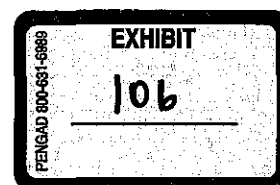
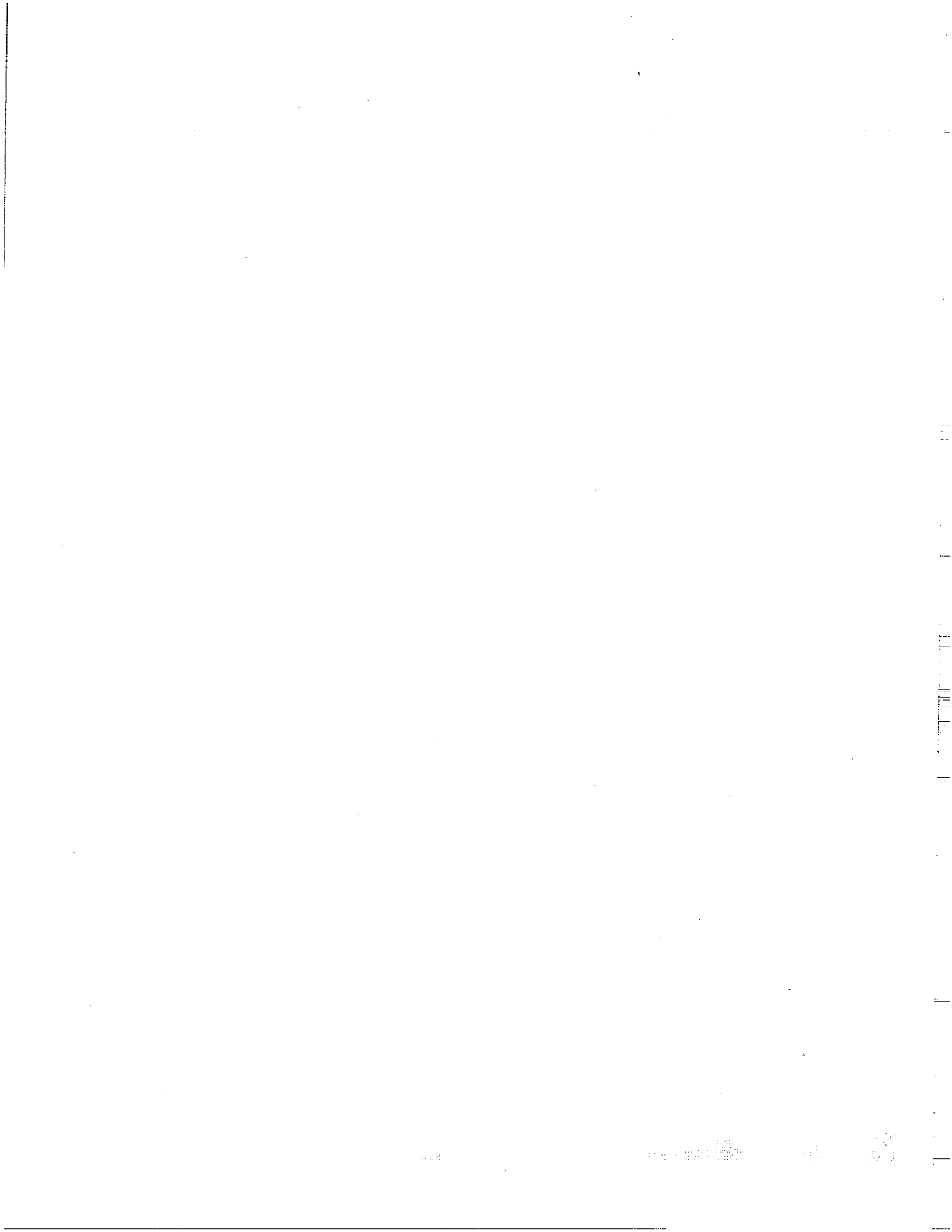


