International Journal of Fauna and Biological Studies
Available online at www.faunajournal.com

IJFBS International Journal of Fauna And Biological Studies

ISSN 2347-2677
IJFBS 2016; 3(2): 14-17
Received: 13-01-2016
Accepted: 05-02-2016

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Extraction of Soil Microarthropods: A low cost Berlese-Tullgren funnels extractor

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Abstract
Construction and operation of a low cost Berlese-Tullgren funnels extractor for extraction of soil microarthropods is described. The extractor is efficiently and rapidly extracts flightless microarthropods from soil samples. It is constructed from materials readily available in the local market in India in which soil arthropods are forced by a temperature gradient to move from soil samples to the collecting tubes. It is economical and easy to procure as compared to the imported counterpart.

Keywords: Berlese-Tullgren funnels, Diversity, Extraction, Microarthropods, Soil.

1. Introduction
Soil microarthropods constitute one of the most species rich communities in forest ecosystems [2]. They have a great impact in functioning of the decomposer as a result of their feeding activities; nutrient regeneration and soil structure have been well documented [6]. In practice, soil microarthropod sampling involves collection of soil which is taken back to the laboratory for extraction. There are several methods to estimate diversity and dynamics of soil micro-arthropods of forest and grassland ecosystems. For qualitative and quantitative studies of living soil microarthropods; one of the most popular method is Berlese-Tullgren extraction method. The Berlese-Tullgren extraction method is the simplest one, efficiently and rapidly extracts flightless microarthropods from soil samples in which soil animals are forced by a temperature gradient move down from the soil sample to the collection vessels [7, 12, 13]. In this method, samples were placed on a coarse sieves fixed across the wide end of a funnel and heated by 25-watt light bulbs fitted into a metal reflectors placed about 25 cm above to the funnel. The heat of bulb dried and warmed up the sample, causing animals to move downwards and fall through the sieve into the funnel, which directs them into a collecting tube below. The collecting tube can contain water or alcohol to prevent the animals from escaping.
The aim of this paper is to present a low cost Berlese-Tullgren funnel extractor with affordable budget capable to handling of bulk samples. The design was developed and proved highly effective for qualitative and quantitative studies of soil micro arthropods. It is constructed from materials readily available in the Indian local market. It is economical and easy to procure as compared to the imported counterpart.

2. Literature review
Soil is a vigorous living matrix that is a crucial part of the terrestrial ecosystem. Soil contains great numbers of various living organisms assembled in complex and varied communities [5]. A high proportion of soil fauna are the arthropods. The most abundant are collembolans, mites, while other arthropods include spiders, millipedes, centipedes and insects [10, 11]. Tullgren extraction method is an active procedure that relies on the migration of arthropods from the sample. Crawling microarthropods move down through the soil sample in response to changes in heat and humidity gradients [8]. Berlese-Tullgren funnels have been used for extracting arthropods from soil and litter samples for many years but are generally considered too costly for students and researchers. Many researchers have stressed the need for slow application of heat to the samples for maximum efficiency [1, 4, 9]. Dombos [3] constructed a modified extractor for sampling of springtails populations from small volume soil cores in high sample size. Van Straalen & Rijninks [14] studied the rate of accuracy of the extraction with respect to the external factors, such as soil type, species and age of soil arthropods. The effects of refrigeration time on extraction efficiency of soil acarina have also documented [2, 6].

3. Results and Discussion

Funnels Design

The construction of the extractor is based on a steel shelf (Fig.4). Overall dimension of the extractor for three units are 6.3x3x1.5 ft. Heat is provided by 25Watt electric ordinary lamp bulbs with metal reflectors and control by switches. 6-holed three plywood panels with 12 mm thickness are fitted at equal distance to hold plastic funnels loaded with samples in small sieves and covered with big stainless steel sieves. Three good quality moisture and pest proof wooden frames are fitted for electrification to hold the bulbs for heating. The collecting medium is usually tap water or alcohol to prevent the animals from escaping. A details of the material used in the construction of low cost extractor, available in Indian market is described (Fig. 4, 5 & 6).

