ANNUAL POPULATION CHANGES OF SOIL NEMATODES IN THE FIELD OF CONTINUOUS CROPPING OR ROTATION

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A. Observations at the Experimental Field of Fujisaka Branch of Aomori Prefectural Agricultural Experiment Station at Towasa, Aomori Prefecture.

Experimental Procedures

Experimental field : The experimental plots on the cropping systems were established in 1930 on the field of black colored volcanic ash soil. Area of a plot is 200 square meter (10 m × 20 m). Plots under continuous cropping were constructed from non-replicated 6 plots of potato, rape, wheat, barnyard millet, soybean, or corn. Plots under rotation were constructed from 6 plots under 6-years rotation of these crops (replicated 1 system), 12 plots under 3-year rotation (replicated 4 systems), and 8 plots under 2-year rotation (replicated 4 systems).

Nematode survey : Soil samples of all these plots were collected annually from 1973 to 1977 (from 38th to 42nd year after the establishment of the experimental plots) in October or early November, after harvest of the summer crops. Soil samples of each plots were composed of 5 subsamples collected from rhizosphere. Nematodes were extracted from the thoroughly mixed soil samples by replicated Baermann's funnel technique ; 20 g. wet soil was settled on a sheet of muslin cloth sheeted on a riddle within the watered funnels at 25° C for 7 days.

Results

Crop rotations of the above systems were clearly effective for the control of northern root-knot nematode, soybean cyst nematode and pin nematode, and longer the rotation systems the better. However, such an effect of the rotations was not recognized at all against to Cobb's root-lesion nematode (see Tables 1-4).

Table 1. Number of plant-parasitic nematodes per 20 g. wet soils of the continuously cropping plots of Aomori Agr. Expt. Sta., at Towada, Aomori prefecture*

Crop	Year	Nematodes**			
		P.p.	M. h.	H. g.	Para. sp.
Soybean	1973	72	69	3	90
	1974	184	89	18	36
	1975	389	117	3	11
	1976	1,530	193	10	41
	1977	279	57	54	24
Barnyard millet	1973	51	0	0	5
	1974	189	0	0	8
	1975	103	0	0	0
	1976	92	0	0	0
	1977	216	0	0	0

Rape	1973	97	0	0	0
	1974	301	0	0	0
	1975	500	0	0	0
	1976	230	0	0	0
	1977	271	0	0	0
Wheat	1973	10	0	0	0
	1974	133	0	0	0
	1975	200	0	0	0
	1976	107	0	0	0
	1977	119	0	0	0
Potato	1973	19	42	0	0
	1974	63	225	0	0
	1975	138	144	0	0
	1976	50	56	0	0
	1977	77	48	0	0
Corn	1973	390	0	0	0
	1974	515	0	0	0
	1975	627	0	0	0
	1976	967	0	0	0
	1977	890	0	0	0

* : Larvae + adults in P.p. and Para. sp., 2nd stage larvae in M.h. and H.g.
** : P.p. = *Pratylenchus penetrans,* M.h. = *Meloidogyne hapla,*
H.g. = *Heterodera glycines,* and Para. sp. = *Paratylenchus* sp.

Table 2. Number of plant-parasitic nematodes per 20 g. wet soils of the 6-years rotation plots.

Plot no.	Year	Crops cultivated*	Nematodes			
			P. p.	M. h.	H. g.	Para. sp.
1	1973	Pot.-(Rape)	50	0	0	0
	1974	(Rape)-S.s.-(W.)	374	0	0	0
	1975	(W.)-Pe-tsai	1,345	0	0	0
	1976	B.m. - (W.)	808	0	0	0
	1977	(W.)-Soybean	422	0	0	0
2	1973	Corn	247	0	0	0
	1974	Pot.-(Rape)	210	0	0	0
	1975	(Rape)-S.s.-(W.)	131	0	0	0
	1976	(W.)-Pe-tsai	113	0	0	0
	1977	B.m. - (W.)	319	0	0	0
3	1973	(Rape)-S.s.-(W.)	411	0	0	0
	1974	(W.)-Pe-tsai	2,248	0	0	0
	1975	B.m. - (W.)	553	0	0	0
	1976	(W.)-Soybean	419	0	0	0
	1977	Corn	666	0	0	0
4	1973	(W.)-Pe-tsai	316	0	0	0
	1974	B.m. - (W.)	538	0	0	0
	1975	(W.)-Soybean	123	0	0	0
	1976	Corn	1,262	0	0	0
	1977	Pot.-(Rape)	439	0	0	0