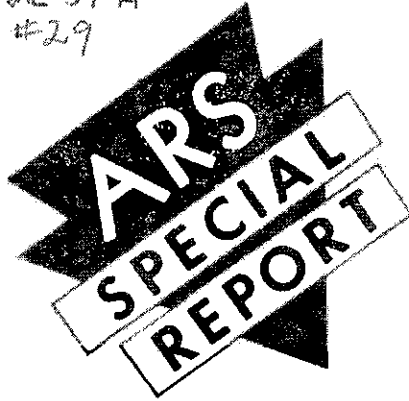
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Soybean Cyst Nematode

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Agricultural Research Service
UNITED STATES DEPARTMENT OF AGRICULTURE

SUMMARY

The soybean cyst nematode, which, depending on the degree of infestation, can cause damage to soybean fields ranging from no measurable amount to complete destruction of the crop, has been found on an area of about 1,400 acres in southeastern North Carolina.

Surveys undertaken since its discovery in August 1954 and warnings sent to soybean-producing States have failed to reveal its presence in any other area.

Research programs, begun in 1955, are continuing. These efforts are directed toward finding vulnerable points in its life history, seeking out soil fumigants, crop rotations, and resistant strains of soybeans that will assist in control or eradication of the pest. It also affects annual lespedeza, common vetch, and snap beans.

Information in this report was furnished by the North Carolina Agricultural Experiment Station, the Field Crops Research Branch, the Horticultural Corps Research Branch, and the Plant Pest Control Branch,
Agricultural Research Service

SOYBEAN CYST NEMATODE

THE SOYBEAN CYST NEMATODE, which is capable of causing total destruction of a crop of soybeans and which was previously known only in the Orient, has now been found on about 1,400 acres in southeastern North Carolina. It was first observed in the United States in August 1954.

Described by a Japanese scientist in 1952 as a new species, the soybean cyst nematode has as additional hosts in Japan the adzuki bean (a food bean in the Orient but grown only for experimental purposes in the United States), the Spanish runner bean (also seldom grown in this country), and the snap bean (which is the same species as the kidney bean, *Phaseolus vulgaris*). Preliminary host-range studies in North Carolina in 1955 indicate that it also parasitizes annual lespedeza and common vetch.

Soon after discovery of the pest, North Carolina authorities asked the Federal Government to assist in making a survey to determine the extent of the infestation. Early in 1955 the State entomologist issued a notice of the presence of the pest in North Carolina to all State plant regulatory officials. A soybean cyst nematode control program was organized in March 1955; cooperating agencies include the North Carolina State Department of Agriculture, the North Carolina State College of Agriculture, and appropriate branches of the Agricultural Research Service, U. S. Department of Agriculture. Preliminary research on the nematode under North Carolina conditions was started by the State Experiment Station and the Field Crops Research Branch in the spring of 1955. The Plant Pest Control Branch, Agricultural Research Service, in July issued an alert to the States in which soybeans are produced commercially, urging them to be on the lookout for unexplained losses.

Surveys of the infested area have established the presence of the pest in fields totaling 1,400 acres on 74 premises. Except for a fairly restricted area in New Hanover and Pender counties, north of Wilmington, the nematode has not been found elsewhere in North Carolina or in the neighboring States of South Carolina and Virginia.

In March 1956 North Carolina imposed a quarantine on the movement of soil, plant parts, machines, and other materials that might spread the infestation.

DISCOVERY AND SURVEY

IN AUGUST 1954, a farmer in the Castle Hayne area, New Hanover County, asked a plant pathologist of the North Carolina Agricultural Experiment Station what could be causing the yellowing and stunting of his soybeans. Samples of plants and soil were sent to an Experiment Station nematologist at Raleigh, who tentatively diagnosed the cause as the soybean cyst nematode. This organism is very difficult to distinguish from the clover cyst nematode. Several thousand cysts (egg-filled bodies of females) and numerous male nematodes were found per pint of soil examined. Samples were sent to a nematologist of the U. S. Department of Agriculture, at Beltsville, Md., who confirmed the diagnosis.

Although proof is lacking, it has been suggested that the nematode slipped into the country shortly before the start of World War II in a shipment of Easter lilies from Japan. The soybean was first mentioned in American literature in 1804; the Nation is fortunate that this soybean pest waited nearly 150 years to follow it. The nematode does not attack bulb plants, but cysts may have adhered to bulbs while in transit. In the intervening years the nematode slowly spread through the soil, moved mostly by cultivating and harvesting equipment, in an area where soybeans are used as a cover crop for bulbs or in rotation with bulb or vegetable crops.

