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RESEARCH ARTICLE

Potential of oribatid mites in biodegradation and mineralization for enhancing plant productivity

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ABSTRACT: The degradation of litter is an essential process of the soil ecosystem leading to nutrient cycling and is mediated by a heterogeneous group of soil organisms. Oribatid mites represent one of the predominant agents of litter biodegradation in the soil. The ubiquitous presence and extensive diversity of this group of mites make them integral to the process of mineralization of litter in almost all types of soil ecosystems. However, an overall assessment of the mineralization potential of different groups of oribatid mites depicts the relative advantage of lower groups of oribatids, namely the lohmannoid and phthiracaroid members, in the degradation of leafy and woody elements of litter. Degradation of such complex materials primarily necessitates additional qualities such as strong and well developed oral and holding appendages, and the presence of the necessary enteric microflora and associated enzymes, for on-going degradation. In-depth field and laboratory studies of two representative species of the above two groups of mites, viz. *Atropacarus* (*Hoplophorella*) *chaliyamensis* Haq and Xavier, 2005 and *Heptacarus hirsutus* Wallwork, 1964, with the vegetable crop *Vigna unguiculata*, clearly demonstrated that these species make a remarkable contribution to the process of nutrient cycling. The combined feeding activity of these two species on the woody elements of litter was found to enhance the release of nitrogen, phosphorous and potassium, as evidenced by the increased concentration of these minerals in fecal pellets. However, a decrease in the levels of calcium and magnesium was observed in the fecal pellets. The decrease in calcium may be accounted for by its immediate requirement in body maintenance. The impact of oriculture on plant productivity was evidenced through increased plant growth, higher yield and better quality of the pods produced by the treated plants.

Keywords: Oribatid mites, biodegradation, nutrient cycling, fertility, plant productivity, oriculture.

INTRODUCTION

Oribatid mites constitute one of the suborders of Acari and are well known for their taxonomic diversity, wide geographic distribution and adaptive radiation in terms of habitats. Subias (2018) compiled a list of more than 11,000 species of oribatid mites described so far. These mites enjoy cosmopolitan distribution with varying abundance across different geographic realms (Gergócs and Hufnagel, 2015). They are predominant in soil ecosystems but have invaded many of the terrestrial habitats and even aquatic habitats (Maraun et al., 2007; Schatz and Behan-Pelletier, 2008; Norton and Behan-Pelletier, 2009; Murvanidze et al., 2016; Behan-Pelletier and Norton, 2016). The majority of the soil oribatids feed on plant litter, fungi, and algae (Behan-Pelletier and Norton, 2016). Varied food preference has been noticed within the litter feeding oribatid mites in the soil and has been investigated extensively for a better understanding of the extent of contribution of these mites to the functioning of the edaphic community (Macfadyen, 1961; Haq, 1994).

Oribatid mites exhibit profound diversity in the soil ecosystem. They have invaded a wide range of microhabitats and adopted an array of nutritional habits. The items of food they ingest range from bacteria, algae, and fungi to leafy and woody materials in the plant litter and to the wastes of living animals and dead remains of animals. Accordingly, they have been grouped into three major feeding guilds, namely the microphytophages, macrophytophages and panphytophages (Schuster, 1956; Hartenstein, 1962). Luxton (1972) further sub-categorized them into a still narrower range of feeding guilds and assigned many more categories to accommodate marginal species showing deviations from the major food and feeding habits.

Jacot's findings (1930, 1936, 1939) on the nutritional habits of oribatid mites were instrumental to the assumption that some species of these mites are specific in their food requirements. Furthermore, Forsslund (1938), Gourbiere et al. (1985) and Lions and Gourbiere (1988) showed that some species of these mites specifically feed on fungal hyphae and spores whereas others live mainly on leaf and needle litter. Harding and Stuttard (1974) stated that the food spectrum of oribatid mites includes algae, lichens, mosses, and pollen. Riha (1951) showed the affinity of oribatid mites for dead collembolans and worms. Rockett and Woodring (1966), Muraoka and Ishibashi (1976), Rockett (1980) and Stefaniak and Seniczak (1981) reported feeding of food items mentioned in the above categories by various specious of oribatid mites. Accordingly, mites have been grouped into three major feeding guilds, namely the microphytophages, macrophytophages and panphytophages. However, these guilds are not sufficient

to accommodate many other feeding habits observed among them. There has been tremendous progress in the knowledge of feeding guilds in oribatid mites over the years. Zinkler (1971), Luxton (1972, 1979) and Urbasek and Stary (1994) have contributed knowledge on the enzyme activity of these mites. Haq (1984) demonstrated the role of microbes in the nutrition of *Heptacarus hirsutus* and Haq (1987) reported on the biodegradation of cellulose in the gut of the same species. This has provided greater insight into the possible relationships between the functional aspects of feeding trends of mites and their ability to biodegrade plant structural polysaccharides. The feeding guilds of oribatid mites are related to the carbohydrase activities of these mites (Siepel and Dijkman 1993). Additional aspects of nutritional biology were reviewed later (Schneider et al. 2004). The functional attributes of oribatid mites in the soil ecosystem were reviewed by Haq (1994, 1996), and the contribution of these mites towards the degradation of plant litter and nutrient recycling (Fig. 1) and maintenance of soil fertility were identified as major roles being played by these mites in the soil (Haq, 2016).

Among the Oribatida, members of the lower taxa like the lohmannoid and phthiracaroid groups have been observed to be more competent in the process of degradation of the residues of higher plants in the litter and the mineralization process through a series of studies on different aspects of the nutritional biology of various groups of oribatid mites (Haq and Prabhoo, 1976; Haq, 1976, 1982, 1992, 1994, 1996, 2007a; Haq and Konikkara, 1988; Haq and Xavier, 2005). The current study was carried out to analyze the practical impacts of these groups of mites in biodegradation, mineralization, nutrient cycling and the enhancement of soil fertility. Two species of mites, namely *Atropacarus* (*Hoplophorella*) *chaliyamensis* Haq and Xavier (2005) representing Phthiracaridae, and *H. hirsutus* Wallwork, 1964 representing Lohmanniidae were selected as the model organisms for the current study. The ability of these two species for wood degradation has been repeatedly established through earlier studies of the author (Haq, 1987, 2016). Specifically, the current work is undertaken to determine the enhancement of soil fertility by these mites in the growth and yield of the vegetable plant, *Vigna unguiculata*.

MATERIAL AND METHODS

Sampling, extraction and identification of mites

The mites for the current study, *A.* (*H.*) *chaliyamensis* of the family Phthiracaridae and *H. hirsutus* belonging to the family Lohmanniidae, were collected from 4 different locations on Beypore and Chaliyam beaches (Figs 2-3), near Calicut University Campus, Malappuram District, Kerala, India. These two species of mites were abundantly available on driftwood, logs, leaf and woody litter samples on the beach shore (Figs 4-6). The extraction of mites from the above litter samples were carried out via the technique of Berlese's Funnel Apparatus (1905) and the Open Brass Funnel Apparatus designed by Haq and Ramani (2002) (Fig. 7). The former species was erected as a new species by Haq and Xavier (2005) and the latter species were identified by comparison with the characters given in the original description provided by Wallwork (1964). No other specific terminologies have been used here.

Nutritional Biology

Analysis of gut content

Live individuals of the selected species of mites were collected from the beaches and subjected for gut content analysis following the protocol described in Haq and Prabhoo (1976) and Haq (1982, 2007b).

Analysis of laboratory feeding

Live mites collected from the beaches were reared in the laboratory in plastic culture vessels and their feeding preferences were screened, as described by Haq and Pabhoo (1976) and Haq (2007a).

Analysis of the morphology of mouthparts

The mouthparts of the mites were dissected from collected specimens, processed and examined according to the methods illustrated by the author (Haq, 2007a).

Analysis of gut enzymes

A qualitative profile of the enzyme contents of the mites was determined according to the protocol described by the author (Haq, 1984, 1987, 2007a).

Analysis of gut microbiota

The gut microbiota of the mites was investigated by inoculating the freshly deposited fecal material of the mites in basic culture media for the bacteria and fungi as described by Haq (1984, 2007a) and Haq and Konikkara (1988).

Biomass Reduction

A known weight of driftwood pieces obtained from the two seashores was offered to 10 adult mites of both *A.* (*H.*) *chaliyamensis* and *H. hirsutus* in separate laboratory culture vessels and the mites were kept under observation at standard laboratory conditions for one month, providing the same quantity of the same food material on completion of feeding on each batch of food material offered. The number of days utilized for completion of every batch of food and the final mass of fecal pellets generated from each batch of food were recorded. The average reduction in the quantity of biomass observed was calculated from the data obtained.