	1973	B.m - (W.)	286	0	0	0
	1974	(W.)-Soybean	345	0	0	0
5	1975	Corn	307	0	0	0
	1976	Pot.-(Rape)	177	0	0	0
	1977	(Rape)-S.s.-(W.)	149	0	0	0
	1973	(W.)-Soybean	205	0	0	0
	1974	Corn	562	0	0	0
6	1975	Pot.-(Rape)	218	0	0	0
	1976	(Rape)-S.s.-(W.)	627	0	0	0
	1977	(W.)-Pe-tsai	796	0	0	0

* : Pot. = potato, S.s. = soiling soybean, W. = wheat, and B.m. = barnyard millet.
( ) : winter crops.

Table 3. Number of plant-parasitic nematodes per 20 g.wet soils of the 3-years rotation plots.

Plot no.	Year	Crops cultivated	Nematodes			
			P.p.	Mh.	H.g.	Para.sp.
		System-I				
	1973	(W.)-Soybean	170	0	0	0
	1974	Barn. millet	340	0	0	0
1	1975	Potato-(W.)	124	0	0	0
	1976	(W.)-Soybean	324	0	0	0
	1977	Barn, millet	300	0	0	0
	1973	Barn. millet	356	0	0	0
	1974	Potato-(W.)	168	0	0	0
2	1975	(W.)-Soybean	703	0	0	0
	1976	Barn. millet	488	0	0	0
	1977	Potato-(W.)	172	0	0	0
	1973	Potato-(W.)	73	0	0	2
	1974	(W.)-Soybean	834	0	9	0
3	1975	Barn. millet	386	0	0	0
	1976	Potato-(W.)	259	0	0	0
	1977	(W.)-Soybean	448	0	0	0
		System-II				
	1973	(W.)-Pe-tsai	200	0	0	0
	1974	Corn	566	0	0	0
1	1975	Potato-(W.)	100	0	0	0
	1976	(W.)-Pe-tsai	216	0	0	0
	1977	Corn	832	0	0	0
	1973	Corn	452	0	0	0
	1974	Potato-(W.)	127	0	0	0
2	1975	(W.)-Pe-tsai	646	0	0	0
	1976	Corn	1,611	0	0	0
	1977	Potato-(W.)	197	0	0	0
	1973	Potato-(W.)	70	0	0	0
	1974	(W.)-Pe-tsai	1,195	0	0	0
3	1975	Corn	803	0	0	0
	1976	Potato-(W.)	259	0	0	0
	1977	(W.)-Pe-tsai	1,093	0	0	0

Table 3. continued.

Plot no.	Year	Crops cultivated	Nematodes P.p.	M.h.	H.g.	Para. sp.
		System-III				
1	1973	(W.)-Clover	110	0	0	0
	1974	Corn	636	0	0	0
	1975	Potato-(W. + Clover)	80	0	0	0
	1976	(W.)-Clover	368	10	0	0
	1977	Corn	780	0	0	0
2	1973	Corn	299	0	0	0
	1974	Potato-(W. + Clover)	186	0	0	0
	1975	(W.)-Clover	265	213	0	0
	1976	Corn	1,093	0	0	0
	1977	Potato-(W. + Clover)	112	0	0	0
3	1973	Potato-(W. + Clover)	22	0	0	0
	1974	(W.)-Clover	674	304	0	0
	1975	Corn	238	0	0	0
	1976	Potato-(W. + Clover)	178	0	0	0
	1977	(W.)-Clover	321	0	0	0
		System-IV				
1	1973	(Rape)-S.s.-(W.)	191	0	0	0
	1974	(W.)-Soybean	353	0	0	0
	1975	Potato-(Rape)	85	0	0	0
	1976	(Rape)-S.s.-(W.)	636	0	0	0
	1977	(W.)-Soybean	454	0	0	0
2	1973	(W.)-Soybean	128	0	0	0
	1974	Potato-(Rape)	237	0	0	0
	1975	(Rape)-S.s.-(W.)	96	0	0	0
	1976	(W.)-Soybean	538	0	0	0
	1977	Potato-(Rape)	264	0	0	0
3	1973	Potato-(Rape)	158	0	0	0
	1974	(Rape)-S.s.-(W.)	252	0	0	0
	1975	(W.)-Soybean	431	0	0	0
	1976	Potato-(Rape)	438	0	0	0
	1977	(Rape)-S.s.-(W.)	132	0	0	0

Table 4. Number of the nematodes per 20 g.wet soils in the 2-years rotation plots.

