FRONTIERS
of PLANT SCIENCE

FALL 1973

THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION NEW HAVEN

New Caterpillar Pests

NOW IT'S THE RED-HUMPED OAKWORM

By Paul Gough

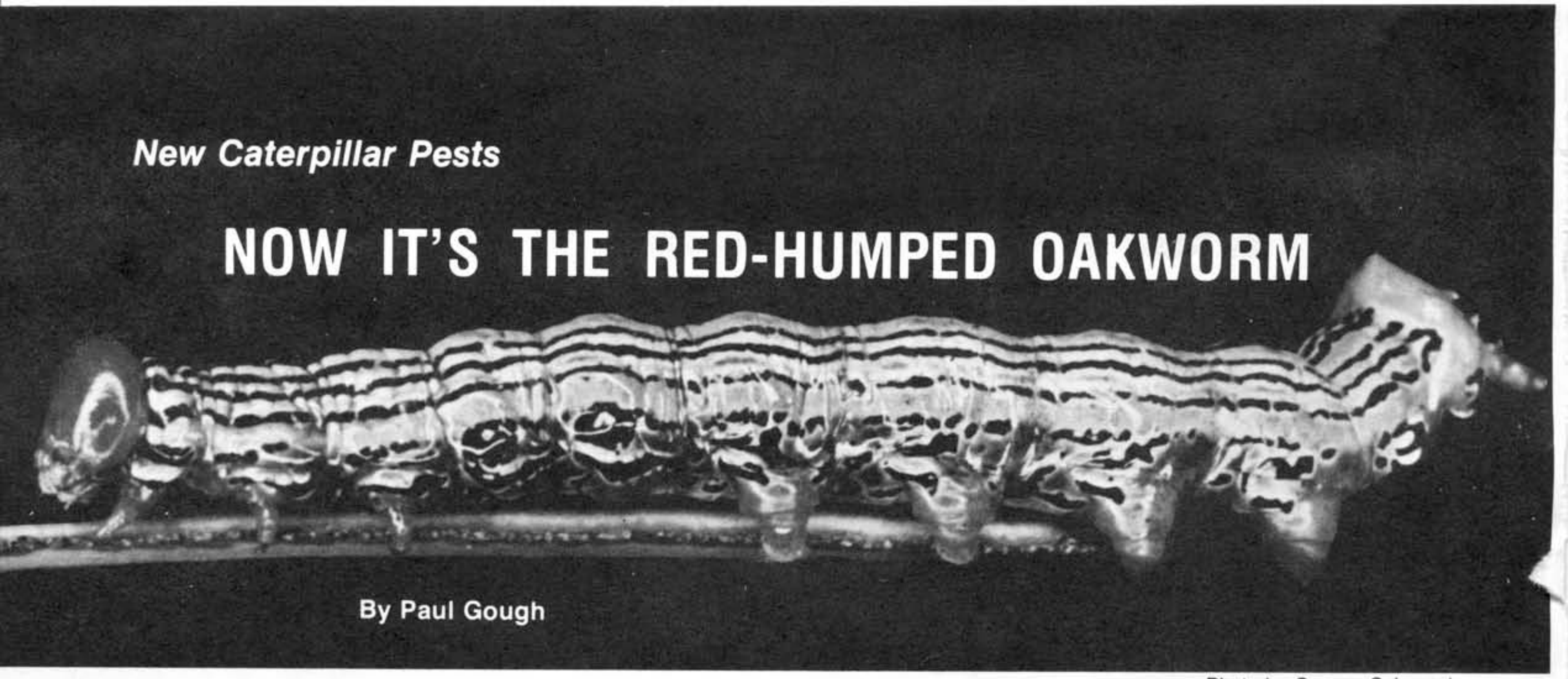


Photo by George Schuessler

WITH tree branches bare of leaves and winter fast approaching, scientists trying to find ways to control defoliating insects can relax, at least until next spring. Right?

Wrong!

As a matter of fact, Experiment Station entomologists have found themselves faced with several new insect pests this year in addition to their long-time early season adversary, the gypsy moth.

The relative newcomers in the lepidopterous rogue's gallery are the red-humped oakworm and the fall webworm. The webworm has been around in outbreak proportions before, but the oakworm is a new problem. Both prefer to dine on fall foliage.

These late season defoliators have created a unique problem since they rarely appear in sufficient numbers to attract much attention. And, to make matters worse, all of these insects are abundant in the northeastern section of the state.

Since the insects, depending on the particular species, are spending the winter as eggs or cocoons, awaiting the proper signals to awaken and become active again, it would appear there is little that one can do besides hope they'll go away and never come back.

Unfortunately, this is not likely to happen, so entomologists John Anderson and Harry Kaya are working in the Experiment Station laboratories in hopes of getting a head start for next year when the caterpillars make their almost inevitable reappearance.