Nutrient Analysis

In order to assess the nutrient turnover effected through the feeding activity of the mites on plant litter, the levels of nitrogen, phosphorous, potassium, calcium, and magnesium were estimated in decomposing wood samples and fecal pellets produced by the mites after

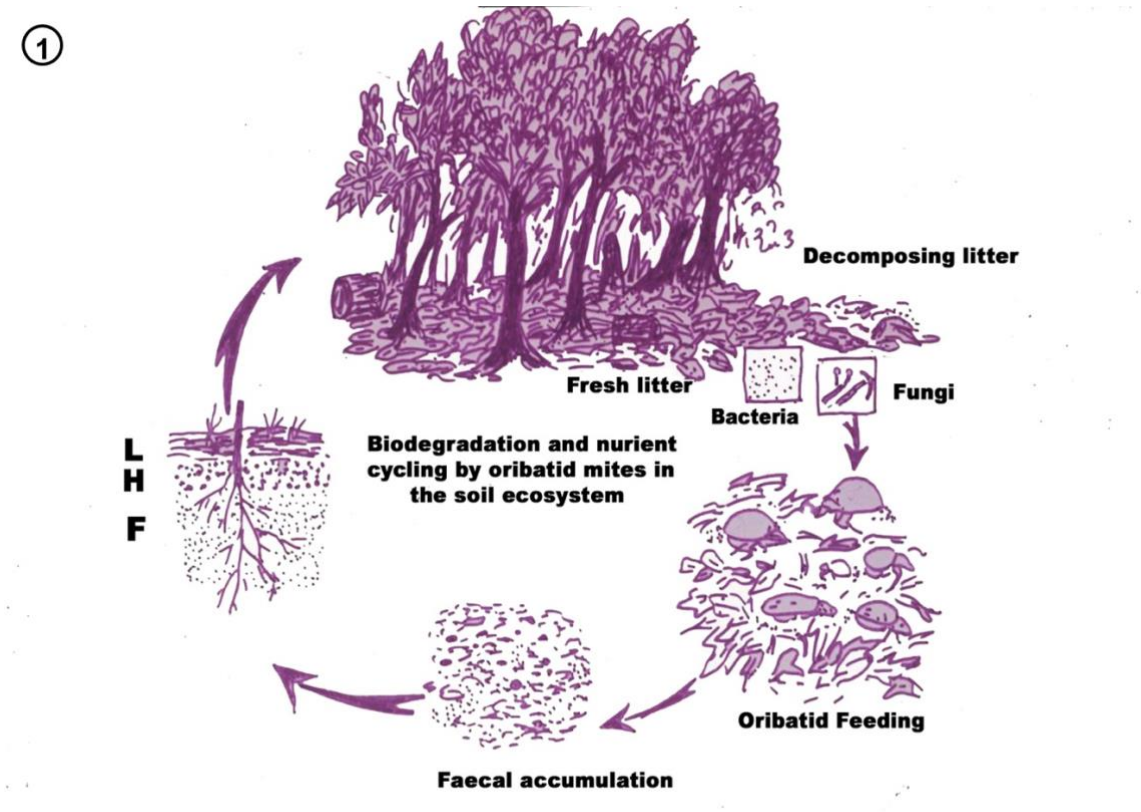

Figure 1. Nutrient cycling.

Figure 2. Beypore Beach with plant litter accumulation on shore.

Figure 3. Chaliyam beach with drift wood and litter accumulation.

Figure 4. Logs of drift wood – A,C) Chaliyam beach, B,D) Beypore Beach.

Figure 5. Samples of various stages of decomposing litter (A-D) used as source for extraction of the mites.

Figure 6. Degraded leafy and woody litter maintained in the field and the laboratory (A-D) for mass culturing of the mites.

Figure 7. Open Brass Funnel Apparatus used for extraction of mites (Haq and Ramani 2002).

Figure 8. Field trial plot of oriculture with *Vigna unguiculata* plants, A) Experimental and control plants after two weeks of growth, B) Experimental and control plants after four weeks of growth, C) Experimental and control plants after eight weeks of growth, D) Experimental plants with matured pods.

feeding, as per the methods elaborated by Haq (1996, 2007a).

Breeding Biology

The mites were reared in individual culture chambers with a Plaster of Paris-charcoal base. They were offered driftwood pieces, as this food was ideal for supporting the development of these mites (Haq, 1987). The cultures were maintained at 27±2°C and relative humidity of 80-82%. The cultures were monitored daily for detection of spermatophores and eggs. As and when the eggs were found, the pieces of wood with eggs were transferred to specially prepared culture vessels comprised of glass rings fixed on a Plaster of Paris-charcoal base in a Petri dish. Cover slips were used as lids for the individual culture cells within the Petri dish. Life history studies of the mites and their individual stages of development were undertaken with their preferred foods and monitored using appropriate microscopes.

RESUTLS

Nutritional profiles

The laboratory feeding experiment showed similar nutritional profiles for both species, including gut content analysis (Table 1). In general, the phthiracarid, *A.* (*H.*) *chaliyamensis* showed a preference towards the woody tissues of higher plants, except for occasional feeding on leaf litter. *A*. (*H*.) *chaliyamensis* showed a wood boring habit (Figs 10 C, 11 D), as the adults often tunnelled into the wood pieces offered as food and laid eggs inside the tunnels. The lohmanniid mite, *H. hirsutus* exhibited a general preference towards high cellulose content litter like the driftwood from the beach and midribs and the veins of dried leaves. The larvae and nymphs often remained within the tunnels (Figs 14 A,B) which were often filled with the fecal material of the mite. The feeding activity was initiated with the soft mesophyll tissue, followed by the veins and the midrib region. This type of feeding often resulted in the appearance of skeletonized leaves in the cultures. When offered the dried roots of *Calotropis gigantea*, the mites bored holes into the root tissues and created tunnels in the woody parts of the roots. Macrophytophagous phthiracarid mites primarily depend on woody tissues and hence are designated as xylophages. The chelicerae of *A*. (*H*.) *chaliyamensis* were broad and stout with denticulate body (Fig. 16 C). They possess well developed and highly sclerotized chelae. The movable and fixed digits of the chelicerae carried 4-5 teeth to help cut the hard wood pieces while feeding on them. The rutellar dendites bear dorsal concavities called vestibules, to accommodate large wood pieces for the masticatory action of the chelicerae of *A*. (*H*.) *chaliyamensis*. The chelicerae of *H*. *hirsutus* were less broad, elongated or with round base (Fig. 16 D). The movable digit had 3-4 teeth and the fixed digit had 2-3. This helped in the cutting of food particles. The rutella had a broader distal end and a narrow proximal end bearing 3-4 notches distally. It appeared flat, more or less triangular in shape and strongly sclerotized.

Gut enzyme profile

Qualitative analysis of the gut enzymes of the mites is presented in Table 1. The carbohydrate digesting enzymes, maltase, cellulase and cellobiase, indicated their ability to digest simple as well as complex carbohydrates.

Gut microbiome

Microbial assay of the fecal materials of the mites confirmed the occurrence of different forms of bacteria in the gut, as indicated in Table 1. Both the mite species harbored Gram+ and Gram- bacteria in their gut, with microbial diversity greater in *H*. *hirsutus*.

Biomass reduction

Estimates of total biomass reduction in samples of selected litter items offered to the mites in the laboratory cultures are presented in Table 2. The percentage reduction in the biomass by *A.* (*H.*) *chaliyamensis* was greater.

Nutrient analysis

Quantitative changes in the concentration of N, P and K as a result of feeding by *A.* (*H.*) *chaliyamensis* and *H. hirsutus* are presented in Table 3. All three macronutrients increased in quantity after feeding and digestion. Therefore, feeding on and digestion of litter released the nutrients bound in the plant tissues.

Breeding Biology

Atropacarus (*Hoplophorella*) *chaliyamensis*

Adult females of *A.* (*H.*) *chaliyamensis* (Fig. 10 A) oviposited mostly inside the wood and rarely among the fecal pellets (Figs 10 B,D). The eggs were inserted into the wood channels and pits (Fig. 11 D) created by their feeding activities. The eggs were mostly solitary (Fig. 12 E) and seldom found in clusters but always within the area of their fecal material. The incubation period ranged from 4-5 days (Table 4). On the third day of incubation, a tiny black patch appeared within the egg shell. This patch darkened and became prominent afterwards. This was followed by splitting of the egg along the patch, releasing the larva. Soon after hatching, the larvae remained immobile for about two hours and afterwards showed signs of feeding and remained concealed within the wood piece. The larvae excavated the wood during their feeding activity. The active period of larva ranged from 4-5 days (Table 4). The physical activities of the larvae became restricted towards the end of this period. The larvae gradually became inactive, swollen in appearance, sluggish in habit and stopped feeding. Then they became immobile when entering the first quiescent phase. At the end of the 3-4 days of quiescent period (Table 4) the process of moulting lasted 2-3 hours. At the beginning of this process, a vertical slit appeared on the notogaster and increased in size. After about 15-30 minutes, the prodorsum came out. Up and down movements of the body resulted in the casting off of the exuvium.