First published announcement of the presence of the pest in the Western Hemisphere appeared in the Plant Disease Reporter, January 15, 1955.

Immediate steps were taken: Japanese scientific papers (with summaries in English) were studied to learn how the pest behaved before it left home. Research projects were set up to study its life history and habits in the new location. Surveys were initiated to find how far the nematode had spread--always a prerequisite to regulatory and control operations.

Infestation was, and is, heavy in the Castle Hayne area. In most of the fields it was relatively easy to find live cysts attached to the roots of soybean plants or live males in the soil. During 1955 the pest was found on 770 acres in 50 properties. In some fields only dead cysts were found, which are so similar to those of the clover cyst nematode that no trustworthy way had yet been found to distinguish between them. Dead cysts were found on 157 acres in 20 properties.

Means of spread were important to the survey. Samples were taken of the soil and dust clinging to two combines that were used to harvest infested fields. Per pound of soil taken from these machines, an average of 4,156 cysts were recovered, of which 16.5 percent contained eggs with viable larvae. Inspectors then began to check custom combining machinery that was used outside the immediate area of Castle Hayne. This led to 91 infested acres in 3 properties north of the county line in Pender County (which in 1954 harvested 3,450 acres of soybeans). Other means of spread include wind (by which the pest was moved 200 yards in one instance tested), water, animals, farm workers and machinery, local traffic, or any means capable of spreading small amounts of infested soils.

Considering the number of cysts found in soil on combines, inspectors made a closer study of combining practices: The beans are threshed and put in burlap bags on the combine, and are tied shut and pushed off the combine onto the ground for later transport by truck. A load of bagged-up beans was taken from Castle Hayne to Goldsboro, about 80 miles north, on a truck. Inspectors swept the body of the truck after it was unloaded at an oil mill. Among half a gallon of soybeans and 3 or 4 ounces of soil they found one living cyst. A cyst contains 50 to 600 eggs, averaging about 200.

Up to February 17, 1956, some 8,500 soil samples had been taken on 6,182 acres in 12 North Carolina counties. In 1954 these 12 counties (including New Hanover and Pender) harvested 77,360 acres of soybeans. The range was from 160 acres in Henderson County to 34,200 in Beaufort County, both of which grow some soybeans in rotation with bulbs. Outside of the two infested counties, New Hanover and Pender, the nematode was not found elsewhere in North Carolina.

Special effort went into a survey of 7 southeastern Virginia counties where bulbs and soybeans are grown in the same fields. Eighty-four soil

samples were collected on 21 properties having an area of 232 acres. The samples were processed at the Virginia Agricultural Experiment Station. No soybean cyst nematodes were found.

Four counties of northeastern South Carolina were surveyed and a determined search was made for bulb-soybean plantations. No such associations were found, and survey and observation of many soybean fields revealed no symptoms of infestation by this nematode.

Inasmuch as nearly 20 million acres of soybeans are grown in the United States, the Plant Pest Control Branch sent a letter to the following States to alert them to the problem: Alabama, Arkansas, Delaware, Florida, Georgia, Illinois, Indiana, Iowa, Kansas, Kentucky, Louisiana, Maryland, Michigan, Minnesota, Mississippi, Missouri, Nebraska, New Jersey, New York, Ohio, Oklahoma, Pennsylvania, South Carolina, South Dakota, Tennessee, Texas, Virginia, West Virginia, and Wisconsin. Table 1 shows soybean acreages in the States of principal production. The value of the 1954 U. S. soybean crop for all purposes was estimated at about \$1 billion.

This alert has been in the hands of interested officials of the soybean-growing States since July 1, 1955. None has so far reported the presence of the pest or symptoms that could not otherwise be explained.

THE NEMATODE

THE SOYBEAN CYST NEMATODE (*Heterodera glycines* Ichinohe) is one of numerous kinds of tiny, almost transparent eelworms that infest the soil, plants, and animals the world over. The root meaning of the word nematode is "threadlike." The name is very descriptive of these little animals before the cyst-forming stage as revealed by a low-power microscope. The male of the species is about one-twentieth of an inch long--the width of a period in ordinary print--and about 1 one-thousandth of an inch in thickness. The female, when distended into an egg-bearing cyst, is slightly shorter but much thicker so that it can easily be seen by the unaided eye.

