Journal of Nematology 20(2):184–190. 1988.


# Current Research on the Major Nematode Problems in Japan[1]

M. Ichinohe[2]

*Abstract:* Among important nematode species occurring in Japan, current research achievements with the following four nematodes are reviewed: 1) Soybean cyst nematode (SCN), *Heterodera glycines*—breeding for resistance, race determination, association with *Cephalosporium gregatum* in azuki bean disease, and isolation of hatching stimulant. 2) Potato-cyst nematode (PCN), *Globodera rostochiensis*—pathotype determination (Ro 1), breeding for resistance, and control recommendations. 3) Pinewood nematode (PWN), *Bursaphelenchus xylophilus*—primary pathogen in pine wilt disease, life cycle exhibiting a typical symbiosis with Japanese pine sawyer, *Monochamus alternatus*, and project for control. 4) Rice root nematodes (RRN), *Hirschmanniella imamuri* and *H. oryzae*—distribution of species, population levels in roots, and role of these nematodes in rice culture.
*Key words: Bursaphelenchus xylophilus, Globodera rostochiensis,* glycinoeclepin A, hatch stimulant, *Heterodera glycines, Hirschmanniella imamuri, Hirschmanniella oryzae,* Japanese pine sawyer, *Monochamus alternatus,* pathotype, pine wilt disease, pinewood nematode, potato-cyst nematode, rice root nematode, soybean cyst nematode, symbiosis.

Contributions to the science of nematology in Japan have been achieved mostly in the last 30 years. A recently compiled bibliography of Japanese nematology contained 6,624 references since 1879, of which 5,492 or 83% were published between 1955 and 1984 (20).

Around 1955 the agricultural administration in Japan began to endeavor to raise production of field crops, such as vegetables, beans, sugarbeets, and cereals, instead of paddy rice which had been promoted by the government since World War II. In each prefecture, nematode specialists began to examine soil samples for the first time from nurseries and fields on a nationwide scale.

In 1971 the Japanese Nematological Society was founded, and since then 16 volumes of the *Japanese Journal of Nematology* have been issued, including nearly 200 articles concerning the taxonomy, bionomics, and control of various nematode groups. New approaches to nematology appear, particularly in studies of insect-parasitic nematodes, biological control utilizing microorganisms, physiology of *Caenorhabditis,* and others.

This paper reviews the current research conducted in Japan since the 1960s on some of the most important nematode species.

## Soybean Cyst Nematode

The soybean cyst nematode (SCN), *Heterodera glycines* described by the author (6) in 1952, has been causing serious damage to soybean and azuki beans throughout Japan since the first discovery in 1915 (5). The characteristic symptom of affected soybeans is yellow, round patches of plants occurring in spots in a field approximately 50 days after seed germination. Later the patches gradually enlarge in diameter.

Breeding programs for soybeans resistant to SCN have been conducted since the 1950s in three regions—Hokkaido, northeastern mainland, and central mainland—utilizing soybean maturity groups 0–II, III–V (mostly IV), and IV–V, respectively. Among hundreds of cultivars or strains of soybeans tested, a native selection called 'Geden-shirazu' was found to be highly tolerant to SCN (13), and from this selection a new cultivar 'Nema-shirazu' was released

Received for publication 20 May 1987.
[1] Symposium paper presented at the annual meeting of the Society of Nematologists, 19–22 July 1987, Honolulu, Hawaii.
[2] Formerly, Hokkaido National Agricultural Experiment Station. Hitsujigaoka 1, Toyohira-ku, Sapporo, Japan 004. Present address: Technical Advisor, Ishihara Sangyo Kaisha, Ltd., 10-30, Fujimi 2-Chome, Chiyoda-Ku, Tokyo, Japan 102.
The author thanks R. McSorley, University of Florida, Gainesville, Florida; T. Nishizawa, National Institute of Agro-Environmental Sciences, Tsukuba, Japan; and Y. Mamiya, Forestry and Forest Products Research Institute, Tsukuba, Japan, for reviewing the manuscript, and N. Minagawa and T. Mizukubo from National Institute of Agro-Environmental Sciences, Tsukuba, Japan, for helpful assistance and comments.

in 1961 in the northeastern mainland and 'Toyosuzu' in 1966 in Hokkaido (29).