The newly moulted nymph remained stationary for some time near the exuvium; after that, it moved away in search

Table 1. Evaluation of nutrition profiles of oribatid mites

Species	Lab Feeding	Gut content	Enzyme Profile	Gut Microbiome	Oviposition	Feeding category
A. (*H.*) *chaliyamensis*	Fungi/Algae: No Leaf: Yes Wood: Yes	Fungi/Algae:No Leaf: Yes Wood: Yes	Maltase Cellulose Cellobiase	Gram+: 3 Gram –: 2 Fungi: 0	Wood Dried roots	Macrophytophage: Xylophage
H. hirsutus	Fungi/Algae: No Leaf: Yes Wood: Yes	Fungi/Algae: No Leaf: Yes Wood: No	Maltase Cellulose Cellobiase	Gram+: 1 Gram –: 6 Fungi: 0	Wood Dried roots Leaf	Macrophytophage: Xylophage

Table 2. Biomass reduction of litter samples by feeding of oribatid mites under laboratory conditions

Mite species	Initial weight (mg)	Final weight (fecal pellets)* (mg)	% reduction of biomass*	No. of days utilized*
A. (*H.*) *chaliyamensis*#	10	9.12	8.8±0.421	2.2±0.105
H. hirsutus#	10	9.25	7.5±0.312	2.3±0.213

#Food offered: Drift wood
*Average of 5 triplicate samples

Table 3. Estimation of nutrient release from litter by feeding of oribatid mites

Nutrients analysed	% Increase	% Decrease
Nitrogen (N)	0.48 ±0.03	NA
Phosphorus (P)	0.06±0.12	NA
Potassium (K)	0.05±0.01	NA
Calcium (Ca)	NA	0.30±0.04
Magnesium (Mg)	NA	0.21±0.02

*Average of 5 triplicate samples

of food. The protonymph continued to be active for about 14-15 days and then entered the second quiescent phase which lasted for 3-4 days (Table 4).

Moulting of the second quiescent phase released the deutonymph. These nymphs excavated the wood during their feeding activity which resulted in the formation of irregular tunnels within the wood. The activity of the nymph could be easily distinguished by their fecal pellets, especially around the tunnels. The active period of the deutonymph ranged from 17-18 days after which it became quiescent. The third quiescent phase extended up to 4-5 days (Table 4) and terminated with the emergence of the tritonymph.

The tritonymph had an active period of 19-20 days, at the end of which it entered into the fourth quiescent phase. This lasted for 6-7 days and was followed by the emergence of the adult (Table 4). The newly moulted adults appeared pale yellow in colour with a light-pinkish tinge. The colour changed to wheat-yellow on the fourth or fifth day after emergence. Adults often wandered outside the wood tunnels (Figs 10 A, 11 A,B) whereas the immatures remained mostly inside (Figs 10 C, 11 C,D). The results of this study indicated that the development of *A.* (*H.*) *chaliyamensis* from egg to adult is completed within 81-89 days (Table 4).

Heptacarus hirsutus

The development of *H. hirsutus* (Figs 13 A,B) involved egg, larva and three nymphal stages interrupted by four quiescent stages between the successive larval stages to the adult stage. Gravid females of *H. hirsutus* generally carried 1 or 2 eggs at a time, which were visible through the integument under microscope. The female deposited solitary eggs on food material like leaf litter packed with fecal pellets. Most of the time the eggs were burried in tunnels bored into the wood. A longitudinal section of the wood tissue revealed the presence of all the life stages, well protected inside the tunnels (Figs 14 A,B). The number of eggs laid by a female under laboratory conditions varied between 6 and 8. However, the egg laying capacity of the adult female increased to 9-16 within one year on retaining the same culture conditions, including the rearing vessels. The incubation period ranged from 22-26 days (Table 5). About 13 days after oviposition, a conical projection appeared towards the animal pole of the egg. Gradually the colour of the egg turned light brown. Prior to hatching, a crescentic area developed along the antero-median portion of the egg, which appeared to be thin. This area gradually got stretched out due to the pressure exerted by the developing larva. As a result, the stretched area ruptured a little ahead of the middle and the slit extended in both directions. The larva stretched out its legs and crawled out through the slit. The egg case after emergence of the larva can be visible for 2-3 days with the half opened slit portion facing upward. The larva which emerged out of the egg appeared very sluggish. It continued to remain in the tunnel bored into the wood. Immediately after emergence, the larva remained motionless for about half an hour. This inactive period represented the hardening period. Then it gradually became active and started feeding on the woody tissue. Continued feeding by the larva led to the extension of the feeding tunnel. As feeding progressed, the tunnel became packed with fecal pellets and the larva subsequently selected an adjacent region of the wood for further feeding. This active period of larval life continued for about 16-18 days (Table 5). By this time, it had increased in size and attained a light brown colour. After the active period, it entered into the quiescent phase for 9-12 days (Table 5). During this time, it suspended all life activities and remained motionless. The quiescent phase ended with moulting. The slow moulting process required 2-4 hours. The body during this phase became slightly swollen and translucent. The postero-lateral region of the notogaster developed a few weakened areas on either side along which narrow slits appeared. Through one of these slits the last pair of legs protruded. The slit became extended in both directions due to the pressure exerted by the emerging individual. The moulting individual gradually came out with a backward thrust of its body. This helped to anchor the last pair of legs of the larva to the ground which allowed backward movement of the body and forward movement of the front pairs of legs simultaneously to push and release the moulting skin forward. Step by step the process finally resulted in the emergence of the protonymph from the larval quiescent stage. The process of moulting remained more or less the same in all the successive stages. Moulting of the first quiescent phase released the protonymph.

Unlike most of the other species of oribatid mites, *H. hirsutus* can multiply in large numbers in woody logs, particularly of marine nature. Availability of this preferred food in large quantities encourages the mite to bore deeply into the wood, creating numerous channels that accommodate sufficient colonies for further feeding activities. Feeding by the protonymph was more active. The individuals fed on the woody tissue for about 19-22 days, followed by the second quiescent phase which lasted for 9-11 days (Table 5). Moulting of the second quiescent phase gave rise to the deutonymph. The deutonymph continued to be active for 22-24 days, feeding voraciously on the wood after which it entered the third quiescent phase. The deutonymphal quiescence period lasted for 10-12 days (Table 5). It underwent moulting as in earlier phases, emerging as the tritonymph. The tritonymph, the largest among all nymphal stages, had an active life of 22-26 days. Active feeding by the tritonymph extended the tunnel deeper into the wood (Figs 14 A,B). There was active participation of the various stages of *H. hirsutus* in feeding and the final production of eggs by the matured females (Figs 14 C,D) in laboratory cultures. The fourth quiescent phase extended for 10-12 days (Table 5). The adult emerged after the moulting of the fourth quiescent phase. It appeared brown in colour and produced large numbers of fecal pellets (Fig.15 D). The total development from the egg to the adult stage ranged between 141-154 days for *H. hirsutus* (Table 5).

Field trial of soil fertility enhancement

The impact of oribatid mite feeding on plant materials enabled decomposition of these components and the increase in soil fertility was assessed by using a field experiment on *V. unguiculata* (Figs 8. A-D). Table 6 illustrates the influence of oribatid mite feeding on vegetative growth of the plant with reference to the increase in length of the plant. The rate of increase in plant length was much higher in the plants grown with oribatid mite exposure than the control. The length of pods was another parameter compared in the field trials. Table 7 shows the difference in pod length of the experimental and control plants. The average difference in pod length of the two sets of plants varied between 1.7 and 2 cm. Apart from this, feeding of preferential food items encourages better life activities rendering sufficient encouragement for reproductive potential. Such activity may increase the rate of degradation. This in turn could increase the rate of soil mineralisation also (Figs 9. A-H).