For many years it was considered impossible that these tiny worms could damage plants by feeding on them, even when they appeared in myriads in the soil. But such judgment reckoned without the peculiar adaptation of this and thousands of other types of nematodes. The worm has six lips and inside its mouth is a short sharp stylet or spear measuring about 1 one-thousandth of an inch. The stylet is hollow, like a hypodermic needle, and it is incredibly sharp and durable. Piercing a hole in a soybean rootlet with this amazing mouth-dagger, the nematode wriggles inside the root. Moving along until it reaches a set of vascular bundles, the nutrient pipelines of the plant, it fixes itself there and begins to tap this abundant food supply. After that, life is easy. All the nematode has to do is to suck in the juices through its hypodermic tongue and devote itself to growth and reproduction.

But life becomes difficult for the plant. In order to feed, the nematode injects chemicals that predigest the plant food for its own use, but these substances interfere with the plant's metabolism and cause it to become dwarfed and yellow. Millions of nematodes working on thousands of plants in a field cause a disease of soybeans first called "yellow dwarf" disease in Japan, Korea, and Manchuria, where the soybean cyst nematode has its only known habitat outside of North Carolina. The nematode-caused

Table 1.--Soybean acreages in States of principal production¹

State	Grown for all purposes		Grown for beans	
	Average 1944-53	1955	Average 1944-53	1955
	1,000 acres			
Illinois.....	3,804	4,642	3,611	4,530
Minnesota.....	925	2,371	870	2,335
Iowa.....	1,735	2,248	1,685	2,223
Indiana.....	1,704	2,202	1,557	2,114
Missouri.....	1,154	1,987	1,070	1,930
Ohio.....	1,077	1,264	1,015	1,245
Arkansas.....	515	1,030	431	933
Mississippi.....	385	752	222	544
North Carolina.....	390	423	255	285
Kansas.....	361	348	322	300
Tennessee.....	245	287	130	185
South Dakota.....	48	272	46	263
Nebraska.....	46	252	44	245
Virginia.....	182	237	122	172
Kentucky.....	194	206	103	130
South Carolina.....	78	183	52	150
Michigan.....	112	170	96	165
Alabama.....	179	157	59	106
Louisiana.....	107	152	31	56
Maryland.....	87	141	58	116
Wisconsin.....	73	91	37	71
Georgia.....	72	89	20	35
Delaware.....	66	80	53	71
North Dakota.....	19	80	17	79
Oklahoma.....	50	48	29	30
Pennsylvania.....	58	46	24	21
New Jersey.....	36	41	17	23
Florida.....	² 12	40	² 9	34
New York.....	9	8	6	6
West Virginia.....	18	7
Texas.....	7	6
UNITED STATES.....	13,740	19,860	11,987	18,397

¹ Data reported in Crop Production, 1955, an annual summary issued by the Agricultural Marketing Service.

² Short-time average.

disease has been known since 1915, when Japanese scientists attributed it to a strain of the sugar-beet nematode, which it greatly resembles. During the 1940's it was considered rather to be a variant of the pea cyst nematode, which it also resembles. Finally, in 1952, it was identified as a separate species.

The soybean cyst nematode goes through four stages as a worm. It molts once while still in the egg and emerges as a second-stage larva. This is the so-called infectious stage, when the nematodes, still young and slender, penetrate into plants. They molt twice more within the host and become adults. In the first two stages they are sexless; after the third molt sexual differentiation begins, and after the fourth molt males and females are easily distinguished. The female remains within the plant, feeding and enlarging until her swollen body causes the rootlet to crack open and her body protrudes, remaining attached by the neck. This may be an adaptation for mating with the males, which emerge from the plant in the adult stage and are free-living in the soil, and for the later release of young larvae. After mating, some eggs are deposited in a gelatinous mass outside the body of the female, but some 50 to 200 eggs are retained within the distended lemon-shaped body. The female body is at first white, changing to yellow, and, after death, it turns to an olive brown. This is the tough-skinned cyst characteristic of the cyst nematodes. The cyst, which is highly resistant to decay, protects the eggs until they are ready to hatch. This resistance to decay and other adverse conditions is of great significance in the survival and dissemination of cyst nematodes.