Plot no.	Year	Crops cultivated	Nematodes P.p.	M.h.	H.g.	Para. sp.
		System-I				
1	1973	(W.)-Soybean	147	0	0	55
	1974	Barn. millet-(W.)	134	3	0	16
	1975	(W.)-Soybean	253	0	0	0
	1976	Barn. millet-(W.)	172	3	0	0
	1977	(W.)-Soybean	261	0	0	30
2	1973	Barn. millet-(W.)	80	0	0	11
	1974	(W.)-Soybean	414	0	5	5
	1975	Barn. millet-(W.)	87	0	0	0
	1976	(W.)-Soybean	400	0	0	40
	1977	Barn. millet-(W.)	168	0	0	27

		System-II				
	1973	(W.)-Soybean	135	0	0	0
	1974	Potato - (W.)	140	0	0	0
1	1975	(W.)-Soybean	454	0	0	0
	1976	Potato - (W.)	112	0	0	0
	1977	(W.)-Soybean	384	0	3	16
	1973	Potato - (W.)	63	0	0	2
	1974	(W.)-Soybean	912	0	8	13
2	1975	Potato - (W.)	170	0	0	0
	1976	(W.)-Soybean	536	0	0	70
	1977	Potato-(W.)	110	0	5	0
		System-III				
	1973	(W.)-Buckwheat	143	0	0	0
	1974	Potato - (W.)	209	0	0	0
1	1975	(W.)-Buckwheat	759	0	0	0
	1976	Potato - (W.)	132	0	0	0
	1977	(W.)-Buckwheat	300	0	0	0
	1973	Potato - (W.)	31	0	0	0
	1974	(W.)-Buckwheat	521	0	0	0
2	1975	Potato - (W.)	187	0	0	0
	1976	(W.)-Buckwheat	181	0	0	0
	1977	Potato - (W.)	151	0	0	0
		System-IV				
	1973	(Rape)-Buckwheat	152	0	0	0
	1974	Potato-(Rape)	354	0	0	0
1	1975	(Rape)-Buckwheat	374	0	0	0
	1976	Potato-(Rape)	271	0	0	0
	1977	(Rape)-Buckwheat	386	0	0	0
	1973	Potato-(Rape)	52	0	0	0
	1974	(Rape)-Buckwheat	297	0	0	0
2	1975	Potato-(Rape)	239	0	0	0
	1976	(Rape)-Buckwheat	204	0	0	0
	1977	Potato-(Rape)	241	0	0	0

B. Observations at the Experimental Field of Central Agricultural Experiment Station, MAF, at Konosu, Saitama Prefecture.

Experimental Procedures

Experimental field : The experimental plots were established in 1972 on a field of "Kanto" loam (volcanic ash) soil where soybean and wheat had been cropped continuously for several years, as a summer crop and a winter crop, respectively. Area of a plot is 100 square meters (5 m × 20). Plots under continuous cropping were constructed from non-replicated 4 plots of upland rice, taro, peanut, or sweet potato. The crops were selected from point of view of local profitability as field crops for these area. A plot under rotation was planted with above mentioned crops as 4-year rotation.

Nematode servey : Same to A.

Results

Occurrence of specific nematode parasites were observed on each of the plots under successive cropping since 2nd or 3rd year of the experiment.

Four-years rotation of these crops impeded the occurrence of the parasitic nematodes; ring nematode, upland rice cyst nematode, northern root-knot nematode, southern root-knot nematode, and coffee root-lesion nematode (see Table 1).

Table 1. Number of nematodes per 20g. wet soils of the harvested plots in autumn*.