The fall webworm is more of an aesthetic problem than a threat to trees, according to Dr. Anderson. Their webs are highly visible on trees at the edges of open fields, waterways and highways. They tend to attack trees like wild cherry which are not normally found in proximity to people, although they also attack ash and hickory which do grow in yards.

But the red-humped oakworm, which actually has orange humps, likes to feed on oaks, which can be found just as easily in a yard as in the forest. And since they forage for food in groups, they are extremely distasteful to have around, especially when they wander around houses looking for other trees to defoliate.

An aerial survey made in early October by Robert Moore of the Station staff, after the caterpillars stopped feeding, showed that more than 40,000 acres of forest in northeastern Connecticut had been defoliated by the red-humped oakworm.

This hardly compares with the half-million acres struck by the gypsy moth and elm spanworm the past few springs, but it establishes a record since the only previously recorded red-humped oakworm outbreak occurred in Bloomfield in 1940 when 10 acres or less were affected.

Drs. Anderson and Kaya have been making periodic trips to eastern Connecticut to collect caterpillars for further study ever since the problem sprang up with little warning in the late summer.

They have been particularly interested in dead or dying caterpillars because they are ever hopeful that they might find a germ or insect like the tiny wasps that decimated the elm spanworm population in 1972.

At this point, the search has not been too fruitful, but winter is long and there are many potential hazards the dormant oakworms must face before they can reproduce and continue their picnic in the woodlands.

While Dr. Kaya has found the bacterial insecticide *Bacillus thuringiensis* effective against these late season caterpillars, the scientists are searching for agents in nature which keep the caterpillars under control.

In their search for pathogens,

they have dissected dead caterpillars in assembly-line fashion in a basement room of the Jenkins Laboratory.

Dr. Anderson starts by placing a dead caterpillar on a sterile microscope slide where it is cut open with a flame sterilized scalpel. The slide is then passed to Kaya who examines it under the high-powered light microscope for evidence of pathogens. They switch places periodically.

In a number of cases, the cause of death is unknown, or at least not immediately visible upon examination.

But, Anderson and Kaya have already uncovered several causes of death for a significant number of caterpillars, including two insect-specific fungi. They have also found one unknown pathogen and a protozoan and several fly parasites.

They are most excited by the find of fungi. Already, these have been cultured on two different media. It is probable that these agents had never been isolated from the red-humped oakworm.

Attempts have been made to infect gypsy moth caterpillars and other oakworms with the fungi, but with little success.

The oakworms studied in the laboratory were nourished by oak leaves, but this food source was rapidly depleted by hungry caterpillars and by the annual fall foliage housecleaning. Fortunately for future studies, however, oakworms spend the winter in brown pupal cases.

There is a possibility that these pupae may be attacked by any number of parasites or predators or be affected by cold weather as they spend the long winter awaiting the opportunity to emerge as adult moths.

Drs. Anderson and Kaya are making frequent collections of oakworm pupae and intend to study them carefully. Any parasites will eventually emerge, while healthy pupae will become moths next July and be ready to lay eggs on the underside of leaves. Greg Piontek and Barbara Tracanna, entomology technicians, have been helping out with the laboratory phases of this project.

While these investigations will not solve the problems created by the red-humped oakworm, they will help prepare the scientists for the coming caterpillar season.