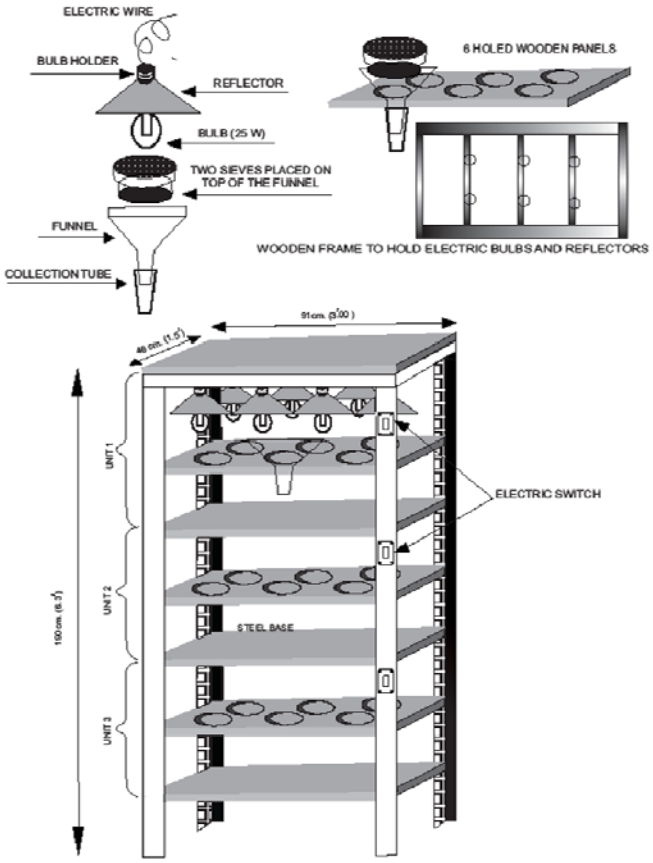

Fig 4: Schematic illustration of low cost Berlese-Tullgren funnels extractor

1. ***Steel Shelf*:** A steel shelf with dimensions 6.3x3x1.5 ft.
2. ***Wooden Panels*:** Three good quality 6-holed plywood panels with 12 mm thickness to hold the funnels in each hole.
3. ***Wooden Frames*:** Three good quality wooden frames for electrification of bulbs and metal reflectors.
4. ***Metal Reflectors*:** Eighteen good quality metal reflectors to avoid the wastage of heat.
5. ***Ordinary Lamp Bulbs*:** Eighteen ordinary lamp bulbs (25-Watt) for constant heating source to warm up the samples.
6. ***Electric Switches*:** Three good quality unbreakable electric switches to control the extractor.
7. ***Steel Sieves*:** Eighteen pairs of good quality stainless steel sieves of two sizes (15.5 & 18.5 cm diameter) with 22 mesh sizes to fix across the wide end of each funnel. (Fig.6).
8. ***Plastic Funnels*:** Eighteen good quality virgin plastic funnels (20.5 cm diameter).

Fig. 5: Fabricated a low cost Berlese-Tullgren Funnels extractor in laboratory

Fig 6: Enlarged view of Funnels

Initial soil temperature was recorded 26 ºC. During the extraction of soil microarthropods maximum temperature was recorded 41.5 ºC (Fig.1).