Nematodes	Year	Continu. crop.				Rotation					
		A	B	C	D	A	B	C	D	A	B
Aphelenchoidea spp.	1972	194	10	86	526	134					
	1973	118	25	275	244		38				
	1974	38	130	58	134			60			
	1975	31	63	57	245				243		
	1976	1	4	3	5					5	
	1977	1	3	7	18						12
Criconematidae spp.	1972	2	0	0	1	3					
	1973	26	0	0	0		0				
	1974	20	0	0	0			0			
	1975	7	0	0	0				0		
	1976	2	1	0	0					2	
	1977	9	0	0	0						1
Ditylenchus spp.	1972	7	4	5	8	86					
	1973	28	1	7	3		4				
	1974	11	4	6	4			5			
	1975	0	0	1	1				1		
	1976	3	0	0	0					13	
	1977	150	1	2	4						49
Helicotylenchus spp.	1972 ~ 1975	0	0	0	0	0	0	0	0		
	1976	0	0	0	1					0	
	1977	0	0	0	10						0
Heterodera elachista	1972	0	0	0	0	0					
	1973	54	0	0	0		0				
	1974	248	0	0	0			0			
	1975	61	0	0	0				0		
	1976	31	0	0	0					7	
	1977	151	0	0	0						5
Meloidogyne hapla	1972	0	0	0	0	0					
	1973	0	0	0	0		0				
	1974	0	0	1	0			0			
	1975	0	0	48	0				0		
	1976	0	0	15	0					0	
	1977	0	0	64	0						0
Meloidogyne incognita	1972	2	0	4	11	0					
	1973	0	0	0	39		0				
	1974	0	0	0	314			0			
	1975	0	0	0	85				5		
	1976	0	0	0	91					0	
	1977	0	0	0	89						0
Pratylenchus coffeae	1972	0	0	0	0	0					
	1973	0	0	0	0		0				
	1974	0	47	0	0			0			
	1975	0	9	0	0				0		
	1976	0	117	0	0					0	
	1977	0	59	0	0						0
Tylenchus spp.	1972	23	2	6	7	48					
	1973	52	2	12	11		8				
	1974	88	4	24	21			57			
	1975	1,006	5	12	43				48		
	1976	12	2	2	1					18	
	1977	54	3	66	41						80
Unidentified saprophagous spp.	1972	327	194	194	424	257					
	1973	464	214	362	360		206				
	1974	373	291	296	262			326			
	1975	174	190	217	198				343		
	1976	208	222	73	70					424	
	1977	1,136	185	368	352						567

*Total number of larval and adult stages except only 2nd stage larvae in the cyst nematode and the root-knot nematodes.
Continu. crop. = plots under continuous cropping,
Rotation = a plot under the rotation.
A = upland rice, B = taro, C = peanut, D = sweet potato.

Studies on a bacterial (sporozoan) parasite of the root-knot nematodes.

Table 1. Percent of parasitized nematodes after 10 days incubation at 28 C in the soil infested densely with the bacteria (sporozoa).

Nematodes (Stages)	% of parasitized nematodes	Nematodes (Stages)	% of parasitized nematodes
Aphelenchoides sp. (L & A)	0.0	*Pratylenchus coffeae* (L & A)	0.0
Aphelenchus sp. (L & A)	0.0	*P. penetrans* (L & A)	0.0
Helicotylenchus sp. (L & A)	0.0	*P. vulnus* (L & A)	0.0
Heterodera elachista (2L)	0.0	*Paratrichodorus porosus* (L & A)	0.0
Meloidogyne hapla (2L)	96.7	*Tylenchulus semipenetrans* (L & A)	0.0
M. incognita (2L)	100.0	*Tylenchus* spp. (L & A)	0.0
M. javanica (2L)	97.7	Saprophagous spp. (L & A)	0.0

Table 2. Degradation of parasitic activity of the bacteria (sporozoa) stored in sealed bottles with wet or air dried soil at different temperature conditions.*

Bacterial soil**	Period of storage	− 15 C	5 C	25 C
Wet	0 month	100	100	100
	2	74	79	76
	6	72	71	74
	24	4	5	3
Air dried	0	100	100	100
	2	63	58	55
	6	—	—	—
	24	6	6	3

* : Relative activities measured by the parasitism to *Meloidogyne* larvae inoculated to the stored soils.
** : Water contents : Wet = 22.7 %, Air dried = 5.3 %.