DISCUSSION

Soil is a unique and complex entity supporting the growth and development of all floral and faunal communities. It provides a holistic platform for the life activities of innumerable organisms including autotrophs and heterotrophs. The soil ecosystem acts as a reservoir for many materials, including old and dead components of flora and fauna being deposited continuously. These

Table 4. Postembryonic development of *A.* (*H.*) *chaliyamensis* under laboratory conditions

No	E	L	IQ	PN	IIQ	DN	IIIQ	TN	IVQ	Total
1	4	11	3	14	3	17	5	19	6	82
2	4	12	4	15	4	18	5	20	6	88
3	5	12	4	15	4	18	5	20	6	89
4	4	11	3	14	3	17	4	19	6	81
5	4	11	3	14	3	17	4	19	6	81
6	5	12	4	15	3	18	5	20	7	89
7	4	11	3	14	3	17	4	19	6	81
8	4	11	3	14	3	17	4	19	6	81
9	4	11	3	14	3	17	4	19	6	81
10	4	11	3	14	3	17	4	19	6	81
Range	4-5	11- 12	3-4	14-15	3-4	17-18	4-5	19-20	6-7	81-89

E: Egg, L: Larva, I-IVQ: Quiescent periods, PN: Protonymph, DN: Deutonymph, TN: Tritonymph

Table 5. Postembryonic development of *H. hirsutus* under laboratory conditions

No	E	L	IQ	PN	IIQ	DN	IIIQ	TN	IVQ	Total
1	22	16	9	19	9	22	10	26	11	144
2	23	16	9	19	9	22	10	22	11	141
3	23	16	9	19	10	24	10	22	10	146
4	26	16	11	21	9	22	12	25	10	152
5	23	18	9	19	11	24	12	26	12	154
6	23	18	9	19	11	22	10	22	11	145
7	25	16	12	22	9	22	10	22	11	149
8	23	17	12	21	10	22	10	24	11	150
9	23	16	9	19	9	23	10	26	10	145
10	22	16	9	19	9	22	12	22	11	142
Range	22-26	16-18	9-12	19-22	9-11	22-24	10-12	22-26	10-12	141-154

E: Egg, L: Larva, I-IVQ: Quiescent periods, PN: Protonymph, DN: Deutonymph, TN: Tritonymph

Table 6. Comparison of the length of *Vigna unguiculata* plants (in cm) grown under oriculture with control plants (up to 60 days age)

Age of plants in days	Control	Experimental	Difference in length (in cm)
10	55.2	62.6	7.4
20	61.0	96.8	35.0
30	72. 8	162.4	89.6
40	88.9	220.6	131.7
50	104.6	296.2	191.6
60	154. 2	324. 5	170.3

*Average of 10 plants

Table 7. Comparison of pod length (in cm of *Vigna unguiculata* in oriculture and control trials

No.	Experiment I	Control I	Difference in length	Experiment II	Control II	Difference in length
1	24	22	2	23	22	1
2	25	20	5	24	21	3
3	23	23	0	25	20	5
4	24	22	2	24	22	2
5	22	21	1	25	22	3
6	24	20	4	22	19	3
7	24	23	1	23	21	2
8	23	22	1	24	22	2
9	22	22	0	22	22	0
10	24	23	1	24	24	0
Average	23.5	21.8	1.7	23.6	21.6	2.0

structural elements of living organisms are a rich source of nutrients but in a bound condition. Therefore, release of the bound nutrients and energy in the waste materials of living organisms is crucial for the maintenance of the ecosystem.

This essential need of the ecosystem is being fulfilled by the collective efforts of the detritivorous faunal components, particularly the arthropod community inhabiting the soil ecosystem. Arachnids have been identified as the most important mediators of the process of decomposition in the soil (Tadros, 1976).

Oribatid mites represent one of the active links in the decomposer food web by playing multiple roles in the decomposition process. They are the only group among the arachnid members that are contributing to the soil structure (Norton, 1985).

The microscopic size and lower energy requirements of *A.* (*H.*) *chaliyamensis* and *H. hirsutus* make their contribution to the process of decomposition less recognizable than that of other arthropods and annelids but this seeming disadvantage is compensated by their abundance, diversity and adaptability (Tadros, 1976; Wallwork, 1976; Mitchell and Parkinson, 1976; Petersen, 1982a,b; Petersen and Luxton, 1982; Haq, 1994; 2007a). The unique adaptive strategies of oribatid mites have helped them in the invasion and colonization of many habitats, including those which are generally not suitable, as in the case of the Arctic region (Behan-Pelletier and Hill, 1978). Surprisingly, the mites in the present study, viz. *A.* (*H.*) *chaliyamensis* and *H. hirsutus* have acqired considerable environmental adaptability, even as marine or littoral zone inhabitants. Therefore, the contribution of this acarine group to the process of nutrient cycling in the soil ecosystem cannot be overlooked.

Three major feeding guilds have been identified among the oribatid mites, based on the type of food materials they prefer within the soil habitat (Schuster, 1956; Hartenstein 1962; Luxton, 1972; Haq, 1994, 1996). Of these, microphytophages represent the group with direct involvement in the litter decomposition process while the macrophytophages facilitate the process indirectly and panphytophages play a dual role, both direct and indirect (Haq, 1987). Materials of higher plant origin like dried leaves, twigs and decaying wood and roots constitute the principal food items of macrophytophages (Luxton, 1972; Behan-Pelletier and Hill, 1978; Haq, 1982; Haq and Ramani, 1991; Ramani and Haq, 1991; Haq, 1994, 2007a, 2016). The macrophytophagous oribatid mites are armored with strong, well-developed mouth parts for the trituration of woody materials. The rate of consumption is determined by several factors such as the nutrient content of the materials, nutritional requirements of the species concerned and also food processing and assimilatory efficiencies of the mite species (Haq, 2016). The nutritive value of the three principal categories of food materials in the decomposition food web decreases in the order of fungi > foliage material > woody elements (Slansky and Scriber, 1985). Nitrogen is an important nutritional requirement for all organisms. The organic matter of plant origin is poor in nitrogen in comparison with fungi. The nitrogen content of 36.2g of wood has been equated to that of 1g of fungal spores (Merrill and Cowling, 1966). The proportion of digestion among the detritivores is 6-35% against that of 42-97% in the case of the fungal feeders (Berrie, 1976; Cummins and Klug, 1979). This shows relatively poor processing ability of the former. In addition, the macrophytophagous oribatid mites have poor food assimilation efficiency that ranges between 10-15% against that of the 50-65% assimilatory efficiency of the microphytophages (Luxton, 1972). The dependence of these mites on food items of low nutrient content, coupled with their poor digestive and assimilatory powers, demands the consumption of enormous quantities of food. This is a positive factor from the ecological point of view as the contribution of these mites towards biodegradation of litter in the soil ecosystem will be always higher when compared to other feeding guilds of oribatids (Haq, 1994).

Oribatid mites exhibit wide ranging nutritional habits within the soil ecosystem (Schuster, 1956; Hartensten, 1962; Woodring, 1963; Shereef, 1970; Luxton, 1972; Haq and Prabhoo, 1976; Haq, 1982, 1994, 1996, Maraun et al., 1998). This imposes selection of preferred microhabitats and distribution pattern for individual categories of these mites so as to ensure the availability of surplus food for each species. In the current study, collection localities of

Figure 9. Oribatid mites and their feeding on decaying wood and leaf litter – A) *A*. (*H*.) *chaliyamensis* initiating feeding on decaying wood piece offered as food, B) Cut opened wood piece showing feeding galleries of *H. hirsutus*, C) Feeding galleries of *H. hirsutus* filled with fecal pellets, D) Mass of fecal pellets formed after feeding on leaf litter by *H. hirsutus*, E) Skeletonized leaf, F) Eggs of *H. hirsutus* in laboratory cultures, G) Immatures of *H. hirsutus* feeding on leaf tissue, H) *A.* (*H.*) *chaliyamensis* colonizing a piece of dried root offered for feeding in the laboratory culture.