Since the maturation date of Nema-shirazu is rather late, breeding of earlier maturing soybeans was attempted by inducing mutations with $^{60}Co$ gamma irradiation of seeds. In 1966 two cultivars were developed: 'Raiden' which matures 25 days earlier than Nema-shirazu and 'Raiko' which matures 15 days earlier, usually about 25 October (25). The highly resistant 'Peking' has been used as a gene source since the 1960s, and a resistant cultivar named 'Suzuhime' was released in Hokkaido in 1980 (30). This cultivar is highly resistant to races 1, 3, and 5, which are common in Japan.

Race determination in Japan began in 1979, and so far races 1, 3, and 5 have been identified (10,11,21,26). Similar studies have been concentrated in Tokachi Province of Hokkaido, the largest soybean growing area in Japan, and 72 out of 84 populations tested were race 3, 11 populations were race 1, one was race 5, and none were races 2 or 4 (27).

A disease of azuki bean in Hokkaido, called defoliation disease and caused by the soil-born fungus *Cephalosporium gregatum,* has become so serious that 66% of the azuki bean acreage was affected in 1970 and 85% in 1976, resulting in a drastic decrease in azuki bean cultivation in the last decade. Affected plants exhibit early defoliation following discoloration and wilting of lower leaves in late August, which then gradually proceeds upwards. In the course of the investigation, the incidence of the disease appeared to be closely associated with nematodes, because treatment with nematicides, such as ethylene dibromide or D-D, resulted in perfect control of the disease (1). A recent investigation (23) reported that the rate of plant infection significantly increased when plants were inoculated with both fungal conidia and second-stage juveniles of SCN, relative to those inoculated with fungus alone. In this study, tests involving inoculation with the two organisms at different times indicated that the highest disease severity occurred when both the fungus and nematode were inoculated simultaneously. Incidentally, this fungus is pathogenic only to azuki beans and not to soybeans or other legumes.

In 1966 hatching of SCN was shown to be stimulated by the root diffusates of host plants such as soybean, azuki bean, and kidney bean (31). Since 1967 attempts to isolate the substance stimulating hatch have continued. The research was quite successful and isolated a substance which was named glycinoeclepin A (19). Its chemical structure was also determined (3). Hatching tests indicated that this substance was efficient at concentrations as low as $10^{-11}$–$10^{-12}$ g/ml of pure glycinoeclepin A in water at 25 C.

The first step of this investigation was to compare the intensity of hatch stimulation of root diffusates from different hosts; kidney bean roots were considered the most efficient source of this material. Kidney beans were grown on 2 or 3 ha every year, and roots were collected at the flowering stage in late July. The roots were washed, fully dried, pulverized, and preserved. Approximately 1,000 kg of roots were obtained from 10 ha, and water was extracted at below 10 C during winter; this yielded 1 kg of hatch-stimulating concentrate. After fractionation and repeated chromatography, guided by bioassays, 1.25 mg of crystalline para-bromophenacyl ester, containing 0.5 mg of pure glycinoeclepin A was isolated. This amount of product enabled the determination of the chemical structure.

## Potato-cyst Nematode

In July 1972 the potato-cyst nematode (PCN), *Globodera rostochiensis,* was found for the first time in potato fields in the western corner of Hokkaido, the northernmost island of Japan. Since the nematode was first found around the small village of Makkari, this population is called the Makkari population to distinguish it from other populations or pathotypes. Five years later PCN was also detected in eastern Hokkaido. The first survey in 1972 revealed 150 ha of infested potato fields; 7 years later the infestation amounted to more than 2,000 ha

(32). Surveys made later than 1979 indicated an annual increase of approximately 350 ha, and by 1985 nearly 4,300 ha were infested.

By courtesy of Rothamsted Experimental Station, England, the Makkari population was compared morphologically and physiologically with British pathotypes A, B, and E, one of which (A) was *G. rostochiensis,* but two of which (B and E) were *G. pallida* (28).

Among several morphological characters checked, the difference in stylet length of the second-stage juvenile was most distinctive between two pathotype groups; one group 21–22 μm long included the Makkari population and British pathotype A, and another 23–25 μm long included British pathotypes B and E. Differential host studies indicated that the Makkari population was the same as British pathotype A, producing only a few females on 'Maris Piper' which possesses $H^1$ genes. From these results, we consider the Makkari population to be *Globodera rostochiensis* pathotype Ro 1 (8).