Table 3a. Soil temperature and parasitism of the parasite to *Meloidogyne incognita.*

Soil temp.	42 days after inoculation (host plant free)*			
	Inoculated**		Control***	
	No. of larvae recoverd****	% of parasitized larvae	No. of larvae recovered****	% of parasitized larvae
20 ± 2 C	24	54	52	0
25 ± 2 C	16	58	24	0
30 ± 2 C	30	47	36	0

* Potted soils were covered with vinyl sheet.
** : Mixed 3.5 l of healthy *M. incognita* infested soil with o.5 l of the diseased (by the bacteria) *M. incognita* soil and potted. 2 reps.
*** : 4.0 l of the healthy *M. incognita* infested soil per pot. 2 reps.
**** Number of *M. incognita* larvae recovered by Baermann's funnel method per 20 g. wet soil.

Table 3b. Soil temperature and parasitism of the parasite to *Meloidogyne incognita.*

Soil temp.	3 month after tomato seedlings were planted at 42nd day					
	Inoculated			Control		
	Root-knot index	No. of larvae recovered	% of parasitized larvae	Root-knot index	No. of larvae recovered	% of parasitized larvae
20 ± 2 C	20	29	38	40	73	0
25 ± 2 C	47	84	40	80	649	0
30 ± 2 C	80	591	43	100	1,953	0

Table 4. Amount of the inoculum and the effect on suppression of *Meloidogyne incognita* density.*

Amount of soils mixed per pot		No. of galls and/or egg masses per 2 g. of rootlet	No. of larvae recovered per 20 g. of wet soil
HM**	BM***		
10.0kg	0.0kg	180	1,632
9.9	01	81	889
9.5	0.5	75	511
9.0	1.0	48	384
8.0	2.0	18	139

* : Susceptible sweet potato were grown for one year in greenhouse. 1/2,000 a pot, 3 reps.
** : Healthy *M. incognita* infested soil.
*** : The parasite-parasitized *M. incognita* infested soil.

Table 5. Effect of the parasite on a population of *Meloidogyne incognita* in potted plants.*

Treatment**	Sweet potato (April, 1974-Feb., 1975)		
	Root-knot index	No. of larvae per 20 g. soil	% of parasitized larvae
Inoculated	53	268	84.0
Control	88	556	0.0

Tomato (Feb., 1975-June, 1975)				
Height of plant	% of dead plant	Roo-knot index	No. of larvae per 20 g. soil	% of parasitized larvae
110cm	0	50	238	96.8
63	100	100	1,678	0.0

Sweet potato (June, 1975-Sept., 1975)		
Root-knot index	No. of larvae per 20 g. soil	% of parasitized larvae
25	9	57.8
100	144	0.0

Sweet potato (May, 1976-Nov., 1976)		Sweet potato (May, 1977-Nov., 1977)	
No. of larvae per 20 g.soil	% of parasitized larvae	No. of larvae per 20 g.soil	% of parasitized larvae
82	57.3	60	82.9
697	0.0	397	0.0

* : 1/2,000 a pot, 6 reps.

** : Inoculated = Mixed 0.5 kg.of the parasite infested soil with 9.5 kg.of the healthy *M. incognita* infested soil for each pot at Feb., 1974; Control = 10.0 kg.of the healthy *M. incognita* infested soil per pot.

Effect of temperature on parasitic ability of the root-knot nematode, *Meloidogyne incognita*.

Single egg mass of the root-knot nematode, *Meloidogyne incognita,* was collected from three districts in Chiba Prefecture in which different crops were cultivated. Each population, reproduced on a susceptible variety of tomato for three or four generations, was inoculated to resistant and susceptible varieties of tomato (res. : "Kyoryoku-goko", sus. : "Fukuju No. 2"), sweet potato (res. : "Norin No. 2", sus. : "Kokei No.14") and tobacco (res. : "NC 95", sus. : "Hicks No. 2") at different temperatures (25 to 33 C). Root-knot index of two populations, "Genshu" and "Kaijo", on three crops was higher on the susceptible varieties than the resistant ones at these temperatures, and it was raised by higher temperature. The "Oami" population, however, indicated a high root-knot index only on resistant variety of tomato as well as on a susceptible one. Reaction on other crops was similar to the other two populations. The parasitic ability to resistant variety in successive generations of which reproduced on resistant variety at higher temperature rose apparently in "Genshu" population to tomato and in "Kaijo" population to sweet potato. Finally parasitic ability to resistant variety of the populations developed as same as to susceptible one under lower temperature (see Figs. 1-4).