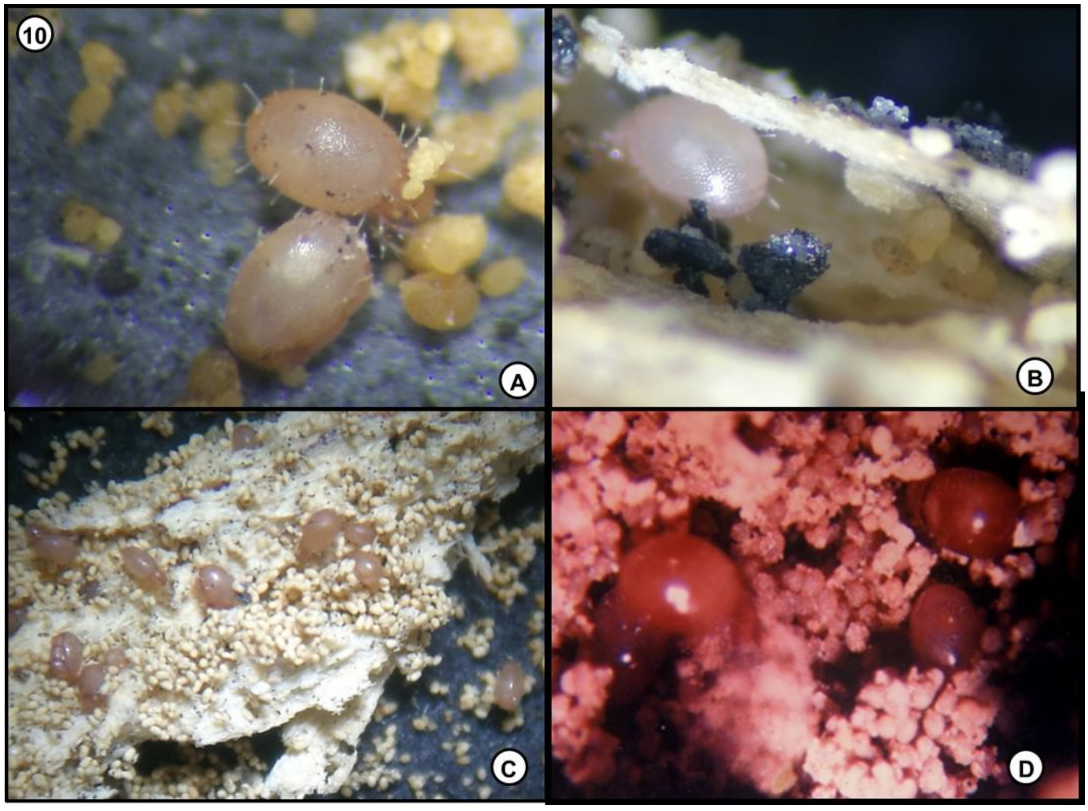

Figure 10. Degradation of wood pieces by *A*. (*H*.) *chaliyamensis* – A) Active adults, B) Formation of channel in the wood by vigorous feeding, C) Trituration of wood by the mite, D) Fecal mass produced by larvae and nymphs after feeding.

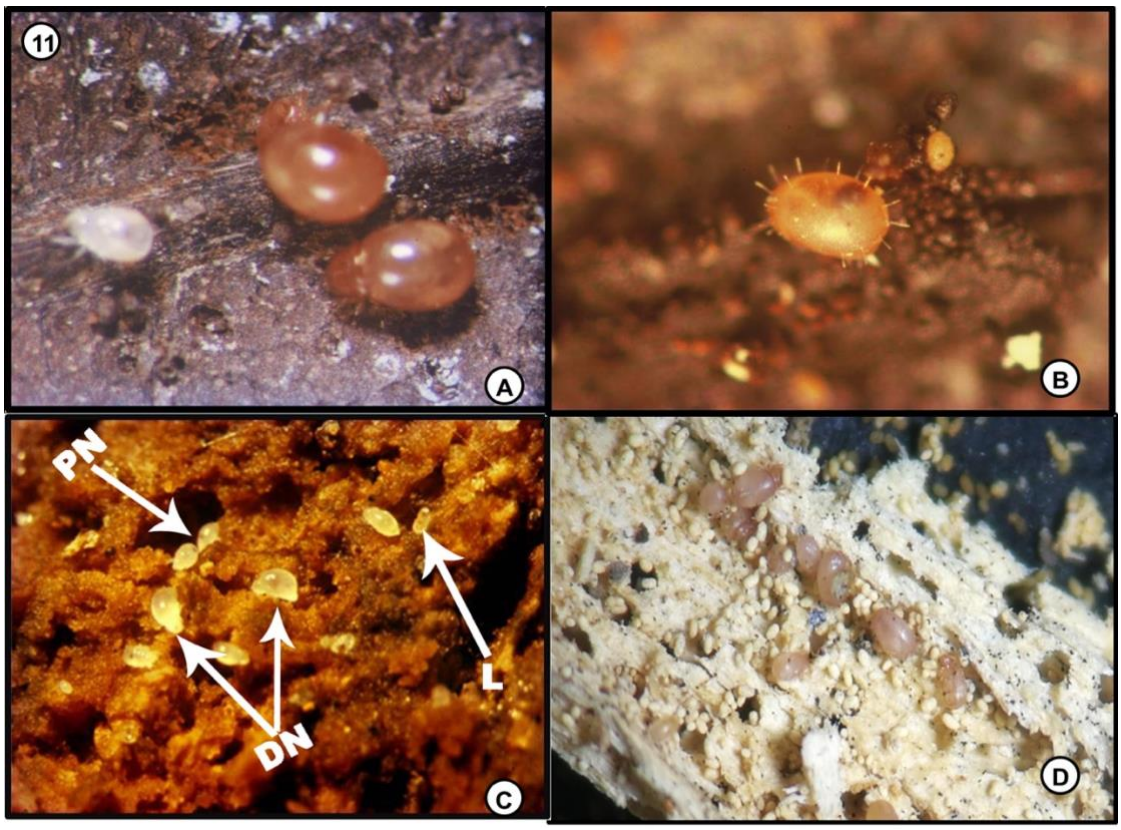

Figure 11. A) Continued feeding on bark tissue by *A*. (*H.*). *chaliyamensis*, B) Production of fecal pellets after feeding on bark tissue,, C-D) Larval and nymphal stages of *A.* (*H.*) *chaliyamensis* from the channels and pits created by the feeding activity (L: Larva, PN: Protonymph, DN: Deutonymph).

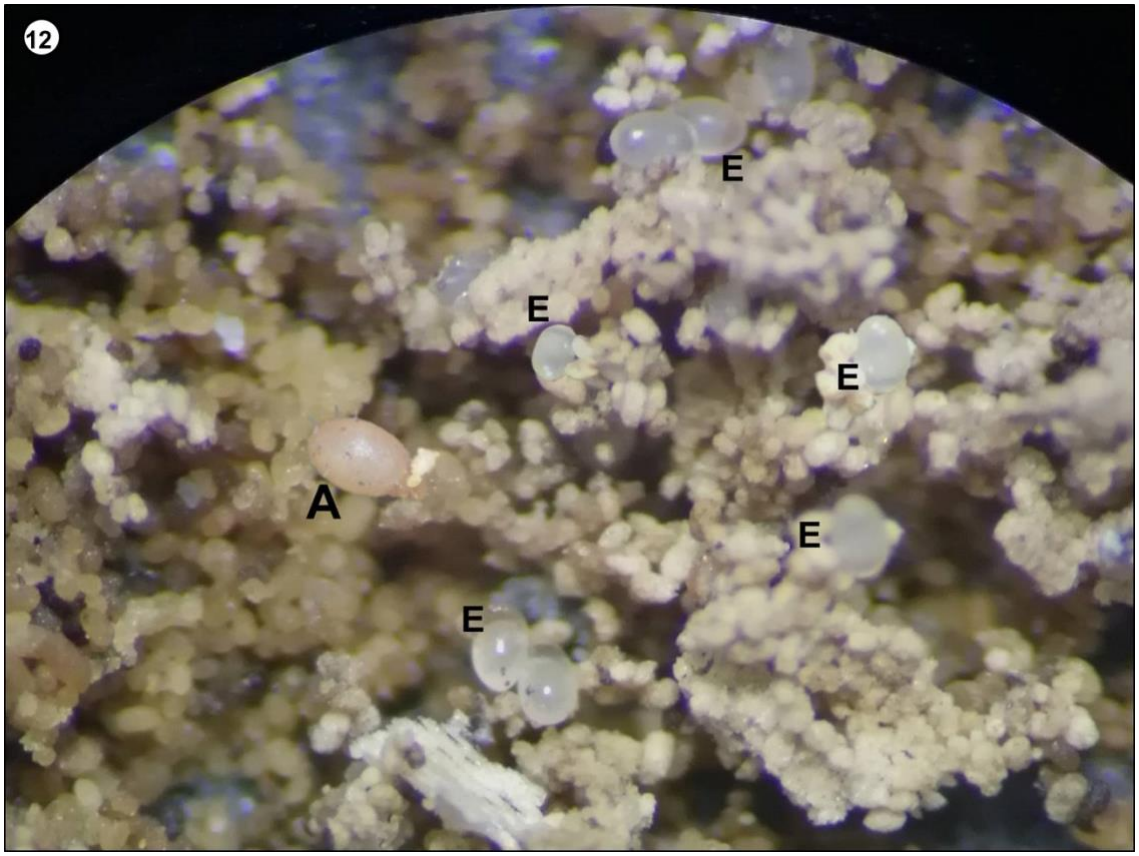

Figure 12. Eggs of *A.* (*H.*) *chaliyamensis* laid among fecal pellets in the laboratory (A: Active adult of *A.* (*H.*) *chaliyamensis*, E: Eggs).