Second-stage larvae develop within the eggs in the cyst. They may hatch immediately, though many may remain unhatched for an indeterminate period. Research has yet to demonstrate how long they remain quiescent and what finally causes them to hatch. Once hatched, they are apparently able to "scent" and move toward the roots of host plants on which they must feed to produce the next generation.

In the latitude and climate of Japan, this nematode can produce three generations in a growing season. Greenhouse tests made at the North Carolina Agricultural Experiment Station indicate that three to four generations could develop during the growing season there, which starts in mid-May and ends in late September.

Host plants were studied in Japan. Pot tests were run on 32 different plants, including legumes, grains, and vegetables. Full-sized egg-bearing cysts developed on the soybean and the adzuki bean. On the snap bean the young females in the roots did not grow to normal size, the cysts were small, and the number of eggs was reduced. The nematodes attacked the Spanish runner bean but development was not complete.

Forty different plant varieties were studied in greenhouse host tests conducted at the North Carolina Station in 1955. Here again the soybean was the favored host. But two new hosts were added, annual lespedeza and common vetch, in both of which the pest reproduced as readily as in soybeans.

Legumes found not susceptible, in North Carolina greenhouse tests, include cowpea, velvet bean, lupine, perennial lespedeza, crotalaria, garden pea, red and ladino clover, alfalfa, and peanut.

Damage done by the soybean cyst nematode to a field of soybeans has been stated in different ways by scientists of the Eastern and Western



Hemispheres. In North Carolina, where the uneven yellow patches have appeared in the fields for anyone to see, the damage has varied from slight in the early stages of infestation to such destruction that no effort was made to harvest the crop.

In Japan nematode-infested plants were weighed, measured, and their pods were counted in comparison with uninfested plants. Height of infested plants ranged from 9 to 16 inches, compared with 24 inches for healthy plants. Nematode-ridden plants ranged in weight from one-tenth to seven-tenths of an ounce, whereas healthy plants weighed 3 ounces. The number of pods per infested plant ranged from an average of 2.4 in a badly infested area to 10.7 in a less heavily infested area; healthy plants had 38 pods.

STEPS BEING TAKEN

Research

SCIENTISTS of the North Carolina Agricultural Experiment Station, working in 1955 with the North Carolina State Department of Agriculture and the Agricultural Research Service of the U. S. Department of Agriculture, in one year of preliminary research assembled an impressive group of findings.

Identification of the soybean cyst nematode as distinguished from the clover cyst nematode, which is widely distributed in several soybean-producing States, has been worked out in a series of meticulous morphological studies by a worker at the experiment station. Until this method was evolved it was necessary to identify the soybean cyst nematode by the time-consuming method of planting cysts or larvae with red or white clover and soybeans to see which they would attack. The problem was further complicated by the fact that the two nematodes have other hosts in common--the snap bean, annual lespedeza, and common vetch.

No readily discernible difference was found in the size, shape, or pattern of markings of eggs or cysts. But, once the second-stage larva leaves the egg and is examined microscopically, many differences are evident. Measuring 150 larvae of each species, it was found that several anatomical measurements are distinctly different. The measurements are consistent, and the averages of the two sets of measurements never overlap. Once recognized, these differences can be detected rapidly by an experienced worker, greatly shortening the time required to identify a soybean nematode. Another difference becomes evident in the adult stage--males of the soybean nematode are very common, whereas males of the clover nematode are seldom found.

Resistance studies were carried out in preliminary greenhouse tests by exposing 309 soybean varieties to natural infestations of the soybean cyst nematode. Approximately one-half of the varieties proved susceptible in these tests, including some that had shown resistance in Japan. Those that showed no infestation will be tested again. Because the growing season of 1955 was generally unfavorable, scientists working on this project felt that some varieties may have escaped infestation because of weather conditions.

Soil fumigation was tried in field tests with two types of commercial nematocides--one a mixture of dichloropropane and dichloropropene (DD mixture) and the other containing 85 percent ethylene dibromide (EDB). Both were applied at the recommended rate for other nematodes, double

that, and quadruple that. The DD mixture was used at the recommended rate of 20 gallons per acre and at 40 and 80 gallons per acre. EDB was applied at the recommended rate of 4.5 gallons per acre and at 9 and 18 gallons per acre.