Since our preliminary tests showed that none of our conventional varieties were resistant to PCN, we immediately started a breeding program. Many potato varieties, strains, and wild species from Germany, England, and Holland were used. To screen for nematode resistance, we employed the new paired tuber method (12) in which a pair of tubers produced by an $F_1$ plant was given the same code number and divided into two groups. One group was used in resistance screening tests in isolated greenhouses, and the other was kept at 2 C in the breeding laboratory. After the initial screening tests, tubers of the latter group could be selected for further agronomic examination if necessary. This method completely eliminated the danger of nematode contamination of potato breeding fields. More than 100,000 tubers were tested, and several promising varieties or lines have been selected. In 1986 the first resistant variety named 'Toyoakari' (22) and two more in 1987 were registered by the Ministry of Agriculture, Forestry, and Fisheries. The female parent of Toyoakari is 'Tunika', and the male parent is a susceptible line of the wild type, with resistance to late blight and powdery scab diseases.

Nematode population densities after planting a host or a nonhost were compared with preplant densities. The population increased sevenfold on a susceptible variety and decreased to 30% on the resistant variety Tunika, a larger decrease than obtained with nonhosts (32). Application of oxamyl (1% granules) to nematode-infested fields increased tuber yields by 30–80% depending on the time of application, dosage, and year (32). Based on the field tests conducted since 1972, Yamada (32) recommended the following control measures against PCN: 1) Since countermeasures vary depending on the nematode density, the nematode population level in a field is measured and designated as low, moderate, or high, if it contains 10 or fewer, 11–99, or 100 or more eggs per gram dry soil, respectively. 2) Crop rotation of at least 4 years is to be practiced in every potato field. A conventional and recommended sequence in Hokkaido is wheat–bean–sugarbeet–potato. Incidentally, the sugarbeet cyst nematode, *Heterodera schachtii,* has not been detected in Japan despite a large sugarbeet acreage. 3) Potato can be planted only on low level fields, possibly with an alternate planting of a resistant and a susceptible potato variety once every 4 years; with the susceptible potato variety, oxamyl granules should be applied at planting time. 4) For moderate and high level fields, autumn treatment with D-D and planting of nonhosts the following year should be made.

## Pinewood Nematode

Great interest has been aroused since the discovery in 1971 of the pinewood nematode (PWN), *Bursaphelenchus xylophilus,* as the causal agent of the pine wilt disease which had long been attributed to insect parasites (14). In 1979 association of PWN with pine wilt disease was also reported in the United States (2). Although a number of references dealing with PWN are available, most have been summarized in reviews by Ikeda (9) and Mamiya (16–18) on which the following review is mostly based.

The first occurrence of pine wilt in Japan was in 1905, when Yano (33) reported it from Nagasaki, Kyushu. He did not determine the cause of the disease, but described disease symptoms that corresponded well with those known currently to be induced by PWN. Pine wilt has spread throughout Japan in stands of three native pines—*Pinus densiflora, P. thunbergii,* and *P. luchuensis.* In 1984 the area infested by the disease was estimated to have reached 610,000 ha, which was 24% of the nation's total of 2.6 million ha of pine forest. Loss of pine wood by PWN was estimated to be 2,430,000 $m^3$ in 1979, decreasing to 1,260,000 $m^3$ in 1985.

During the early 1960s, entomologists came to the conclusion that insects did not play a primary role in pine wilt disease, because the trees were already affected at the time of insect attack by oviposition, although the trees showed no external symptoms (18). Research into the onset of the disease prior to insect attack was then initiated.

In 1971 Kiyohara and Tokushige (14) demonstrated death of trees induced by nematode suspensions injected into holes in stems, branches, or roots and suggested that PWN was the causal organism of pine wilt disease. Death of trees occurred 40–60 days after inoculation during the summer. Dead trees contained nematodes throughout stems, branches, and roots. Old pine trees as well as young seedlings were affected by PWN to the same degree. Three pine species native to Japan are highly susceptible, while no mortality has been observed on stands of *P. taeda* and *P. elliottii* even where they are surrounded by heavily damaged Japanese pines.

Quick death of a nematode-infected tree is characteristic of pine wilt disease, and quite commonly, affected pine trees that appear healthy in early summer show a reddish-brown foliage and die in late summer. The first detectable internal symptom is a reduction and cessation of oleoresin exudation. A simple technique for measuring oleoresin exudation flow, developed for pine bettle research (24), was found useful in diagnosing diseased trees at an early stage, even when no external symptoms appeared. Oleoresin exudation in seedlings ceases completely within 6–9 days after nematode inoculation. Transpiration from foliage of affected trees decreases 20–30 days after inoculation, and foliage wilts immediately.

