NEMATODE LOSSES DURING CENTRIFUGAL EXTRACTION FROM TWO SOIL TYPES[1]

R. McSorley and J. L. Parrado

Respectively, Department of Entomology and Nematology, IFAS, University of Florida, Gainesville, FL 32611; and Tropical Research and Education Center, 18905 S.W. 280 St., Homestead, FL 33031, U.S.A.

Accepted:

25.VIII.1987

Aceptado:

ABSTRACT

McSorley, R., and J. L. Parrado. 1987. Nematode losses during centrifugal extraction from two soil types. Nematropica 17:147-161.

Estimates of losses at selected stages of a centrifugation procedure were obtained for nematodes infesting Rockdale fine sandy loam and Perrine marl soils in southern Florida. The steps investigated ranged from concentrating a sieved subsample before centrifugation to collection of nematodes following a final centrifugation in a sugar solution. Losses in supernatant water following the first centrifugation (in water) were relatively low (<5%), except for *Helicotylenchus dihystera* from Rockdale soil (33.6%) and *Meloidogyne incognita* juveniles from marl soil (22.2%). For most nematodes, greatest losses occurred in the pellet following centrifugation in sugar solution, ranging from 7.38-29.87% on Rockdale soil and from 2.79-15.50% on marl soil, depending on species. Losses in sieving nematodes suspended in sugar solutions were low (≤6.00%). Estimates of maximum extraction efficiency over all centrifugation steps were: 85.8% for *Criconemella onoensis*, 81.6% for *Rotylenchulus reniformis*, 61.2% for *H. dihystera*, and 91.9% for *Quinisulcius acutus* on Rockdale soil; 91.5% for *C. onoensis*, 92.9% for *R. reniformis*, 77.3% for *M. incognita*, and 80.5% for *Tylenchorhynchus martini* on Perrine marl. Considering losses from the centrifugation steps as well as losses during sieving steps prior to centrifugation, maximum extraction efficiencies for sieving and centrifugation were estimated as: 55.1% for *C. onoensis*, 58.6% for *R. reniformis*, and 49.1% for *H. dihystera* on Rockdale soil; 81.3% for *C. onoensis* and 71.6% for *R. reniformis* on Perrine marl soil.

Additional key words: Criconemella onoensis, extraction efficiency, Helicotylenchus dihystera, methodology, Meloidogyne incognita, population assessment, quantitative nematology, Quinisulcius acutus, Rotylenchulus reniformis, Tylenchorhynchus martini.

RESUMEN

McSorley, R., y J. L. Parrado. 1987. Pérdidas en la extracción de nematodos por centrifugación en dos tipos de suelo. Nematrópica 17:147-161.

Las estimaciones de pérdidas en etapas selectivas del procedimiento de centrifugación fueron determinados para los nematodos que infestan los suelos de textura franco arenosa fina de Rockdale y marl de Perrine al sur de la Florida. Los pasos investigados comprendieron desde la concentración de la submuestra tamizada antes de la primera centrifugación hasta la recolección de los nematodos después de la centrifugación final en solución de azúcar. Las pérdidas en el agua residual después de la primera centrifugación (en agua) fueron relativamente bajas (<5%), exceptuando a *Helicotylenchus dihystera* en los suelos de Rockdale (33.6%) y las formas juveniles de *Meloidogyne incognita* en los suelos marl de Perrine (22.2%). Para la mayoría de los nematodos, las pédidas más grandes se produjeron

en el sedimento de suelo después de la centrifugación en solución azucarada, variando de 7.38-29.87% en los suelos Rockdale y 2.79-15.50% en los suelos marl, dependiendo de la especies de nematodo. Las pérdidas en el tamizado de los nematodos suspendidos en la solución de azúcar fueron bajas (≤6.00%). Las estimaciones de máxima eficiencia de extracción a través de todos los pasos de la centrifugación fueron: 85.8% para *Criconemella onoensis,* 81.6% para *Rotylenchulus reniformis,* 61.2% para *Helicotylenchus dihystera* y 91.9% para *Quinisulcius acutus* en los suelos Rockdale; 91.5% para *C. onoensis,* 92.9% para *R. reniformis,* 77.3% para *M. incognita,* y 80.5% para *Tylenchorhynchus martini* en los suelos marl de Perrine. Considerando las pérdidas en las etapas de extracción por centrifugación aquí investigadas, así como las pérdidas durante el proceso de tamizado ya descrito, la máxima eficiencia de extracción para tamizado y centrifugación fueron estimados asi: 55.1% para *C. onoensis,* 58.6% para *R. reniformis,* y 49.1% para *H. dihystera* en los suelos Rockdale; 81.3% para *C. onoensis* y 71.6% para *R. reniformis* en los suelos marl de Perrine.
Palabras claves adicionales: eficiencia de extracción, evaluación de la población, metodología, nematología cuantitativa.

