites, belonging to 11 families, from soil samples for whole years and from three types of selected citrus orchards of four districts of Punjab. Similarly, El-Banhawy *et al.* (2006) reported sixteen species of predacious soil mites of six families from citrus orchards of Nile Delta, Egypt. Navarro-Campos *et al.* (2012) reported fifteen species of eight families from Spain, Imen *et al.* (2018) also recorded nineteen species of ten families of Mesostigmata from citrus orchard soils of Tunisia. Mesostigmata are commonly found from all types of citrus orchards due to application of farmyard manure and irrigation with canal water, pivotal for the improvement of soil quality of citrus orchards. Shannon diversity index ( $H'$ ) varied slightly ( $H'=2.03-2.11$ ) among different type of intercropped citrus orchards and maximum Shannon diversity ( $H'=2.11$ ) was reported from citrus + barseem intercropped orchard while minimum ( $H'=2.03$ ) from citrus orchards without intercropping. These findings are closely resembling with results of Karg (1986) and El-Banhawy *et al.* (2006) who indicated that mesostigmatid mites are commonly found from citrus orchard enriched with organic matter as, citrus + barseem orchards receive more organic matter than other two orchards and no cultivation disturbance after sowing (October) to harvesting (May-June) in Punjab. Abundance of Mesostigmata showed great variations as maximum abundance ( $15.75\pm1.88$ ) was reported from citrus orchards without intercropping during April while minimum ( $3.5\pm0.87$ ) was observed during October. These findings are closely related with results of Imen *et al.* (2018) as cultivation showed short effect on soil inhabiting mites and showed variations. Interaction of orchard type and districts showed slight difference ( $11.08\pm1.16$  to  $6.64\pm0.73$ ) as described by El-Banhawy *et al.* (2006), Wissuwa *et al.* (2012) and Khan *et al.* (2017). This variation is due to change of soil type of each locality. Richness of mesostigmatid soil mites also showed significant variations among different types of citrus orchards ( $S=4.40\pm0.18$  to  $4.06\pm0.17$ ). Impact of intercropping on diversity of mesostigmatid mites is less documented and some workers reported that soil arthropods assemblage showed different response for overlying tree diversity like Badejo and Tian (1999), Hansen (2000) and Eissfeller *et al.* (2013). Similarly, significant richness variation was evidence during different months and interaction with orchard types like  $5.94\pm0.32$  during April to  $2.53\pm0.27$  during October. This variation is due to short term influence of cultivation over soil mites of citrus orchards being practiced during different months for intercropping. Same findings were made by Hulsman and Wolter (1998) who reported that variation is due to desiccation as a result of cultivation and exposure of soil mites to direct sunlight. Community structure of mesostigmatid mites showed significant variations for each type of citrus orchard as Ameroseiidae, Ascidae, Laelapidae,

Pachylaelapidae was found maximum from citrus orchards (without intercropping), Melicheridae and Phytoseiidae from citrus + wheat intercropped while, Parasitidae, Sejidae and Uropodidae was recorded from citrus + barseem intercropped orchards and same finding was reported by Hulsman and Wolter (1998).

**Conclusion:** It is concluded that citrus orchard without intercropping also showed maximum richness. Correlation of abiotic factors with abundance of different families of Mesostigmata showed that rainfall was positively correlated with Phytoseiidae and Melicheridae other families were negatively correlated. Correlation of soil parameters such as EC, P and K was Phytoseiidae, Rhodacaridae, Melicheridae and Sejidae other were positively and OC was negatively correlated.

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