

Nematode Community Structure in Desert Soils: Nematode Recovery¹

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Abstract: The sugar-flotation-sieving (SFS) and Baermann-funnel (BF) methods were compared for nematode extraction efficiency. The SFS method recovered nematodes from more trophic groups whereas greater total numbers of individuals were recovered by BF. In a test to validate the efficiency of SFS, virtually 100% of the nematodes added to desert soil prior to extraction were recovered by four consecutive SFS washings of each soil sample. Estimations of nematode biomass in desert soils based on numbers of nematodes extracted by the two methods were similar unless there was large reserve of eggs in the soil. The biomass of nematodes from a Colorado desert soil was 0.9 g/m² as determined by both methods, whereas BF gave 0.17 g/m² for nematodes from a Mojave desert soil as compared to 0.9 g/m² with SFS. *Key Words:* soil sampling.

Although deserts comprise 15% of the land area of the contiguous United States, there is little information concerning the distribution and ecology of nematodes occurring in such habitats. As a part of the United States International Biological Program, Desert Biome study, the community structure of nematodes in undisturbed desert soils was investigated. The apparent cryptobiotic condition of the nematode fauna during long dry periods required special consideration in nematode population studies. Initial nematode numbers extracted from deserts were low, compared to nematode numbers usually found in more humid soils. In efforts to determine if these differences resulted from poor recovery, two nematode extraction methods, Baermann-funnel (BF) and sugar-flotation-sieving (SFS) (4) were compared using desert soils. Recent papers have compared extraction procedures with different soils (2, 5, 7, 8, 9). The two methods, BF and SFS, are known to require less effort and time than the laborious Cobb's sieving

technique or the centrifugal flotation technique. A disadvantage of the BF technique, however, is that the total numbers of nematodes recovered from soil include nematodes revived from cryptobiosis and larvae hatched from eggs. The SFS technique recovered both dead and cryptobiotic nematodes and lost viable eggs. The purposes of comparing these two techniques were to identify a fast, efficient extraction method which represented the total nematode population in desert soil at the time of sampling and to determine any effects on nematode biomass computation due to extraction methods.

METHODS

Two Mojave desert sites, Victor Valley, California, and Rock Valley, Nevada, and a Colorado Desert site at Deep Canyon, California, were chosen for study. Soil samples of 500 cm³ were collected with a shovel to a depth of 20 cm. Stones larger than 1.3-cm diam were discarded, and the soil samples were placed in plastic bags and held in an insulated chest during transit. Samples were refrigerated (4C) until processed, usually within 72 h. Initial experiments showed nematode recovery was not affected by chilling. Each desert sample was mixed twice

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on a split-sample mixer and a 50-cm³ subsample from each sample analyzed.

In the BF technique, 50-cm³ soil were placed on a BF composed of a 100-cm diam glass funnel (3) and a layer of 1-ply Shopmaster® Heavy Duty Wipes (Crown Zellerbach Corp., San Francisco, California) supported by a wire screen. The funnel was filled with tap water to the bottom of the screen, and nematodes were removed after 48 h.

The SFS extraction procedure of Byrd et al. (4) also was used. In our tests, nematodes collected from the 44- μ m (325-mesh) sieve were poured onto a 38- μ m (400-mesh) sieve. Stirring time before sieving was 30 sec. Preliminary extraction tests, including varying the concentrations of sugar, Separan NP10® (Dow Chemical Company, Midland, Michigan) or the stirring time, did not increase nematode recovery. Each soil subsample was processed four consecutive times by SFS except in the method comparison test and the biomass comparison test where each subsample was processed twice. After each extraction, nematodes were counted, identified, and placed in one of the following trophic groups: (i) fungivorous - *Aphelenchoides* spp., *Aphelenchus avenae* Bastian, 1865, *Ditylenchus* spp.; (ii) microbivorous - mainly Cephalobidae; (iii) omnivorous/predaceous - Dorylaimina; (iv) plant parasites - Tylenchida; and unidentifiable - those deteriorated or unrecognizable. Results were expressed as numbers of nematodes per 500-cm³ soil.