INTRODUCTION

Quantitative nematology demands an accurate assessment of errors associated with the extraction process (5,10,14). Since its initial application by Caveness and Jensen in 1955 (4), centrifugation has been widely used to separate nematodes from soil and debris, particularly the method as modified by Jenkins (9). Efficiency of Jenkins' sieving and centrifugation method, or modifications thereof, in separating nematodes from soil has ranged from 2-65%, depending on nematode species and soil type (3,15,17). In addition to the sieving steps, the centrifugation portion of the procedure is also a multistep operation (1,13). The objective of the present study was to obtain some estimates of the losses occurring at each step in a centrifugation procedure applied to nematode extraction from two soils commonly used for vegetable production in southern Florida. A corresponding analysis of losses during sieving has been presented elsewhere (12).

MATERIALS AND METHODS

A series of experiments were conducted to estimate losses at each step of a centrifugation process. Sieving prior to centrifugation was conducted as described previously (12), and the centrifugation steps were conducted as outlined (Fig. 1).

Soil for these centrifugation experiments was obtained from vegetable fields near Homestead, Florida, and was naturally infested with plant-parasitic nematodes. The soil type used (7) was a Rockdale fine sandy loam (Lithic Eutrochrept, 40.4-52.4% sand, 21.6-31.4% silt, 26.0-28.2% clay, pH = 7.8). All experiments were repeated using Perrine marl soil (Typic Fluvaquent, 3.6% sand, 64.2% silt, 32.2% clay, pH = 7.8).