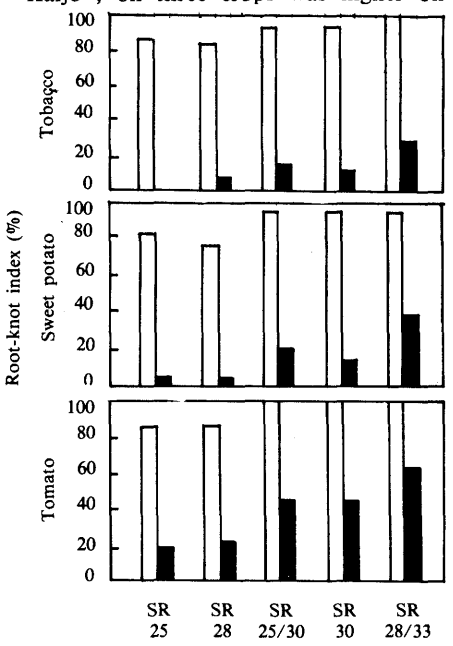


Fig. 1. Effect of temperature on the parasitic ability of Genshu population to susceptible or resistant varieties of three crops.
S : Susceptible variety, R : Resistant variety. 25/30, 28/33 : 12 hr. condition of dark/light.

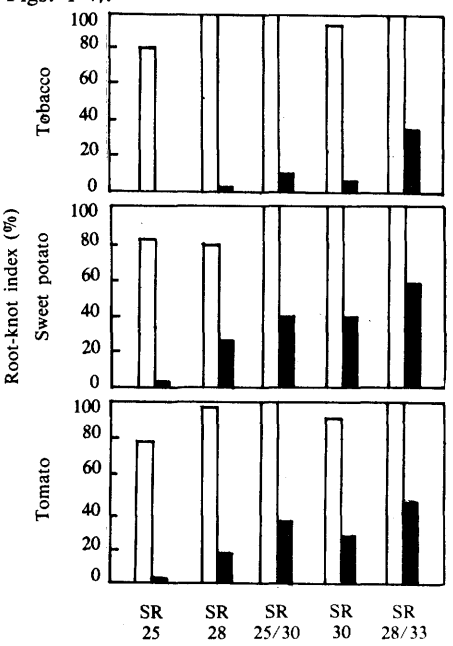
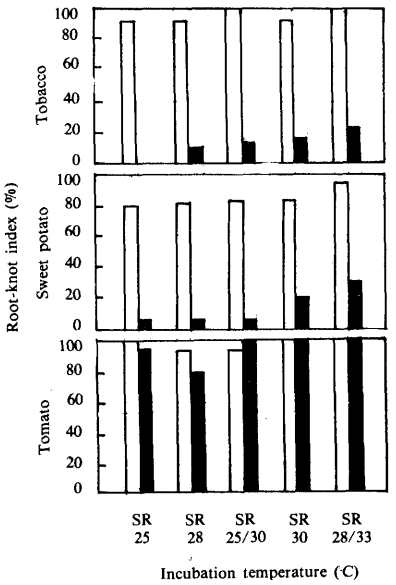


Fig. 2 Effect of temperature on the parasitic ability of Kaijyo population to susceptible or resistant varieties of three crops.
S : Susceptible variety, R : Resistant variety. 25/30, 28/33 : 12 hr. condition of dark/light.



3. Effect of temperature on the parasitic ability of Oami population to susceptible or resistant varieties of three crops. S : Susceptible variety, R : Resistant variety. 25/30, 28/33 : 12 hr. condition of dark/light.