Under natural conditions, nematode infection of pine trees with dauer juveniles occurs via wounds made by maturation feeding of pine sawyer, *Monochamus alternatus,* on living twigs. The transmitted dauer juveniles invade the wood tissues immediately, molt to adults, and begin to feed and reproduce in resin canals (16). During winter and early spring, the dispersal third-stage juveniles, defined as the resting stage which can survive unfavorable conditions, congregate around the pupal chambers of the pine sawyer. In late spring, coinciding with the time of pupation, aggregated nematodes molt to dauer juveniles. Immediately after emerging, adult beetles become contaminated with dauer juveniles. In a severely damaged pine forest, an average of 15,000 dauer juveniles were recovered from a single insect (18). Outlying infection resulting from the introduction of nematode-infected pine logs is the most common pattern of spread of this disease into uninfested areas.

Control of pine wilt disease in Japan consists of three parts: 1) control of the Japanese pine sawyer population by aerial spraying as well as ground sprays, 2) control of PWN population, and 3) breeding pines resistant to PWN.

The low volume aerial spraying of insecticide, reducing the 60 liters of conventional spray by 8 liters per ha, and the technique of gun nozzle spray, a direction controlled spraying nozzle installed in a helicopter, appear very effective in controlling pine sawyers and also in minimizing adverse environmental effects (9). Two promising nematicides, mesulfenphos and morantel-tartalic acid, are now available for direct control of nematode populations. The project of breeding pines resistant to PWN, started in 1978, involves 17 organizations in grafting trees and PWN inoculation tests (9,17). The $F_1$ of an interspecific hybrid between *P. thunbergii* and *P. massoniana* showed high resistance to PWN (17).

Investigation of PWN has disclosed a magnificent symbiosis between nematodes and beetles with pine trees as the common host. Most pine species in the United States, where PWN is considered to originate, are highly tolerant to this nematode. This suggests the possible introduction of PWN from outside, because an introduced pest organism often shows strong pathogenicity to new hosts after acquiring an effective association with the native vector.

## Rice Root Nematodes

High population levels of the rice root nematodes (RRN), *Hirschmanniella imamuri* and *H. oryzae,* have always been found in most rice paddies throughout Japan. Although increased rice production is economically important in Asian countries, relatively little work has been done on the nematode pests of rice (7), except for the white tip nematode, *Aphelenchoides besseyi.*

Since 1968 the bionomics of two species of RRN have been compared by standardized methods in 15 prefectures scattered throughout Japan. Studies especially concentrated on distribution of the two species, seasonal prevalence, and effects on rice growth. Estimation of the nematode population levels in rice roots was made by the Baermann-funnel technique for 48 hours at 20–25 C, using arbitrarily sampled 5-g portions of fresh roots cut into 1-cm-long pieces.

Virtually all rice paddies in Japan are infected with one or both species of *Hirschmanniella,* except for a few paddies in Hokkaido where no RRN have been detected. Of 221 rice paddies examined, *H. oryzae* occurred alone in only 10%, *H. imamuri* occurred alone in 14%, and 72% were inhabited by both species simultaneously.

In transplanted rice roots, nematode populations gradually increase in June and reach a peak between August and September in the north and between August and November in the south. Although nematode populations in rice roots varied considerably among paddies, seasons, soil conditions, or cultural practices, the average number of nematodes per gram of root in the species-differential study conducted from September to November throughout Japan was 56.2 (n = 182) *H. oryzae* and 59.3 (n = 190) *H. imamuri.*

Even after the rice harvest, a comparatively high nematode population was always recovered from rice root stubble. Close examination revealed that from October to February a large portion of the *H. oryzae* population was in the adult stage with fewer juveniles observed, whereas *H. imamuri* was mainly in the third or fourth juvenile stage and only few adults were detected from stubble roots. Since a comparatively high adult ratio of *H. imamuri* has been observed in rice roots at transplanting time, the juveniles of *H. imamuri* that have hibernated in old rice stubble roots apparently continue to mature and emerge into the paddy soil by the time of rice transplanting. According to Kuwahara and Iyatomi (15), there is one generation a year of *H. imamuri* and two generations of *H. oryzae.*

A high population level with a high adult ratio (68.5%) of *H. imamuri* has also been observed at transplanting time in the roots of barnyard grass (*Panicum crus-galli* var. *frumentaceum*), a common rice paddy weed in Japan (4). Paddy weeds as well as rice stubble roots may serve as reservoirs for further nematode infection of rice.

The effect of different nematode population levels on growth of rice plants has been studied in many locations in Japan by means of nematode inoculation or nematicide treatment. In small tests, rice plants inoculated with a high population of RRN exhibited early growth suppression resulting in decreases in the number of tillers and in yield. However, many studies conducted in rice paddies have failed to demonstrate high correlations between nematode population levels and yield reductions.