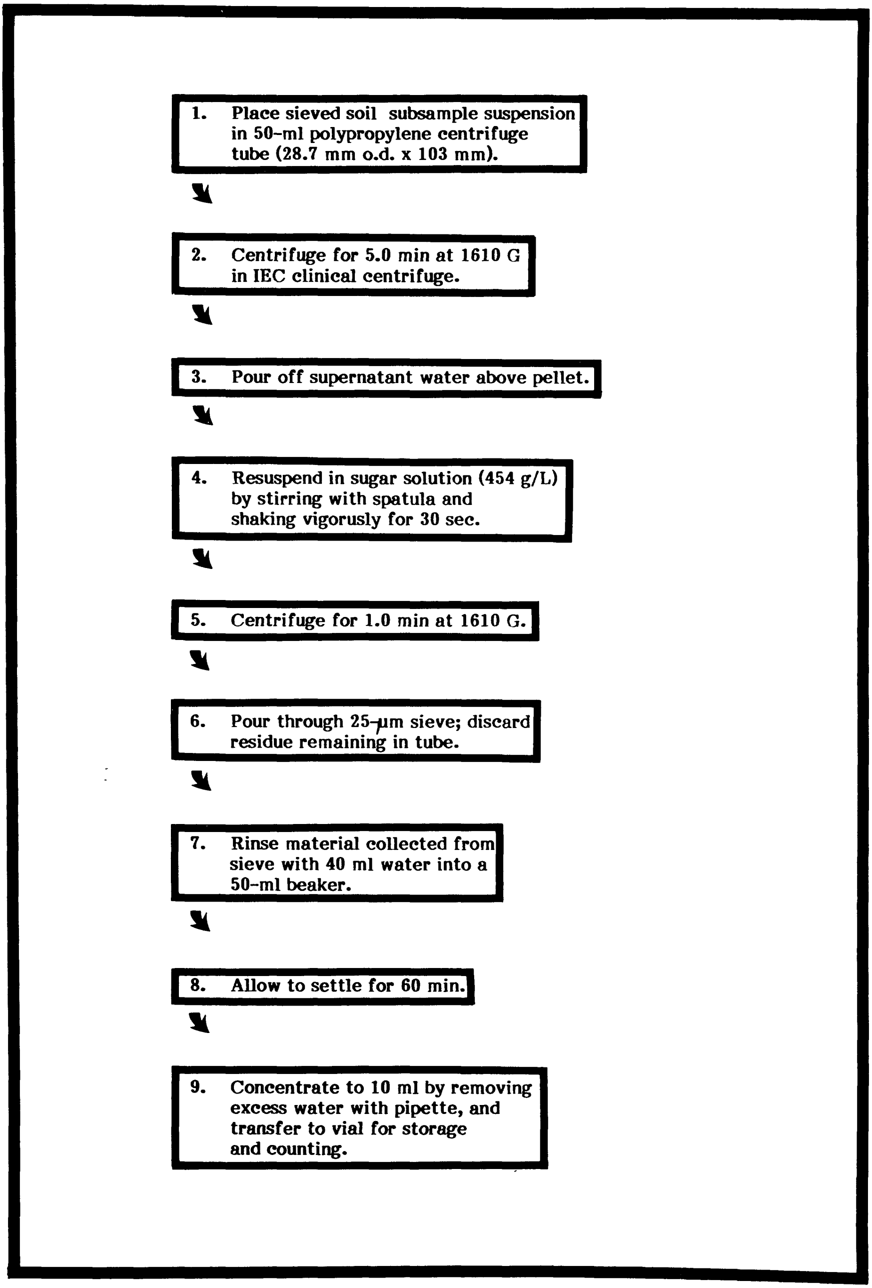

Fig. 1. Steps involved in a typical centrifugation process for extracting nematodes from sieved soil subsamples.

Usually, one step in the centrifugation process was highlighted or varied in each experiment. Other steps were as in Fig. 1, and the counts obtained following the standard procedure (Fig. 1, step #9), hereafter referred to as a "standard count," are used as a standard against which loss estimates are reported, for consistency in comparing losses from different steps and experiments. Subsamples of 100 cm^3, each drawn as a single scoop from a large bulked soil sample, provided material for all treatments and replications involved in a given experiment. Depending on the experiment, standard procedures for analysis of variance or curve fitting were used, as appropriate (6). Specific details of individual experiments are given below.

Losses in supernatant water. Samples from previous experiments (12) on settling time and decanting were used here. In those experiments, nematodes and debris washed from sieved samples were allowed to settle in 200 ml of water for various lengths of time prior to decanting to a smaller volume, which was then transferred to a 50-ml centrifuge tube for the centrifugation procedure (Fig. 1). Centrifugation steps were as outlined, except that supernatant water following the first centrifugation (Fig. 1, step #3) was saved rather than discarded. This supernatant water was passed through a 500-μm sieve to remove debris and then through a 25-μm sieve, from which nematodes were washed and counted directly. In the first experiment, 3 treatments (30, 60, or 120-min settling times) x 6 replications were used, while in the second experiment, 4 treatments (15, 30, 60 or 120-min settling times) x 4 replications were used. Both experiments were performed with Perrine marl as well as with the Rockdale soil.

Modification of the first centrifugation. Following the first centrifugation, excess water is usually poured from the tube, retaining only the pellet at the bottom (Fig. 1, step #3). Since it is possible that some nematodes at the top surface of the pellet could be disturbed and subsequently lost, a modification was introduced in which 5 ml of water above the pellet was retained after pouring the supernatant excess. Paired samples, one using the standard method and one retaining 5 ml of water above the pellet, were compared using three replicates of Rockdale soil and four replicates with Perrine marl soil. Other steps in the centrifugation process were as outlined (Fig. 1), except that a more concentrated sugar solution was added to the samples in which 5 ml of water were left, so that the resuspension (Fig. 1, step #4) was in a 454 g/L sugar solution in both cases.

Losses in the pellet following sugar centrifugation. Procedures for these experiments were very similar to those performed to estimate losses in the supernatant from the first centrifugation. Two experiments were performed with Rockdale soil and two with Perrine marl soil, utilizing the 3 treatments (30, 60, and 120-min settling times) x 6 replications or the 4 treatments (15, 30, 60, and 120-min settling times) x 4 replications described previously. Rather than discarding the residues obtained following centrifugation in sugar (Fig. 1, step #6), the residues were saved and resuspended. Subsequent extraction (Fig. 1, step #4-9) from the resuspended material was used to obtain some estimate of the number of nematodes lost in the pellet, expressed as a percent of those obtained in a corresponding standard count.

A second series of more detailed experiments were conducted to evaluate losses in the pellet following centrifugation with sugar. Four 100-cm^3 subsamples drawn from a large soil sample were suspended and sieved (12), and then centrifuged by the procedures outlined above (Fig. 1). However, instead of discarding sugar pellet residues (Fig. 1, step #6), the residues were resuspended as above and nematodes extracted. This was repeated through four sugar centrifugations, to obtain data on counts obtained from each successive centrifugation, over four replications. The experiment was performed for both Rockdale and Perrine marl soils.

Losses in pouring the sugar suspension. In addition to nematode losses in the pellet of the sugar centrifugation, losses during the sugar centrifugation can occur during the pouring of the sugar suspension itself (Fig. 1, step #6-7), if nematodes pass through the 25-μm sieve or adhere to the sieve following rinsing. To evaluate these losses, 8 replicate 100-cm^3 subsamples were drawn from a large Rockdale or Perrine marl soil sample, and extracted using the standard procedure (Fig. 1). Water poured through the 25-μm sieve (Fig. 1 step #6) was saved, concentrated, and examined directly for nematodes. After nematodes were rinsed from the 25-μm sieve in the standard procedure (Fig 1, step #7), the sieve was rinsed a second time and any nematodes obtained were counted separately from the rest of the sample.