Figure 13. Active adults of *H. hirsutus* (A-B) among fecal mass they produced in laboratory culture.

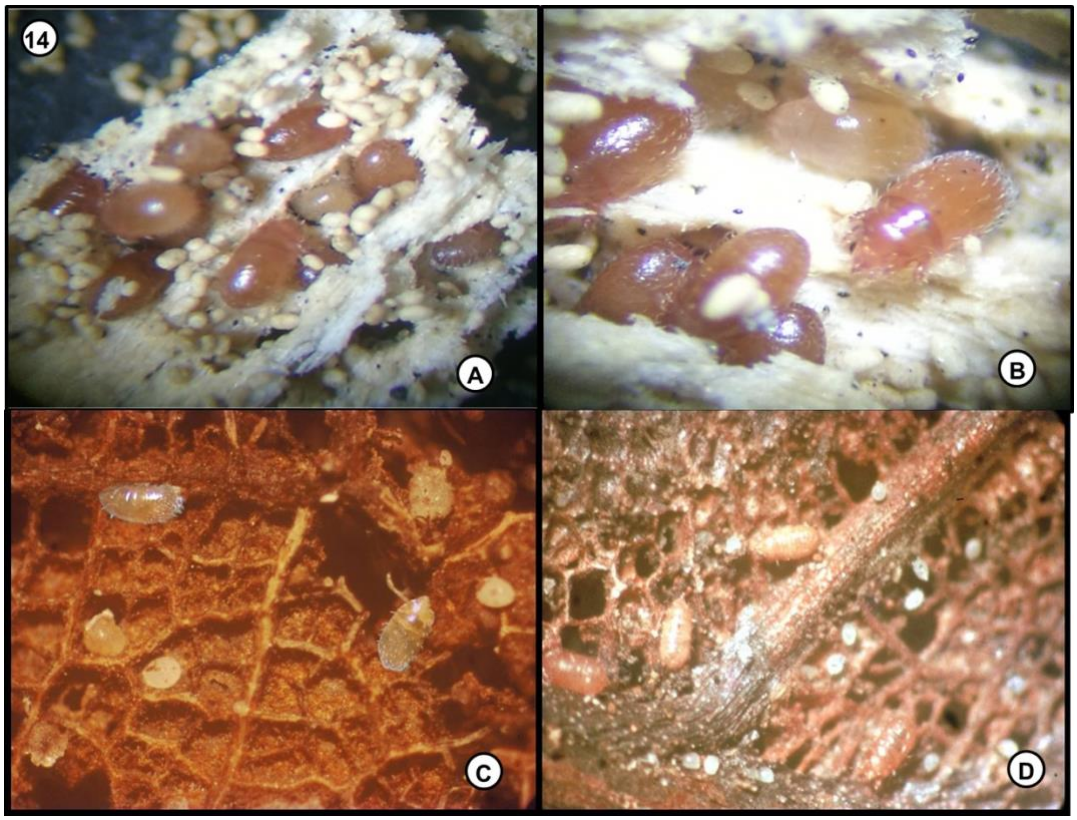

Figure 14. Wood and leaf litter feeding by *H. hirsutus* in the laboratory–A,B) Channels (A) and holes (B) produced in wood pieces by the feeding activities of *H. hirsutus* (Note the presence of larval and nymphal instars packed in pits and channels of the wood pieces given for feeding), C) Adults with eggs on softened leaf lamina offered as food, D) Eggs packed between veinlets in laboratory cultures.

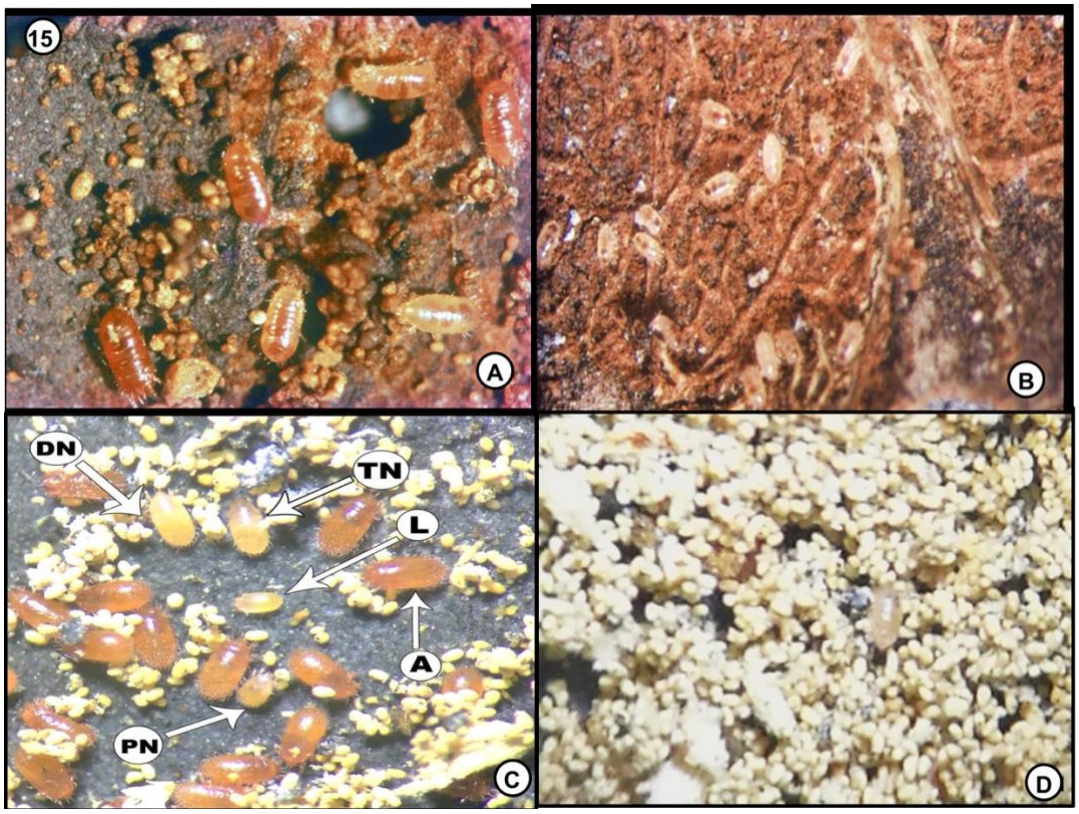

Figure 15. A) Other members of lomanniid mites cultured for similar studies, B) Active participation of various stages of *H. hirsutus* during feeding on leaf tissue and producing eggs, C) A: Adult, L: Larva, PN: Protonymph, DN: Deutonymph, TN: Tritonymph, D) Mass production of fecal pellets after feeding on wood by *H. hirsutus*.