Nematicide trials were conducted with D-D mixtures, 30% EDB, or DBCP (80% E.C.) at appropriate application rates for paddies. D-D treatment resulted in 10–38% yield increases in well-drained paddies, but less in moderately and poorly drained paddies. DBCP treatment increased the average yield by about 10%, but EDB induced plant abnormalities such as short culms, unfolded leaves dense to dark green in color, or exceptionally prolific tillering. Experiments indicated that soil treatment should be accompanied by a reduction in the amount of nitrogenous fertilizer by at least 20–40% (50–60% in heavy clay soil); otherwise the rice plant will grow too rapidly, resulting in heavy fungus infection and lodging.

The actual reduction in rice yield caused by RRN is still controversial. One reason is that the environmental conditions under which a rice plant grows are unique, and the conditions of experiments vary considerably. The fact that virtually every rice paddy in Japan is infested with RRN suggested that the rice varieties which now grow the best in paddies are those relatively resistant to this nematode, because they have been developed for hundreds of years in rice paddies inhabited by the nematodes. More data on the role of these nematodes in rice culture are needed.

## Literature Cited


1. Aota, T., A. Tanii, and J. Akai. 1981. Influence of plant parasitic nematodes on the incidence of brown stem rot of azuki bean. Annals of the Phytopathological Society of Japan 47:99–100 (Abstr.). (In Japanese.)

2. Dropkin, V. H., and A. S. Foudin. 1979. Report of the occurrence of *Bursaphelenchus lignicolus*-induced pine wilt disease in Missouri. Plant Disease Reporter 63:904–905.

3. Fukuzawa, A., A. Furusaki, M. Ikura, and T. Masamune. 1985. Glycinoeclepin A, a natural hatching stimulus for the soybean cyst nematode. Journal of the Chemical Society, Chemical Communications 4:222–224.

4. Goto, M. 1969. Ecological studies on *Hirschmanniella imamuri* S. A. Sher in Shonai-district, Japan. Yamagata Norin Gakkaiho 26:43–51. (In Japanese.)

5. Hori, S. 1915. Phytopathological notes, 5. Soil sickness of soybean caused by a nematode. Byotyugai Zasshi 2:927–930. (In Japanese.)

6. Ichinohe, M. 1952. On the soybean nematode, *Heterodera glycines* n. sp., from Japan. Oyo Dobutsugaku Zasshi 17:1–4.

7. Ichinohe, M. 1972. Nematode disease of rice. Pp. 127–143 *in* J. M. Webster, ed. Economic nematology. New York: Academic Press.

8. Ichinohe, M., T. Nishizawa, and K. Okamoto. 1980. Determination of pathotype of potato cyst nematode. Kenkyu Seika Series No. 127:21–34. Tokyo: Ministry of Agriculture, Forestry, and Fisheries. (In Japanese.)

9. Ikeda, T. 1984. Integrated pest management of Japanese pine wilt disease. European Journal of Forest Pathology 14:398–414.

10. Inagaki, H. 1979. Race status of five Japanese populations of *Heterodera glycines*. Japanese Journal of Nematology 9:1–4.

11. Inagaki, H., Y. Momota, and K. Shimizu. 1981. Race of the soybean cyst nematode in Saitama, Ibaraki, Gunma, and Chiba. Proceedings of the Kanto-Tosan Plant Protection Society 28:130. (In Japanese.)

12. Inagaki, H. 1984. Studies on the ecology and control of the potato cyst nematode, *Globodera rostochiensis*. Research Bulletin No. 139, Hokkaido National Agricultural Experiment Station, Sapporo, Japan. Pp. 73–144.

13. Ishikawa, M. 1958. Nematode resistance of soybean endemic variety 'Geden-shirazu' and selection of promising strain. Nogyo Gijutsu 13:461–463. (In Japanese.)

14. Kiyohara, T., and Y. Tokushige. 1971. Inoculation experiments of a nematode, *Bursaphelenchus* sp., onto pine trees. Journal of Japanese Forestry Society 53:210–218. (In Japanese; English summary.)

15. Kuwahara, M., and K. Iyatomi. 1970. Studies on the bionomics of rice-root nematode, *Hirschmanniella imamuri* Sher and *Hirschmanniella oryzae* (Soltwedel) Luc & Goodey, with special reference to its mode of life and population dynamics. Japanese Journal of Applied Entomology and Zoology 14:117–121. (In Japanese; English summary.)