RESULTS AND DISCUSSION

Losses in supernatant water. Numbers of nematodes lost in the supernatant water as a percent of the nematodes obtained in a "standard count" are summarized by experiment and settling time (Table 1). In general, settling time before decanting had no significant ($P = 0.05$) effect on percent of nematodes lost in the supernatant, except for *Tylenchorhynchus martini* which showed significant ($P = 0.01$) linear or quadratic decreases in nematodes lost as settling time prior to decanting increased. The reason for the greater loss at short settling times is unclear. Previous work (12) indicated that settling time did not affect pellet size in the first centrifugation or numbers of nematodes recovered in a standard count. It is possible that 15 min is insufficient time for suspended organic debris to either float to the top or sink out of suspen-

Table 1. Losses in supernatant water following the first centrifugation.

Nematode	Percent loss in supernatant by settling time (min)[w]				
	15	30	60	120	Mean[x]
Rockdale fine sandy loam, first test[y]					
Criconemella onoensis	—	0	1.39	0	0.46
Rotylenchulus reniformis	—	9.49	6.05	9.36	8.30
Helicotylenchus dihystera	—	41.42	31.80	27.59	33.60
Rockdale fine sand loam, second test[z]					
Criconemella onoensis	0.41	0.03	0.11	0.09	0.16
Rotylenchulus reniformis	1.33	0.83	0.67	1.00	0.95
Quinisulcius acutus	0.20	0	0.20	0.53	0.23
Perrine marl, first test[y]					
Criconemella onoensis	—	0.59	0.08	0.20	0.29
Rotylenchulus reniformis	—	6.11	5.11	3.09	4.89
Perrine marl, second test[z]					
Tylenchorhynchus martini	4.84	2.74	2.14	1.36	2.77
Meloidogyne incognita	17.25	28.19	18.75	21.51	22.24

[w]Losses are expressed as a percent of a standard count, i.e., the number of nematodes recovered in the standard extraction process.
[x]The mean value is averaged across all settling times. No significant ($P = 0.05$) differences with settling time except for *T. martini* in the second test on Perrine marl soil, for which significant ($P = 0.01$) linear and quadratic effects were noted.
[y]Data are means of 6 replications.
[z]Data are means of 4 replications.

sion. Organic debris from the supernatant was not measured, but it is possible that it could have trapped some nematodes or interfered with their normal settling. Losses of several nematode species in the supernatant were low to moderate, but an average of 33.60% of *Helicotylenchus dihystera* were lost from Rockdale soil samples and 22.24% of *Meloidogyne incognita* juveniles from marl soil. Among nematodes recovered from both experiments with Rockdale soil, few *Criconemella onoensis* were lost, averaging only 0.31% over both experiments, while numbers of *Rotylenchulus reniformis* lost varied in the two experiments, averaging 4.62%.

Modification of the first centrifugation. Retaining 5 ml of water above the pellet to reduce disturbance during pouring significantly ($P = 0.01$) reduced the losses in the supernatant following the first centrifugation in 3 of the 4 nematode-soil combinations evaluated (Table 2). However, this reduced loss, with 5 ml of water retained, was not great enough to have a significant ($P = 0.05$) impact on the number of nematodes counted in a standard sample when standard counts from the two techniques were compared (Table 2). Evidently, the standard procedure (Fig. 1) of pouring down to the pellet does disturb the pellet surface sufficiently to cause some (usually small) losses during pouring.

Table 2. Effect of two methods of pouring supernatant from first centrifugation on numbers of nematodes extracted and lost from 100-cm³ subsamples.

Nematode and Location	Nematodes per 100 cm³ soil[x]		
	Supernatant poured down to pellet	5 ml water maintained above pellet	t-statistic[y]
Rockdale fine sandy loam			
Rotylenchulus reniformis in standard count[z]	583	708	−1.13
in supernatant	81.7	13.3	6.93**
Supernatant as % of standard	14.01	1.88	
Quinisulcius acutus in standard count	50.0	66.7	−0.56
in supernatant	6.7	0	4.00**
Supernatant as % of standard	13.34	0	
Perrine marl			
Criconemella onoensis in standard count	1756	1825	−0.44
in supernatant	12.5	6.2	1.39
Supernatant as % of standard	0.71	0.34	
Tylenchorhynchus martini in standard count	100.0	93.8	0.17
in supernatant	13.8	3.8	3.70**
Supernatant as % of standard	13.75	4.00	

[x]Data are means of three (Rockdale soil) or four (Perrine marl) replications.
[y]Asterisks (**) indicate significant differences at P = 0.01.
[z]The standard count is the number of nematodes recovered in the standard extraction process.