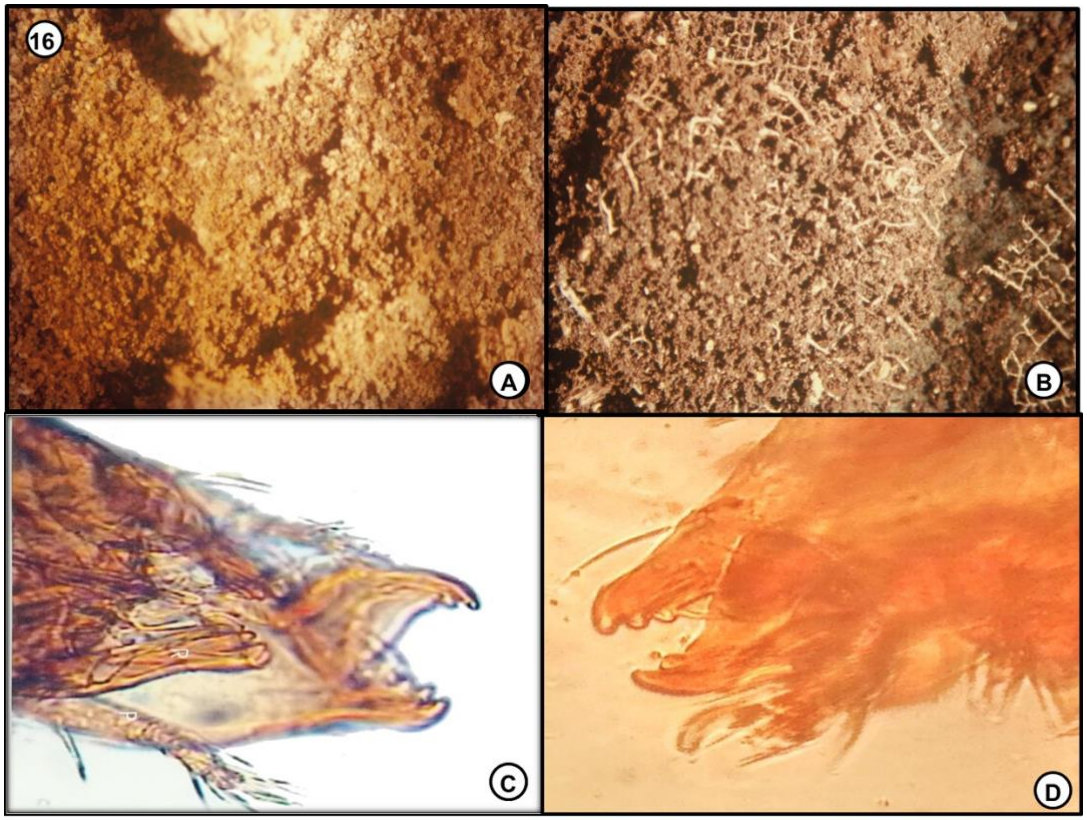

Figure 16. A) Mass production of feces by *A. (H.) chaliyamensis* after feeding on wood under laboratory conditions, B) Mass production of fecal material by *H.hirsutus* after simulatneous feeding on wood and leaf litter, C) Chelicera of *A.* (*H.*) *chaliyamensis*, D) Chelicera of *H. hirsutus*.

the mites comprised of secondary forest ecosystem, grassland, agriculture and monoculture land and beach soil, presenting varied abiotic and biotic factors. Three species of mites, namely *A. longisetosus, O. kuhnelti* and *S. minuta* exhibited their presence across all the four localities having distinct environmental conditions, thereby presenting the eurytypic distribution pattern referred by Wallwork (1976) for species of *Oppia* and *Scheloribates*. Further investigation on feeding habits of these mites have indicated the prevalence of panphytophagy among them. This feeding habit may be owed to their eurytypic distribution as hypothesized, based on various research (Luxton, 1972; Harding and Stuttard, 1974; Behan-Pelletier and Hill, 1978).

The classic nutritional habits assigned for the soil oribatids, namely macrophytophages, panphytophages and microphytophages (Schuster, 1956; Hartenstein, 1962; Luxton, 1972) could be observed among the mite species investigated in this study as well. Macrophytophagy was the dominant food habit among the mites screened during the current study. This trend was recorded among 68% of the species, followed by panphytophagy (19%) and microphytophagy (13%). Although the basic organisation and pattern of the gnathal structures are the same in *A.* (*H.*) *chaliyamensis* and *H. hirsutus*, certain variations of these structures were obvious and can be related to their food and feeding habits. The presence of the vestibule on the rutella observed in *A.* (*H.*) *chaliyamensis* can be related to the xylophagous feeding of the species, as suggested by Woolley (1967). The prominent and strong teeth on the chelicerae of this species resembled the teeth reported among other species of this genus (Dinsdale 1974). The stenarthric infracapitulum with its highly reduced mentum allows maximum rutellar mobility laterally. Dinsdale (1974) noted that the rutellae normally envelop the retracted chelicerae but during feeding the chelicerae are protruded and the rutellae diverge as a result of the associated deformation of the infracapitulum. Furthermore, Grandjean (1957) suggested that during the retraction of the chelicerae the rutellae converge to scrape their outer surfaces and the resulting particles are collected by the mouth. The structure and organization of the mouthparts of *A.* (*H.*) *chaliyamensis* has provided evidence for its ability to triturate comparatively large and stout wood pieces very effectively, involving the application of the maximum power of the rutellae. The involvement of hemolymph pressure and anal and adanal musculature in the defecation process of oribatid mites has been reported (Heethoff and Norton, 2009) and matches with the anatomy of the macrophytophages.

In the current study, both the species of mite showed a preference for the drift wood collected and showed similar feeding activity, with tunnelling into the wood and laying eggs within the tunnels. The wood feeding tendency of *H. hirsutus* is exceptional among the lohmanniid members but was reported by the author earlier for this species (Haq, 2007a). Gut enzyme analysis was used as a complementary assay to confirm the feeding habits of oribatid mites in the laboratory experiments. In a similar study on multiple groups of oribatid mites, Haq and Konikkara (1988) and Haq (2007a) discussed the enzyme profiles of the macrophytophages, panphytophages and microphytophages, and reported that the macrophytophages possess the three carbohydrases, cellulase, cellobiase and maltase. This has been demonstrated again in the current study, confirming the macrophytophage identity of *A.* (*H.*) *chaliyamensis* and *H. hirsutus*.

The occurrence of symbiotic microorganisms in the gut of oribatid mites and their role in the digestion by the latter was reported by Haq and Konikkara (1988) who hypothesized that the microbes were the sources of the enzymes detected in the gut of these mites. *In vitro* cellulose digestion by gut microbes from oribatid mites was reported by the author later (Haq, 2007a). During the current study, diverse microbial colonies were isolated from the gut contents and fecal pellets of both the species of mites, indicating their role in the digestion of the cellulose rich food consumed by these mites. The presence of carbohydrases among a wide range of oribatid mite species and determination of their feeding guilds based on their enzyme profile was reported with more narrow levels of categorization splitting the classic macrophytophage, panphtophage and microphytophage categories into 9 feeding guilds (Siepel and de Ruiter-Dijkman, 1993).

The contribution of oribatid mites to the biodegradation of soil organic matter at the functional level is undisputed and well-illustrated in many terrestrial ecosystems like tropical forest soil, temperate forest soil, grasslands, peat soil and even degraded ecosystems like mines. However, quantitative assessment of the extent of litter biomass turnover by these mites is meagre, though the concept was demonstrated with artificial food (i.e. filter paper) offered to *H. hirsutus* (Haq, 1987). Considering this information gap, the current study estimated the loss of biomass due to the feeding activity of two mite species under laboratory conditions. There was an average reduction of 8.8% and 7.5% in the mass of the wood tissue offered as food to *A.* (*H.*) *chaliyamensis* and *H. hirsutus*, respectively, within a period of 2 to 3 days. This type of study on more species is essential in assessing the contribution of these mites to the biodegradation process.

Chemical analysis conducted in the present study showed a general increase in the concentration of the three macronutrients tested. This has established the potential of oribatid mites in the enhancement of soil fertility through the enzymatic breakdown of litter components in their gut and subsequent release of nutrients to the environment. Information regarding this crucial role played by oribatid mites was provided by a number of earlier workers (Schuster, 1956; Wallwork, 1958; Hartenstein, 1962; Hayes, 1963; Berthet, 1964; Luxton, 1966; Kowal, 1969; Kowal and Crossley, 1971; Hammer, 1972) who stressed the bioprocessing ability of oribatid mites. The relevance of these mites in the recycling of a few essential nutrients like calcium and potassium has been brought to light (Cornaby et al., 1975; Gist and Crossley, 1975; Werner and Dindal, 1987). Norton (1985)

reported that feeding by oribatid mites leads to an increase in the nitrogen content of organic litter. Haq (1996) conducted quantitative analysis of certain macro- and micronutrients in selected items of plant litter after its consumption by oribatid mites and reported a general increase in concentration of nitrogen and phosphorus in all the materials tested. Ramani and Haq (2001) showed that the feeding activity of *H. rimosus* and *Lohmannia* sp. increased the nutrient status of the litter of *Artocarpus integrifolia.* The same authors found that both macro- and micronutrient levels increased. These observations, in conjunction with the present findings, signify the involvement of oribatid mites in soil productivity.

The global distribution and diverse habitats occupied by oribatid mites when coupled with their varied feeding habits (Hayes, 1963; Luxton, 1966, 1972; Haq, 1976, 1982; Haq and Prabhoo, 1976; Behan-Pelletier and Hill, 1978, 1983; Ramani and Haq, 1991) affirm the significant role of these mites in the process of nutrient and energy cycling. The importance of rhizophagous (i.e. feeding on dead roots of plants) oribatid mites in soil aeration, drainage and clearing of dead mass of roots in the soil profile has been reported (Rogers, 1939; Ghilarov, 1971) and hence the significance of *A.* (*H.*) *chaliyamensis* and *H.hirsutus* needs special appraisal.