Soil samples from Victor Valley were pooled, mixed, and a total of 30 subsamples were used for analysis, 10 each for the following procedures: (i) BF, (ii) SFS - two extractions, with an additional recovery on a 30- μ m (500-mesh) sieve to determine the numbers of nematodes that wash through the 420- μ m (40-mesh) and 44- μ m (325-mesh) sieve during the first sieving of the SFS processing, and (iii) SFS - four extractions, 48 h after 30-ml water were added to the soil samples. Data were analyzed by Duncan's multiple range test, and least significant differences (LSD) were computed.

The efficiency of extracting nematodes by the SFS method was determined by adding *A. avenae* to 10 Rock Valley, Nevada, soil subsamples. *A. avenae* was used because it was recovered in low numbers from Rock Valley soils and was easily cultured. Thirteen-

hundred (± 125) *A. avenae* from *Rhizoctonia solani* cultures (6) were pipetted in 10-ml of water onto each of the ten 100-cm³ soil subsamples. The nematodes were then extracted from soil by SFS. Controls consisted of the same soil with 10-ml water added. *A. avenae* were counted, and the percentage of nematode recovery calculated.

The efficiency of the SFS method was tested on two natural desert soils, the Colorado desert site (74.4% sand, 14.0% silt, 11.8% clay) and the Rock Valley site (61.1% sand, 32.4% silt, 6.5% clay). The soil was mixed and ten 100-cm³ subsamples from each soil site was processed by SFS. The results were compared with SFS extractions of two irrigated field soils obtained from the University of California, Riverside (Field A: 45.7% sand, 26.7% silt, and 27.6% clay, a sandy clay loam, and Field B: 66.8% sand, 19.9% silt, and 13.3% clay, a sandy loam) to identify possible differences between desert soils and more humid soils.

To determine if the number of nematodes in the various trophic groups were affected by the method of extraction, a test was run using the two methods. Ten 50-cm³ aliquants of a homogeneous Colorado desert soil were processed by BF and SFS. Data were analyzed by Student's *t*-test.

Nematode biomass is derived from numbers of nematodes extracted and the average length and width of extracted nematodes (1). The effects of extraction methods on biomass calculations were compared in two desert sites as well as in an irrigated field. Twenty soil samples were taken at 10-m intervals, 20-cm deep, along north-south and east-west transect within a 1-hectare, square plot. Each sample was mixed, and two 100-cm³ subsamples were used for the extractions, one subsample each for BF and SFS. Nematode numbers for the biomass study were based on average counts of the 20 transect subsamples. Nematodes were preserved in 2% formaldehyde, and the average length and width of 125-175 representative nematodes from the pooled 20 subsamples were used for biomass calculations. Nematode biomass was calculated as g/m² to a depth of 20 cm.

RESULTS

Numbers of nematodes per 500-cm³ soil extracted from Victor Valley desert soil by the

three procedures were: BF, 2,668; SFS, 1,685; and, water + SFS, 1,524. There were no significant differences between numbers of nematodes recovered by the SFS-4 extractions + H₂O and SFS-2 extractions with the 30- μ m (500-mesh) sieve. The 30- μ m (500-mesh) sieve trapped nematodes that usually are lost after the first SFS sieving, but they represented only 7.8% of the total nematodes recovered.

Nearly all *A. avenae* were recovered from both the "seeded" and natural control soils. An average of 1,380 *A. avenae*/100-cm³ soil of the 1,300 \pm 125 *A. avenae* added to each desert-soil subsample were recovered after four SFS extractions. Only 28 *A. avenae* were recovered from the control soils.