Losses in the pellet following sugar centrifugation. Losses in the pellet following centrifugation in the sugar solution were not significantly (P = 0.05) affected by settling time of the sample prior to decanting for the first centrifugation (Table 3). Losses in this step varied greatly, particularly between the two experiments conducted with Rockdale soil. Losses of *C. onoensis* were extreme in the first experiment, reminiscent of the severe losses of *Criconemella* spp. observed by Henn (8) in pellets following sugar centrifugation, and perhaps, as suggested elsewhere (8), a result of alteration of the density of the sugar solution by suspended soil particles (8,16). Such a wide variation in recovery of *C. onoensis* is disturbing, especially since the Rockdale soil samples used in these two tests were very similar in their properties. Similar trends were observed with *R. reniformis* and *Q. acutus,* although of lesser magnitude than with *C. onoensis.* Thus, the extraction efficiencies for many nematodes may be very sensitive to small changes in the type of soil used.

Numbers of nematodes recovered from the pellet decreased rapidly with successive centrifugations in sugar solution (Table 4). Although a number of models explained a significant (P = 0.05) proportion of the variation observed in these experiments, best results ($r^2 = 0.842$ to 0.979) were obtained with models of the form:

Table 3. Losses in the pellet following centrifugation in a sugar solution.

Nematode	Percent loss in pellet by settling time (min)[w]				
	15	30	60	120	Mean[x]
Rockdale fine sand loam, first test[y]					
Criconemella onoensis	—	65.05	57.79	53.15	58.66
Rotylenchulus reniformis	—	8.28	9.45	13.42	10.38
Helicotylenchus dihystera	—	20.92	28.18	40.51	29.87
Rockdale fine sandy loam, second test[z]					
Criconemella onoensis	18.41	8.78	11.50	14.43	13.28
Rotylenchulus reniformis	1.67	0	0	5.00	1.79
Quinisulcius acutus	2.00	16.25	9.25	2.00	7.38
Perrine marl, first test[y]					
Criconemella onoensis	—	5.14	3.65	3.15	3.98
Rotylenchulus reniformis	—	1.67	3.44	3.36	2.79
Perrine marl, second test[z]					
Tylenchorhynchus martini	7.23	8.61	12.24	10.62	9.75
Meloidogyne incognita	2.60	3.18	8.12	11.89	7.14

[w]Losses are expressed as a percent of a standard count, i.e., the number of nematodes recovered in the standard extraction process.
[x]The mean value is averaged across all settling times. No significant ($P = 0.05$) differences with settling time.
[y]Data are means of 6 replications.
[z]Data are means of 4 replications.

$$\log_{10} y = a - b \log_{10} x,$$

where y is the number of nematodes recovered per 100 cm^3 of soil, x is the number of centrifugations in sugar, and a and b are regression coefficients (e.g., Fig. 2). Note that data from the first centrifugation are the results obtained through the usual method (Fig. 1), and are in fact the standard count. Recovery in the second centrifugation is presented (Table 4) for comparison with results from the previous section (see Table 3), and total loss (implicity assumed to be obtained after four centrifugations) is also expressed as a percent of the standard count (Table 4). Measuring losses through four sugar centrifugations instead of two provided a greater increase in loss estimates on Rockdale soil (2.87-3.75%) than on marl soil ($\leq$0.70%). Presumably, additional nematodes could be obtained (and loss estimates further increased) if additional centrifugations were performed. The equations (Table 4) indicate that recovery of only one nematode per sieving is anticipated after 8.81 and 6.05 centrifugations for *R. reniformis* and *C. onoensis* respectively, on Rockdale soil, or after 5.48 and 3.26 centrifugations of *C. onoensis* and *T. martini* on Perrine marl soil. Thus, beyond four centrifugations, addi-

Table 4. Nematode recovery from repeated sugar centrifugations of 100 cm^3 soil subsamples.

Nematode	Nematodes per 100 cm^3 soil by number of centrifugations in sugar solution[x]					Percent lost in 2nd centrifugation[y]	Percent loss in 2nd-4th centrifugations[y]	Relationship between number of nematodes per centrifugation (y) and number of centrifugations (x)[z]
	1	2	3	4	Total, #2-#4			
Rockdale fine sandy loam								
Criconemella onoensis	200	28.8	3.5	4.0	36.3	14.40	18.15	$\log_{10}y = 2.232 - 2.855 \log_{10}x$, $r^2 = 0.884$***
Rotylenchulus reniformis	1881	256	39.2	14.8	310	13.61	16.48	$\log_{10}y = 3.328 - 3.522 \log_{10}x$, $r^2 = 0.977$***
Perrine marl								
Criconemella onoensis	1881	135	9.5	3.8	148	7.18	7.88	$\log_{10}y = 3.316 - 4.486 \log_{10}x$, $r^2 = 0.979$***
Tylenchorhynchus martini	50	7.5	0	0.2	7.8	15.00	15.50	$\log_{10}y = 1.652 - 2.949 \log_{10}x$, $r^2 = 0.842$***

[x]Data are means of 4 replications.