The ability of oribatid mites to degrade plant materials containing complex organic molecules, which they ingest as food, and the mode of achievement was an integral part of this study. Therefore, tracing the mechanisms involved in the hydrolysis of complex plant polysaccharides like cellulose, lignin and chitin was considered essential for ascertaining their impact in the process of decomposition (Hartenstein, 1962). This was evidenced by the production of large quantities of excrements rich in nutrients by *A.* (*H.*) *chaliyamensis* and *H. hirsutus* (Figs 16 A, B). The decrease observed in the cellulose content of *Quercus* leaf litter after digestion by oribatids like *Hermanniella* sp. and *Peloribates* sp. (Gasdorf and Goodnight, 1963) indicated the cellulolytic activities of this group of mites. Synthesis of the enzyme cellulase is rare among animals. Generally, this unusual capacity is possessed by microbiota. Very few organisms, including silverfish (Lasker and Giese, 1956) and a marine crustacean (Boyle and Mitchell, 1978), possess the ability to digest cellulose without the help of microbes (Waller and La Fage, 1987). Confirmation of this theoretical view was achieved through the enzyme assay of 14 species of oribatid mites (Luxton, 1972; Haq and Konikkara 1988, Haq, 2007a) which established the presence of several carbohydrase groups of enzymes in their intestines. Further evidence of the presence of enzymes like cellulase, carboxymethyl cellulase, xylanase and pectinase that are capable of breaking down plant structural polysaccharides has also been reported (Haq and Konikkara, 1988). Meanwhile, a contradictory result establishing the absence of cellulase activity and cellulolytic gut symbionts in the case of Phthiracarid members has also been provided (Dinsdale, 1974). The presence of cellulase, glucosidase, galactosidase, trehalase, raffinase and lactase in the gut of the xylophagous mite, *H. hirsutus,* was correlated with preferential wood feeding and also the ability to digest ingested fungal particles (Haq, 1984).

Obviously, the above information has clearly revealed the fact that large quantities of litter of higher plant origin supported a rich population of phthiracarid and lohmanniid mites. Therefore, further extention of field cum laboratory studies have been followed on two representative members of the above families. These two groups of mites have been identified as macrophytophages (Haq and Prabhoo, 1976; Behan-Pelletier and Hill, 1978; Haq, 1982, 1984, 1987, 1994, 1996; Haq and Konikkara, 1988). A majority of lohmanniid mites are phyllophagous, which restricts them to areas where leafy litter is available. However, the lohmaniid species of the current study, *H. hirsutus*, is an exception that prefers the woody tissues of higher plants, particularly driftwood and roots of *C. gigantea* in the beach ecosystem (Haq, 1987, 1994, 1996; Seniczak et al., 2018). Therefore, this species can be assigned to the subcategory of xylophages of the major feeding guild, macrophytophages. The members of the family Phthiracaridae are typically known for their wood feeding habit categorized as xylophagous (Haq, 2007a). The results of the gut enzyme profile and microbiome assay of these mites in the current study have provided evidence of their competency in the decomposition of the cellulose rich woody tissues of the higher plants. The actual role of these mites in mineralization and nutrient cycling has been established through the increased major macro- and micronutrient content of the partially digested woody tissue defecated by the mites, as reported for other oribatid mites (Haq, 2007a, 2016).

The current study recorded an increase in macronutrients in the woody litter from the collection sites after its digestion by the two species of mites studied. The trend in percentage increase of the macronutrients in the descending order in the experimental trials was N>P>K. However, the concentration of Ca and Mg decrease after digestion by the mites. The reduction in the Ca content may be attributed to the requirement of the nutrient by these mites. A similar study on the effect of the mite *Scheloribates moestus* Banks feeding on corn and oak litter reported an increase in the quantities of polysachharides and phenols in the fecal matter produced after feeding on corn litter but no change in polysaccharide and phenol contents in faecal pellets of those fed on oak litter, indicating the influence of the type of litter on the process of decomposition and turnover of nutrients associated with digestion (Wickings and Grandy 2011). Hence it appears that more investigations on nutrient conversion in the litter of same plant species by different species of mites and vice versa are required in order to elicit specific details of the nutrient release achieved through digestion by oribatid species during litter decomposition. The experimental evidence of nutrient enhancement of soil by oribatid mites and the potential scope of their use for enhancement of soil fertility using the okra plant as a model system (Haq, 2016) confirmed the nutrient enrichment of plant litter through decomposition by oribatid mites at a gross level.

Further studies carried out on the driftwood pieces, litter and roots of *C. gigantea* collected from Chaliyam and Beypore beaches supported the full growth and reproduction of *A.* (*H.*) *chaliyamensis* and *H. hirsutus* in laboratory cultures. This provides confirmatory evidence for their xylophagous habit, as breeding activity and persistence are the ultimate standards in assessing the food preference of oribatid mites (Haq 1984, 1994, 2007a). The total period of ontogeny ranged from 81-89 days for *A.* (*H.*) *chaliyamensis* and 141-154 days for *H. hirsutus* in the current study. The duration of ontogenic development of oribatid mites is highly dependent on the type of nutrition (Brückner et al., 2018). The development of certain species of mites can take more than one year (Schatz, 1985). The duration of ontogeny of both species studied in the present investigation is comparatively higher with reference to the species of oribatids in the tropics. The longer developmental period of the members of Phthiracaridae and Lohmaniidae appears to be advantageous to the ecosystem when viewed from the angle of litter degradation, as the feeding and efficiency of the nymphal stages of *H. hirsutus* have been rated as par with that of the adult (Haq, 1987). In considering the feeding habit, enzyme profile, microbiome of the gut and nutrient turnover efficiency of *A*. (*H.*) *chaliyamensis* and *H. hirsutus,* the current study has established their competency beyond doubt in the bioprocessing of plant litter and enhancing soil fertility. The longer developmental period of the mites complements their niche in the soil ecosystem. Therefore, these two species have fulfilled the purpose of a model system by demonstrating biodegradation, nutrient release and enhancement of soil fertility.

Extension of the laboratory feeding and breeding studies of the mites was made through a field trial on soil fertility enhancement using *V. unguiculata* as the model plant and *A.* (*H.*) *chaliyamensis* to validate the concept of "oriculture farming practice" (Haq, 2007a,b, 2016). Establishment of a population of mites involved in biodegradation is essential for oriculture farming. Biotic potential is an important determining factor for population growth of any species. The availability of preferred food in surplus is one of the key factors governing biotic potential in the environment. Both the oribatid species used in this study produced a large numbers of eggs and a large population in the laboratory cultures. This facilitated the tracing of their ontogeny in the laboratory, proving their ability to produce large populations when their preferred food is available in surplus.

The final phase of bioprocessing of organic residues in the soil is the release of previously bound nutrients in them in a form freely available for absorption by plants. The macro-, meso- and microfauna of the soil bring about this transformation through their synergistic activities, mostly linked to their nutrition and growth. The contribution of each category of soil fauna towards nutrient cycling is determined by the quantity, quality and composition of the nutrients released during their bioprocessing activities. This aspect is well demonstrated primarily by the preference for woody elements in the case of *A.* (*H.*) *chaliyamensis* and *H. hirsutus* and secondarily by leafy elements of the litter by *H. hirsutus.* Biodegradation ensures the availability of the essential nutrients in soil for plant growth. Timely absorption of optimal amounts of nutrients optimizes plant growth and productivity. Vegetative growth, flowering and seed production are considered the milestones in plant life and are used as performance indicators for the assessment of agricultural productivity. The present study used plant height and pod length as the measurable indices for assessing the productivity of the vegetable crop *V. unguiculata* for the evaluation of the impact of oriculture in soil fertility enhancement.

The current study has demonstrated that oriculture has a positive impact on the vegetative growth of the selected plant *V. unguiculata* through a clear enhancement in the rate of vegetative growth of the plant at different age levels, as envisaged by length of the twiners plant height and pod length as stated above. Oriculture influenced the productivity of the plants as well, with enhanced growth of the pods produced by the treated plants, as per the results of the present study. This is comparable to the outcomes of earlier small scale trials on the same plant where reduction in flowering time and increase in average biomass and number of pods produced by *V. unguiculata* was demonstrated. However, the lohmanniid mite *Meristacarus degradatus* was used as the test species for demonstrating oriculture (Haq, 2007b). The same author (2016) further confirmed the value of oriculture with okra plants by using five species of oribatid mites. Therefore, the potential of oribatid mites to contribute to soil fertility enhancement and organic farming through oriculture (Xavier and Haq, 2005; Haq, 2007a, b, 2016), the long awaited result aimed during the study has been evidenced. The diverse ecological niches occupied by oribatid mites and their ability to tolerate lower soil humidity levels when compared to earthworms make oriculture practice complementary to vermiculture in supporting sustainable agriculture.

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