Approximately 75% of the total nematodes recovered from the Colorado and Mojave desert soils were obtained with two SFS extractions. Nematode numbers recovered from the Colorado and Mojave desert soil were 1,680 and 1,314 per 500-cm³ soil, respectively. The nematode numbers recovered from the irrigated field soils were two-to-three times greater than from the desert soils.

Nematodes in the omnivore/predator group were recovered in greater numbers by the SFS method (Table 1). Due to much variation, differences between the microbivorous and fungivorous nematodes

extracted by the two methods were not significant. More juveniles in the microbivorous and fungivorous groups were recovered by the BF. Plant parasites extracted by the two methods were not compared because of their low numbers in the Colorado desert soil.

Variation in nematode biomass calculations introduced by the extraction method is shown in Table 2. More nematodes with a smaller average size or weight were recovered by BF, whereas SFS recovered fewer nematodes, but of a larger size.

DISCUSSION

The sugar-flotation-sieving method (SFS) is better than the Baermann funnel for extracting nematodes from desert soils. Results from the study show that SFS extracts nematodes more representative of the community.

The SFS method is 75-80% efficient with two extractions of each 100-cm³ desert soil sample. Eighty percent of *A. avenae* added to desert soils were recovered after two SFS extractions. Seventy-five percent of the total nematode recovery from Colorado and Mojave desert soils occurred in the first two of four SFS extractions.

The BF technique has several disadvantages. There is a low recovery of

TABLE 1. Numbers of nematodes in three trophic groups extracted from desert soils by the sugar-flotation-sieving (SFS) and Baermann funnel (BF).

Trophic group	Mean no. nematodes ^a /500 cm ³		Standard deviation of differences between methods
	SFS	BF	
Microbivores	825 NS	1255	1216
Omnivores/ Predators	622*	166	655
Fungivores	325 NS	580	727

^aData are averages of 10 replicates. Asterisk (*) means significant difference, $P=0.05$, NS = difference not significant.

TABLE 2. Numbers, average weight and biomass for nematodes/m² to 20-cm depth between desert soils and an irrigated agricultural field.

	No. nematodes per 500cm ³ soil ^a		Mean nematode wt. ^{b,c} ($\times 10^{-4}$ mg) ^b		Biomass ^c (g/m ²)	
	BF	SFS	BF	SFS	BF	SFS
Mojave Desert	2820	902	1.5	2.6	.17	.09
Colorado Desert	2168	1262	1.0	1.8	.09	.09
Irrigated agricultural field	4174	2124	0.9	2.0	.15	.16

^aData are averages of 20 replicates; samples processed by Baermann funnel (BF) and sugar-flotation-sieving (SFS).

^bData are averages of 100-150 nematode measurements.

^cDetermined by Andr ssy's method.

Dorylaimina and a high recovery of small, microbivorous juveniles. The role of the various trophic groups in the total nematode community structure may therefore be misrepresented by BF. In some desert soils processed by BF, Cephalobidae represented 50-75% of the total nematodes recovered. The eggs in the soil can hatch during the 48-h period in the funnel and increase the total nematode recovery. Few eggs are recovered by the SFS method, and the influence of egg hatch is small.

Nematodes recovered by BF are active, whereas those recovered by the SFS extraction are often coiled, inactive, and apparently in a cryptobiotic state from 15 min to 1 h after addition of the sugar solution to the desert soil samples. This was determined by continuous observation of the nematodes immediately following the SFS extraction. SFS extraction appears to represent the nematodes present in the soil and their condition at the time of sampling.

Biomass as determined by Andrassy's method (1) is dependent on the size and numbers of nematodes. In most instances, extraction methods will affect biomass figures. However, our biomass figures are similar, except when there appears to be a large egg reserve as in the Mojave desert soil (Table 2). When nematodes are extracted by the BF, the increased numbers of nematodes resulting from egg hatch influence biomass and may therefore reflect the potential rather than actual nematode population.

On the basis of these studies, we have

adopted a standard technique for extracting desert nematodes by processing each 100-cm³ soil subsample two consecutive times by the SFS technique.

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