[y]Losses are expressed as a percent of a standard count, i.e., the number of nematodes recovered in the standard extraction process, shown here by counts from the first centrifugation.

[z]Asterisks (***) indicate significant relationship at $P = 0.001$.

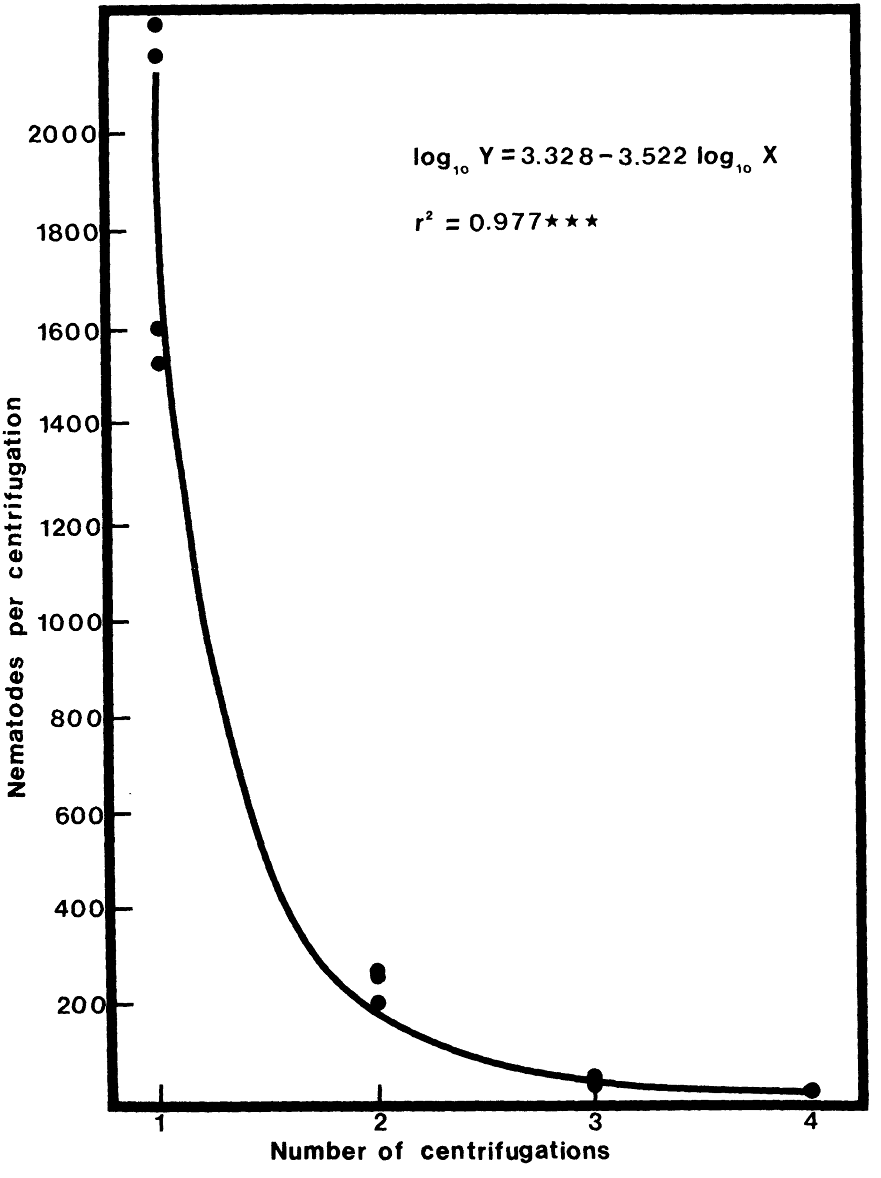

Fig. 2. Relationship between number of nematodes per 100 cm^3 soil recovered per centrifugation and number of successive centrifugations in a sugar solution, for *Rotylenchulus reniformis* from Rockdale fine sandy loam soil.

tional recovery becomes negligible. For example, if 9 centrifugations were performed for *R. reniformis* from Rockdale soil, the estimate of numbers recovered in centrifugations 5 through 9 is given by:

$$\sum_{x=5}^{9} 10^{(3.328-3.522 \log_{10} x)} = 15.77$$

This amounts to only 0.84% of a standard count, increasing the total loss estimate from 16.48 to 17.32%.