16. Mamiya, Y. 1983. Pathology of the pine wilt disease caused by *Bursaphelenchus xylophilus*. Annual Review of Phytopathology 21:201–220.

17. Mamiya, Y. 1984. Perspectives of pine wilt disease in Japan. Pp. 6–25 *in* V. H. Dropkin, ed. Proceedings of the United States-Japan seminar, The resistance mechanisms of pines against pine wilt disease, University of Missouri, Columbia.

18. Mamiya, Y. 1984. The pine wood nematode. Pp. 589–626 *in* W. R. Nickle, ed. Plant and insect nematodes. New York: Marcel Dekker.

19. Masamune, T., M. Anetai, M. Takasugi, and N. Katsui. 1982. Isolation of a natural hatching stimulus, glycinoeclepin A, for the soybean cyst nematode. Nature 297:495–496.

20. Minagawa, N., Y. Ohshima, and K. Nakasono. 1986. Annotated bibliography of nematology in Japan: 1879–1984. Miscellaneous Bulletin No. 67, Kyushu National Agricultural Experiment Station, Chikugo, Fukuoka, Japan. (In Japanese.)

21. Momota, Y., and H. Inagaki. 1983. Races of

the soybean cyst nematode in Iwate, Miyagi, Tochigi, Yamanashi, Hyogo and Kumamoto. Proceedings of the Kanto-Tosan Plant Protection Society 30:192. (In Japanese.)

22. Mori, M. 1986. New potato cultivar for industry 'Toyoakari'. Nogyo Gijutsu 41:554–555. (In Japanese.)

23. Negishi, H., and K. Kobayashi. 1984. Effect of soybean cyst nematode on the incidence of brown stem rot of adzuki bean. Annals of the Phytopathological Society of Japan 50:500–506. (In Japanese; English summary.)

24. Oda, K. 1967. The applicability of measurement of the oleoresin yield in determining the susceptibility of pine trees to beetle infestation. Forest Pest News (Tokyo) 16:263–266. (In Japanese.)

25. Saibai Dainibu Sakumotsu Daisankenkyushitsu. 1970. Genetic report of soybean new cultivars bred with gamma irradiation—'Raiden' and 'Raiko'. Research Bulletin No. 40, Tohoku National Agricultural Experiment Station, Kuriyagawa, Iwata, Japan. Pp. 65–104. (In Japanese.)

26. Shimizu, K., H. Inoue, K. Sunada, K. Honma, and Y. Mitsui. 1981. Races of the soybean cyst nematode in Tokachi district, Hokkaido. Annual Report of Plant Protection of North Japan 32:67–68. (In Japanese.)

27. Shimizu, K., Y. Mitsui, and M. Ichinohe. 1984. Races of the soybean cyst nematode in Tokachi Province, Hokkaido, Japan. Proceedings of the First International Congress of Nematology. P. 84 (Abstr.).

28. Stone, A. R. 1973. *Heterodera pallida* n. sp. (Nematoda: Heteroderidae), a second species of potato cyst nematode. Nematologica 18:591–606.

29. Sunada, K., K. Gotoh, M. Saito, and S. Sakai. 1966. Soybean new cultivar 'Hohrai' and 'Toyosuzu'. Hokuno 33(11):16–28. (In Japanese.)

30. Sunada, K., S. Sakai, K. Gotoh, T. Sanbuichi, T. Tsuchiya, and M. Kamiya. 1981. A new soybean variety 'Suzu-hime'. Bulletin No. 45, Hokkaido Prefectural Agricultural Experiment Stations, Naganuma, Hokkaido, Japan. Pp. 89–100. (In Japanese; English summary.)

31. Tsutsumi, M., and K. Sakurai. 1966. Influence of root diffusates of several host and non-host plants on the hatching of the soybean cyst nematode, *Heterodera glycines* Ichinohe, 1952. Japanese Journal of Applied Entomology and Zoology 10:129–137. (In Japanese; English summary.)

32. Yamada, E. 1987. Studies on ecology and control of the potato cyst nematode, *Globodera rostochiensis* (Wollenweber, 1923) Behrens, 1975. Report No. 16, Hokkaido Prefectural Agricultural Experiment Station, Naganuma, Hokkaido, Japan. (In Japanese; English summary.)

33. Yano, M. 1913. Investigation of the causes of pine mortality in Nagasaki Prefecture. Sanrin Koho 4(Suppl.):1–14. (In Japanese.)