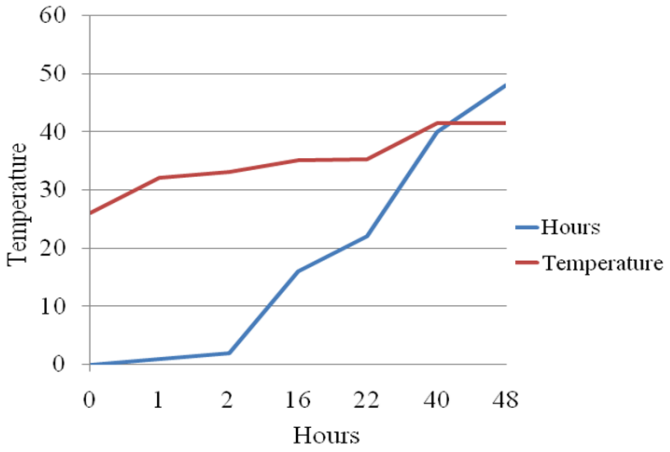

Fig 1: Temperature Profile

Speed & Efficiency of Extraction

Speed was tested by loading the funnels with fresh grassland soil samples on sieves and covered by big sieves to avoid the jumping out the animals from the samples. Samples were heated from above by an ordinary lamp bulbs (25 W), suspended about 20 cm above each sample to continuous heating for maximum extraction of different microarthropod groups. As the surface of the sample becomes heated and desiccated, the animals move down and are driven out from the sample, and collected into the specimen tubes filled with tap water placed below. Maximum extraction of soil micro-arthropods was recorded on second day after 16 to 22 hours of continuous heating at temperature ranges between 35.1ºC to 35.2ºC. Minimum extraction was recorded at 41.5ºC (Fig.2). The maximum population was recorded of Order *Collembola* followed by *Cryptostigmata* mites, *Mesostigmata* mites, *Prostigmata* and *Astigmata* mites. The minimum numbers of individuals were recorded of *Protura* and *Symphyla* groups. After 48 hours of continuous heating no extraction of soil arthropods were found (Table 1). The upper big sieves were served as a lid to avoid escaping the live specimens and also served as ventilator to prevent immediate desiccation of the sample.

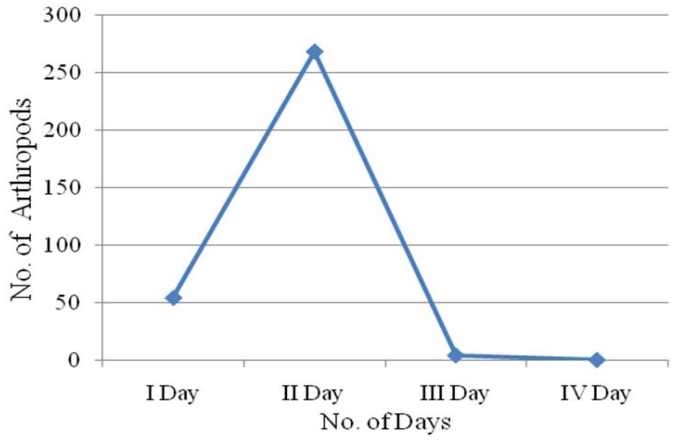

Fig 2: Rate of Extraction

Table 1: Speed of extractor among soil microarthropods groups

Groups	Sieve mesh size (mm)	Extraction time (48 hours)	Extraction time (after 48 hours)
Collembola	22	+	-
Cryptostigmata	22	+	-
Mesostigmata	22	+	-
Prostigmata	22	+	-
Astigmata	22	+	-
Pauropoda	22	+	-
Protura	22	+	-
Diplura	22	+	-
Pseudoscorpion	22	+	-
Symphyla	22	+	-

(+) = Present; (-) = Absent

The efficiency was tested by loading the funnels with grassland soil samples containing known numbers of micro-arthropods of different groups were placed into the extractor and warmed up with ordinary lamp (25 Watt) bulbs continuous for the 48 hours for extraction and calculated percent recovery for each group (Table 2).

Table 2: Percentage recovery of microarthropods groups from grassland sample

Groups	No. of Individuals (n)	%
Collembola	30	100
Cryptostigmata	10	90
Mesostigmata	7	86
Prostigmata	6	100
Astigmata	1	100
Diplura	1	100
Protura	2	50
Pseudoscorpion	1	100
Symphyla	2	50

4. Conclusion

A low cost Berlese-Tullgren funnels method for extraction of soil microarthropods is described. The extractor was built up by both the authors and designed to suit with red gravelly/sandy and black soils for extraction of soil micro-arthropods. Construction of a low cost Berlese-Tullgren funnels extractor is strongly recommended to any researcher embarking on qualitative and quantitative studies of soil microarthropods.

5. Acknowledgements

We thank to the Director, Zoological Survey of India, Kolkata for necessary facilities. Authors are also grateful to the Director, Central Arid Zone Research Institute, Jodhpur for his keen interest, constant encouragement and support for this work.

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