Losses in pouring the sugar suspension. Although previous work had been done to optimize sieve size and rinsing at this step (11), some nematodes still passed through the sieve and were lost (Table 5). In addition, a small percentage (up to 4.66% of a standard count) were retained on the sieve following rinsing to collect nematodes, either as a result of incomplete rinsing or because they became entangled in the meshes of the sieve (Table 5). The totals of the two types of losses at this stage were relatively low (≤6.00%).

Losses from all stages in the centrifugation process are summarized (Table 6). These were especially great for *H. dihystera,* which had a particularly heavy loss in the supernatant following the first centrifugation. Most nematodes showed their greatest losses in the pellet following sugar centrifugation. Total losses over the centrifugation steps evaluated ranged from 7.68-63.47%, depending on nematode species and soil type (Table 6). These figures could be converted to extraction efficiency (2,5), defined as number recovered divided by number present, by the relation:

$$EF = 100/(100 + y),$$

where EF is extraction efficiency and y is the number of nematodes lost per 100 counted, *i.e.,* a percent of the standard count. The efficiency of centrifugation was relatively high (≥ 77%), except for *H. dihystera,* for which efficiency was 61.2% (Table 6). As in the previous study (12), these represent maximum extraction efficiencies, since the steps used to evaluate losses do not remove all nematodes completely.

As practiced here, nematode extraction from soil involves two major portions, sieving and centrifugation. Combined losses (Table 6) are available from the data presented here and from a related study of losses incurred during sieving (12). In the case of *H. dihystera,* more nematodes were lost than counted in a sample. Extraction efficiencies computed over both sieving and centrifugation steps were somewhat lower (49.1-58.6%) for Rockdale soil than for Perrine marl (71.6-81.3%). Much of this discrepancy results in the greater efficiency of sieving Perrine marl soil, although the efficiency of centrifugation was also somewhat higher with marl soil.

Table 5. Losses of nematodes on or through a 25-μm sieve following centrifugation in sugar solution.

Nematode	Nematodes per 100 cm^3 soil[y]				Losses as percent of standard count[z]		
	Standard count[z]	Adhering to sieve	Passing through sieve	Total on or through sieve	Adhering to sieve	Passing through sieve	Total on or through sieve
Rockdale fine sandy loam							
Criconemell onoensis	78.1	1.0	0.8	1.8	1.28	0.96	2.24
Rotylenchulus reniformis	1738	13.6	12.1	25.8	0.78	0.70	1.48
Quinisulcius acutus	31.2	0.2	0.1	0.4	0.80	0.38	1.22
Perrine marl							
Criconemella onoensis	1534	12.5	4.2	16.8	0.81	0.28	1.09
Tylenchorhynchus martini	56.2	2.6	0.8	3.4	4.66	1.33	6.00

[y]Data are means of 8 replications.
[z]The standard count is the number of nematodes recovered in the standard extraction process.

Table 6. Estimated nematode losses during a centrifugation process.

Extraction (Source of data)	Losses as percent of nematodes counted in a standard count, by soil type[y]: Rockdale fine sandy loam				Perrine marl			
	Criconemella onoensis	*Rotylenchulus reniformis*	*Helicotylenchus dihystera*	*Quinisulcius acutus*	*Criconemella onoensis*	*Rotylenchulus reniformis*	*Meloidognye incognita*	*Tylenchorhynchus martini*
Losses in supernatant water (Table 1—mean of 1 or 2 tests)	0.31	4.62	33.60	0.23	0.29	4.89	22.24	2.77
Losses in sugar pellet (Table 4, or Table 3, if not in Table 4)	18.51	16.48	29.87	7.38	7.88	2.79	7.14	15.50
Loses on or through 25-μm sieve (Table 5)	2.24	1.48	—	1.22	1.09	—	—	6.00
Sum of centrifugation losses	20.70	22.58	63.47	8.83	9.26	7.68	29.38	24.27
Losses during sieving (12)	60.8	47.9	40.1	—	13.7	32.0	—	—
Total losses from sieving and centrifugation	81.5	70.5	103.6	—	23.0	39.7	—	—
Maximum efficiency of centrifugation[z]	82.8	81.6	61.2	91.9	91.5	92.9	77.3	80.5
Efficiency of sieving (12)[z]	62.2	67.6	71.4	—	88.0	75.8	—	—
Maximum efficiency of sieving and centrifugation[z]	55.1	58.6	49.1	—	81.3	71.6	—	—

[y]A standard count is the number of nematodes recovered in the standard extraction process according to steps outlined in Fig. 1. Dashes (—) indicate no data obtained.