At recommended and double rates of application, with both nematocides, there was increased plant growth and yield over unfumigated plots. At quadruple rates there was stunting and evidence of toxic effects on the plants. Living cysts were found in all of the plots after fumigation, indicating failure of these materials at the rates applied or methods used to eradicate the soybean cyst nematode. But, these tests must be regarded as preliminary and will be repeated.

Eradication treatments were tested on daffodil bulbs and gladiolus corms, a great many of which are grown in and shipped from the infested area. While daffodils and gladiolus are not susceptible to the nematode, bulbs or corms of these plants might easily become contaminated with cysts and serve as carriers in introduction of the pest. The tests began with accepted bulb-dipping treatments that are in current use for fungus rots, including hot water with 1 part of 40-percent formalin to 200 parts of water, and 85-percent sodium trichlorophenate. The same treatment as that given the bulbs was applied to screened soil containing nematode cysts, enclosed in cloth bags. After treatment the cysts were placed in pots containing nematode-free soil, and soybeans were planted. In 6 or 8 weeks the soil and roots of the soybeans were examined for the presence of new cysts, and the efficacy of the treatment was determined.

Using hot water, it was found that when bulbs are steeped or soaked in water at 120° F. for 30 to 60 minutes it kills all soybean cyst nematodes. A similar result was obtained when bulbs were soaked 15 to 30 minutes in water at 130° F.

Using 40-percent formalin in a 0.5-percent solution by volume at 111° F., all nematodes were killed after soaking 3 or 4 hours. This solution was not fatal to the pests when they were immersed for an equal length of time at a temperature of 77° to 56° F.

Using 85-percent sodium trichlorophenate, which is widely employed to treat bulbs and corms for fusarium rot, at the rate of 2 pounds to 100 gallons of water at prevailing temperatures, all soybean nematodes were killed after soaking for 1 hour. When soaked for 15 minutes, the same result was obtained in solutions of 3 and 6 pounds to 100 gallons of water. Scientists say that the sodium trichlorophenate treatment can be applied very cheaply. For materials alone, 2 tons of bulbs can be dipped for less than \$1. This, of course, does not represent the total cost, which includes investment in dipping equipment and the charge for labor.

As a control, bulbs and small bags of nematode cysts collected for the purpose were run through water at the prevailing temperature, and through water containing a detergent. Neither had any effect on the viability of the organisms. Two other chemicals, 97.5-percent phenylmercury acetate and 5-percent ethylmercury phosphate, failed to kill the soybean cyst nematode.

Air-drying experiments were tried on nematode-infested soil in order to measure the mortality of nematodes in the normal practice of air-drying bulbs. In one experiment, two soil samples weighing 3.2 grams (0.1 oz.) were taken from soil that had been removed from an infested field. Hatched larvae were counted in one sample, which had not been air-

dried, and 52,314 larvae were found. The other sample was dried at room temperature for 42 days and hatched larvae were counted. Forty-two were found. Thus, assuming that both samples of equal weight had originally contained an equal number of nematodes, air-drying had reduced the number 1,245-fold.

In another experiment, soil samples of similar size were taken from pots in which diseased soybeans had been grown in the greenhouse. From the sample not air-dried, 44,158 larvae were counted. The other sample was air-dried for 72 days and hatched larvae were counted. They numbered 70--a 630-fold reduction.

Quarantine

Following a hearing of interested parties at Raleigh, March 29, 1956, the North Carolina State Board of Agriculture the same day issued a quarantine covering infested areas and premises. Regulated articles may be moved out of quarantined areas only when accompanied by a permit, which is issued when the following conditions, quoted from the quarantine regulations, are met:

- (1) Living soybean cyst nematode in any stage of development may not be moved or transported except for scientific purposes as authorized by the State entomologist, unless a certificate or permit has been issued therefor.
- (2) Soil as such or soil attached to plants or articles may not be moved from the quarantined area to any point outside thereof unless such soil, under the supervision of the inspector, has been sterilized, fumigated or otherwise treated so as to kill all nematodes.
- (3) True bulbs, corms and rhizomes may not be moved out of the quarantined area until at least a 60-day drying period, after digging, has elapsed and they have been thoroughly cleaned of soil.
- (4) Root crops and tubers may not be moved out of the quarantined area unless all soil has been removed by washing.
- (5) Farm tools, implements, and other construction and maintenance equipment may not be moved out of the quarantined area, unless all soil has been removed by washing.
- (6) Crates, boxes, burlap bags, or other farm product containers used for harvesting in infested fields may not be used for marketing.
- (7) Except as noted above, portions of plants without roots attached (such as cut flowers) may be moved from the quarantined area without certification.
- (8) Compliance with subsections (2), (3), (4), and (5) of this section shall not be necessary when such treatment or washing is found by an inspector to be unnecessary because the particular portion of the quarantined area involved is found by the inspector not to be infested or not to be so located or used as to be exposed to infestation.

THE OUTLOOK

Economic Importance of Host Plants

THE SOYBEAN CYST NEMATODE has a limited number of hosts. However, these hosts occupy extensive acreages in many States.

Soybeans.--This is the principal host, and soybeans are a big crop in the United States--grown for all purposes on nearly 20 million acres in 31 States (table 1). They have a farm value of about \$1 billion.

Lespedeza.--According to the North Carolina host-range studies, the soybean cyst nematode attacks annual lespedeza quite as willingly as soybeans, and goes through the full cycle of penetration and reproduction in this host. Lespedeza is grown in 18 States for hay and seed, in addition to its role as pasture, green-manure, and cover crops. In 1954 a total of 3,702,000 acres of lespedeza was harvested for hay, of which about 10 percent is estimated to be of the perennial variety, and 580,500 acres of annual lespedeza for seed, making a total of 4,282,500 acres grown for these two purposes in Alabama, Arkansas, Delaware, Georgia, Illinois, Indiana, Kansas, Kentucky, Louisiana, Maryland, Mississippi, Missouri, North Carolina, Oklahoma, South Carolina, Tennessee, Virginia, and West Virginia. Largest producers of lespedeza hay are Tennessee, Kentucky, and North Carolina; States producing most lespedeza seed are Missouri, North Carolina, and Kentucky. Farm value of the seed for the country as a whole was placed in 1954 at \$15.8 million.

Common Vetch.--As tested in North Carolina, this plant is attacked as readily by the soybean cyst nematode as are soybeans and lespedeza. In areas where it is used as a winter cover crop, common vetch is of considerable importance to farmers. Because it grows as a weed in some places, along roadsides and fencerows, it might serve as an alternate host when other plants suitable to sustain the nematode are not available.

Snap Beans.--As shown by host tests in Japan and North Carolina, the soybean cyst nematode does not develop as well on this plant as on more favored hosts, growing fewer and smaller cysts. No information has been developed on the damage this nematode does to snap beans. Snap beans, which had a value of \$85.7 million in 1954, are grown commercially on 309,400 acres in 32 States: Alabama, Arkansas, California, Colorado, Connecticut, Delaware, Florida, Georgia, Illinois, Louisiana, Maine, Maryland, Massachusetts, Michigan, Mississippi, Missouri, New Hampshire, New Jersey, New York, North Carolina, Ohio, Oklahoma, Oregon, Pennsylvania, Rhode Island, South Carolina, Tennessee, Texas, Utah, Virginia, Washington, and Wisconsin. Farm value of the crop harvested for the fresh market in 1954 was \$43.2 million and the farm value of snap beans used for processing was \$42.5 million. States of heaviest production, in the order given, were Florida, New York, Wisconsin, Maryland, South Carolina, and North Carolina.

Research

Rotation.--The North Carolina Agricultural Experiment Station has leased a tract of nematode-infested land for 5 years, beginning in 1956, to carry out a series of crop rotations. Soybeans and other host plants will be left out of rotations for 1 to 5 years to determine whether and in what period of time the nematode can be starved out by this means.

Soil fumigation trials will be continued.

Resistance studies will be continued, with some 150 varieties of soybeans that did not become infested during the 1955 trials. Early research in this direction will be aimed at complete resistance to the nematode. Tolerance will be sought later, if resistance cannot be found.

In general, an attitude of optimism prevails, among State and Federal workers associated with this problem, that research will point the way to control of the pest.