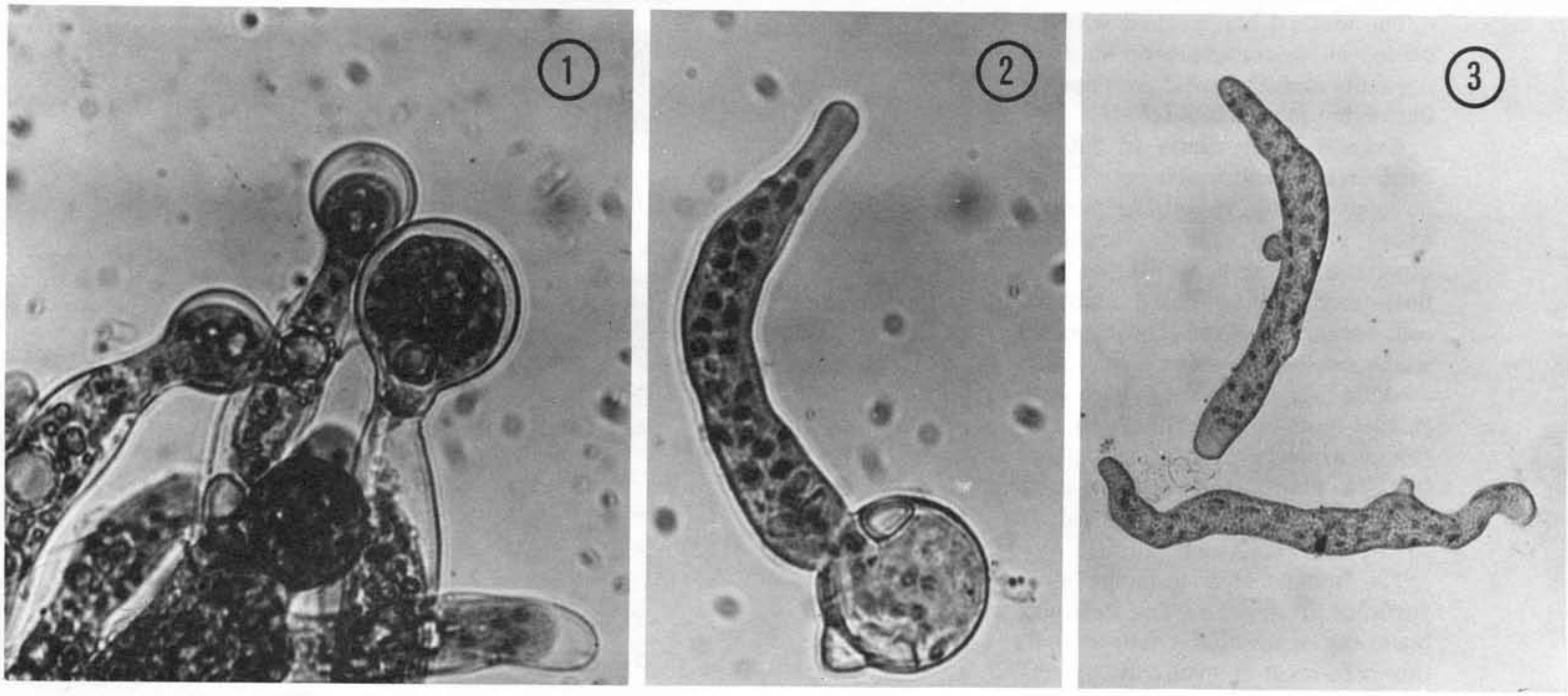
A better understanding of the natural mortality factors should help entomologists predict whether the insects will survive in sufficient numbers to be a nuisance again in 1974.

While they are watching the oakworm, Drs. Anderson and Kaya have also been studying another insect which is on the increase in the same general area.

These tussock moths, which are closely related to the gypsy moth, hibernate for the winter as tiny caterpillars, and are expected to continue their growth in the spring.

The entomologists intend to conduct the same kind of studies with the tussock moth, so while insects may take the winter off, an entomologist can hardly afford to miss a day in the fight against leaf-eating arthropods.

A fungus found in oakworm caterpillars. (1) Developing conidia at the tips are propelled away (2) Conidium producing hyphal body (3) Hyphal bodies. Cover Photo. Conidiopores in the process of developing conidia.



PLANT BREEDING WITHOUT SOWING SEED

By Peter R. Day

THE PAST 50 years have seen great increases in the yields of food crops, many resulting from plant breeding. During the past five years, however, the rate of increase has begun to fall, while the demand for food and its cost have continued to grow. It seems as though the old methods of breeding for increased yield are not working fast enough.

Station scientists are developing new plant breeding methods using cultured cells of crop plants that may produce new varieties more rapidly. A single petri dish of agar medium, which will accommodate tens of thousands of cells, could be equivalent to a field with an equal number of plants. The trick is to devise ways to select only those cells that will give rise to improved plants. Success depends on our ability to make plant cells grow like microbes, to eliminate the ordinary cells, and to convert the more desirable cells back to entire plants at will.

The method has worked with tobacco, an important crop in Connecticut, and has great promise for improving food plants too.

Excised stem tissue of tobacco can be readily cultured in petri dishes on agar medium containing mineral salts, vitamins, plant hormones, growth substances, and sugar. In a flask containing a liquid medium, cell suspensions can be produced which can be transferred to a solid medium. At moderately high densities, most cells will form cell clumps which in turn generate plantlets. The plantlets grow into normal tobacco plants if transplanted to soil in the greenhouse.

The first problem is to introduce variation from which the desirable traits can be selected. One way to do this is to treat the cells with a mutagen which causes chemical changes in the genes. Ultraviolet light is a convenient mutagen. There are also several chemical mutagens that are easy to use but dangerous to handle since they are also carcinogens.

To induce mutations, cells spread on agar medium are placed under a germicidal ultraviolet lamp for a few minutes, or are bathed in a solution of a chemical mutagen that can be removed later by washing. Most mutations either kill or cripple the cells, but some may enable a cell to survive conditions that otherwise might kill it.