[z]Extraction efficiency = 100/(Total loss + 100). Values in the table are expressed as percent (i.e., extraction efficiency x 100%).

The heavy losses and correspondingly low extraction efficiencies observed here are not unexpected in view of the low recoveries achieved with related sieving and centrifugation procedures in other extraction efficiency studies (3,15,17). In any multistep procedure, some losses probably occur at every step. If the number of steps cannot be minmized, then the critical steps should be identified and perhaps modified. It is evident from the work presented here that certain steps in centrifugation are subject to great losses, while relatively insignificant losses occur at other steps. In addition, steps in which losses are variable (e.g., losses in the pellet from sugar centrifugation) must be identified and modified or an understanding of the factors influencing the variability must be achieved. Future studies of extraction efficiency and resultant modifications are essential in obtaining accurate estimates of nematode populations for quantitative work.

LITERATURE CITED

1. AYOUB, S. M. 1980. Plant nematology an agricultural training aid. NemaAid Publications, Sacramento. 195 pp.
2. BIRD, G. W. 1981. Integrated nematode management for plant protection. Pp. 355-375 *in* B. M. Zuckerman and R. A. Rohde (eds.), Plant Parasitic Nemtodes. Vol. III. Academic Press, New York. 508 pp.
3. BYRD, D. W., JR., K. R. BARKER, H. FERRIS, C. J. NUSBAUM, W. E. GRIFFIN, R. H. SMALL, and C. A. STONE. 1976. Two semi-automatic elutriators for extracting nematodes and certain fungi from soil. J. Nematol. 8:206-212.
4. CAVENESS, F. E., and H. J. JENSEN. 1955. Modification of the centrifugal-flotation technique for the isolation and concentration of nematodes and their eggs from soil and plant tissue. Proc. Helminth. Soc. Wash. 22:87-89.
5. FERRIS, H. 1987. Extraction efficiencies and population estimation. Pp. 59-63 *in* J. A. Veech and D. W. Dickson (eds.), Vistas on Nematology. E. O. Painter Printing Co., DeLeon Springs, Florida. 509 pp.
6. FREUND, R. J., and R. C. LITTELL. 1981. SAS for linear models. A guide to the ANOVA and GLM procedures. SAS Institute, Inc. Cary, North Carolina. 231 pp.
7. GALLATIN, M. H., J. K. BALLARD, C. B. EVANS, H. S. GALBERRY, J. J. HINTON, D. P. POWELL, E. TRUETT, W. L. WATTS, G. C. WILSON, JR., and R. G. LEIGHTY. 1958. Soil survey (detailed-reconnaissance) of Dade County, Florida. U.S. Government Printing Office, Washington. 56 pp.
8. HENN, R. A. 1987. Development of sampling strategies for management studies of *Hoplolaimus galeatus* on St. Augustinegrass

(*Stenotaphrum secundatum*). Ph.D. dissertation, University of Florida, Gainesville. 134 pp.
9. JENKINS, W. R. 1964. A rapid centrifugal-flotation technique for separating nematodes from soil. Plant Dis. Reptr. 48:692.
10. MCSORLEY, R. 1987. Extraction of nematodes and sampling methods. Pp. 13-47 *in* R. H. Brown and B. R. Kerry (eds.), Principles and Practice of Nematode Control in Crops. Academic Press, Sydney. 447 pp.
11. MCSORLEY, R., and J. L. PARRADO. 1981. Effect of sieve size on nematode extraction efficiency. Nematropica 11:165-174.
12. MCSORLEY, R., and J. L. PARRADO. 1987. Nematode losses during sieving from two soil types. Nematropica 17:125-146.
13. SOUTHY, J. F. (ed.). 1986. Laboratory methods for work with plant and soil nematodes. Her Majesty's Stationary Office, London. 202 pp.
14. VIGLIERCHIO, D. R., and R. V. SCHMITT. 1983. On the methodology of nematode extraction from field samples: Baermann funnel modifications. J. Nematol. 15:438-444.
15. VIGLIERCHIO, D. R., and R. V. SCHMITT. 1983. On the methodology of nematode extraction from field samples: comparison of methods for soil extraction. J. Nematol. 15:450-454.
16. VIGLIERCHIO, D. R., and T. T. YAMASHITA. 1983. On the methodology of nematode extraction from field samples: density flotation techniques. J. Nematol. 15:444-449.
17. WEBER, D. E., and A. S. WILLIAMS. 1968. Evaluation of the centrifugal-flotation technique for the quantitative recovery of three types of endoparasitic nematodes. Nematologica 14:18 (Abstr.).

Received for publication:

9.VII.1987

Recibido para publicar:

[1]Florida Agricultural Experiment Station Journal Series No. 8278.