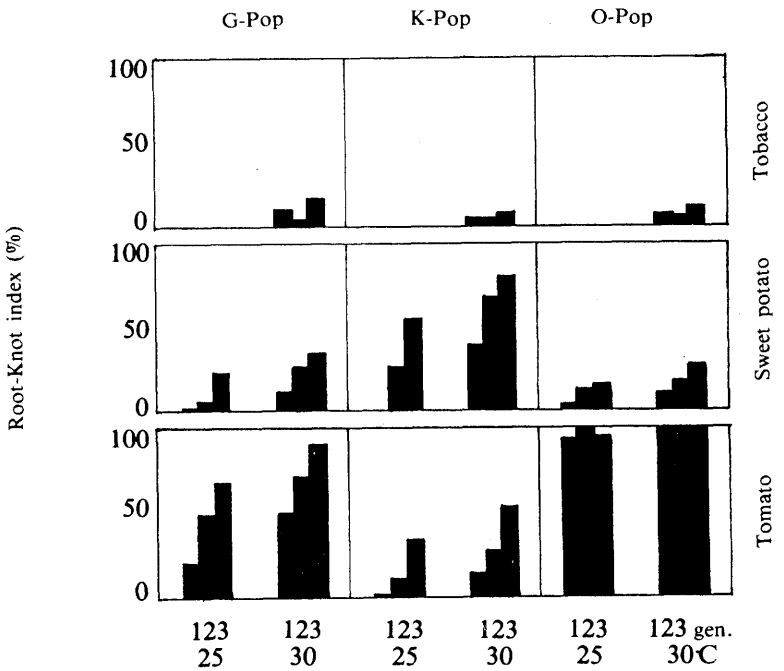


Fig. 4. Changes of parasitic ability in successive generation of three root-knot nematode populations to three resistant crops at different temperatures. gen. : generations, G-Pop : Genshu population, K-Pop : Kaijo population, O-Pop : Oami population.

Meloidogyne collection in Japan for the IMP.*

	Locality		Host plant	Identified or suspected species name
	Prefecture	City or Town		
1	Hokkaido	Memuro	Azuki bean	*Meloidogyne hapla*
2	"	Akaigawa	Potato	*M. hapla*
3	"	Yoichi	Apple	*M. mali*
4	Aomori	Towada	Soybean	*M. hapla*
5	Yamagata	Sakata	Cucumber	*M. hapla*
6	Nagano	Matsushiro	Chinese yam	*M. javanica*
7	"	Misato	Apple	*M. mali*
8	Tochigi	Senjogahara	Strawberry	*M. hapla*
9	Saitama	Konosu	Sweet potato	*M. incognita*
10	"	"	Mulberry	*M. incognita*
11	"	Kitamoto	Peanut	*M. hapla*
12	Chiba	Yachimata	Cucumber	*M. incognita*
13	Shizuoka	Omaezaki	Sweet potato	*M. incognita*
14	Aichi	Hekinan	Carrot	*M. incognita*
15	Tottori	Sakaiminato	Welsh onion	*M. incognita*
16	Shimane	Taisha	Grape vine	*M. incognita*
17	"	"	Chinese yam	*M. incognita*
18	"	"	Mulberry	*M. thamesi*
19	"	Matsue	Spinach	*M. incognita*
20	"	Izumo	Peach	*M. incognita*
21	Okayama	Okayama	Chrythanthemum	*Hypsoperine* sp.?
22	Hiroshima	Oshirabe	Spinach	*M. incognita*
23	Nagasaki	Aino	Potato	*M. hapla*
24	Kumamoto	Nishigoshi	Sweet potato	*M. incognita*
25	"	"	Tobacco	*M. javanica*
26	"	Aso	(Natural grassland)	*M. hapla*
27	Miyazaki	Shintomi	Seet potato	*M. incognita* (resistant braking)

* : As of December, 1977.

Discussion

Madamba: How do you propagate the nematode parasites and what method do you use for inoculation? Do you think it is an obligate parasite?

Nishizawa: First we obtain healthy 2nd stage larvae of *Meloidogyne*. The larvae are introduced to the soil where the parasites are present. The parasites are thought to be obligate ones. They persist in the soil as long as 2 years, may be as a type of cysts.

Chunram: What technique do you use to separate nematodes from soil?

Nishizawa: Usually, I use direct Baermann's funnel method. In special cases I use sieving and decantation technique or centrifugal floatation technique.

Sasser: Are you continuing the immunological study for identifying those local populations which you found in Japan?

Nishizawa: Yes, I am doing it.