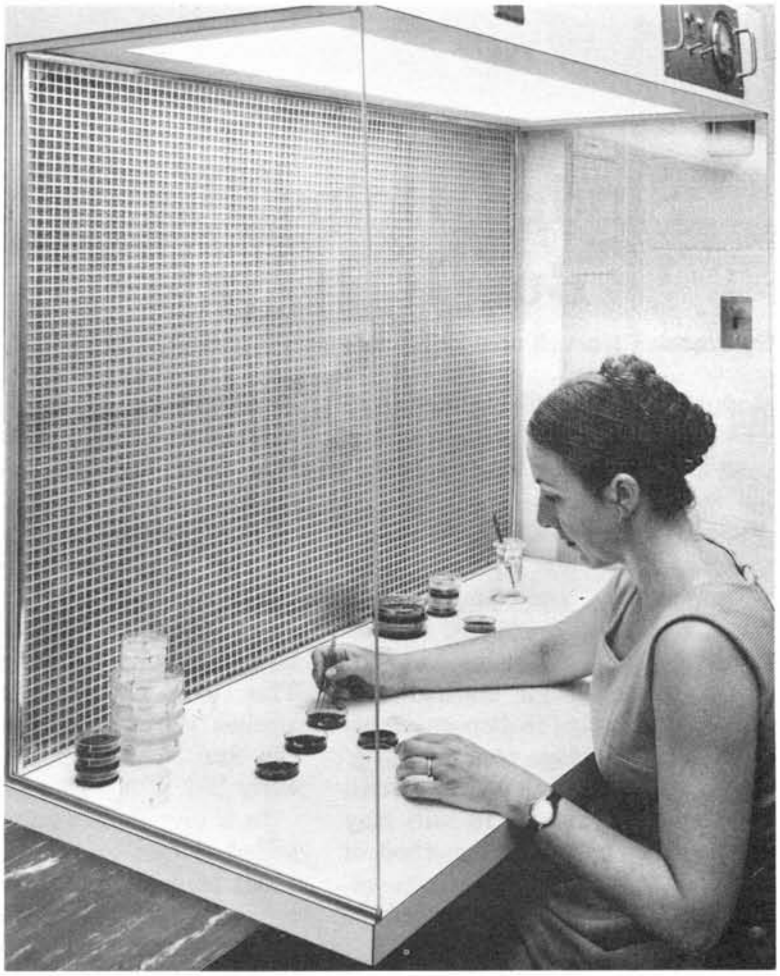
For example, ordinary susceptible cells may be killed by a bacterial toxin, but if they have been treated with a mutagen, some of the mutant cells may survive and may grow into cell clumps that resist the disease caused by the bacteria and their toxin.

Our collaborator, Dr. Peter Carlson of the Brookhaven National Laboratory, obtained tobacco plants resistant to wildfire disease in this way from mutagen-treated haploid cells.

Here at the Station, we are attempting to recover mutant haploid cells which metabolise glycolic acid at different rates than normal tobacco cells.

Glycolic acid is a key substance in photorespiration. This process occurs in the light and releases newly fixed carbon as carbon dioxide inside the leaf. Photorespiration does not occur to a significant extent in high-yielding crops like corn and sugarcane, but it appears to set limits on the total amount of carbon fixed by tobacco and food crops like wheat and soybean (see Zelitch, *Frontiers of Plant Science,* Fall, 1972). It may be possible to increase the photo-

Haploid plants emerging from anthers placed on culture medium.



Anther and tissue transplanting carried out in a stream of clean air by Sandra Anagnostakis.

synthetic yield of food by regulating glycolic acid metabolism.

We have obtained cell cultures, after mutagen treatment, that grow well on levels of glycolic acid that are toxic to normal cells. We also seek mutant cells that are unable to use glycolic acid as a carbon source. Both kinds of mutants should tell us more about how we can genetically "turn off" or "turn down" photorespiration and more about the effect on plant growth.

It is much easier to select mutants from haploid microbes than from diploid higher plants. Diploid cells have two sets of genes, one from each parent, while haploid cells have only one set of genes. In such a cell, a mutant gene is expressed without interference from the other gene copy present in a diploid cell. Fortunately in tobacco, and in several other plants, haploids can be produced by culturing pollen.

To do this, the pollen-bearing anthers are removed aseptically from the flower buds and placed on a culture medium. After a period of up to 10 weeks, the anthers give rise to embryo-like haploid plants that have roots, stems and leaves.

These can be transplanted to soil and grow into tobacco plants that appear normal. Haploid plants, however, are usually slightly smaller than diploid plants and their flowers are sterile.

Unless it can be turned into a diploid plant, a mutant haploid is a laboratory curiosity. Development of root and shoot in a small clump of cells depends on the correct balance of hormones and growth regulators in the culture medium. This balance has been worked out for several plants.

A proportion (10-25%) of the tobacco plantlets formed by cell clusters become diploid during the process of growth. The frequency of spontaneous change to diploid can be increased by treatment with drugs like colchicine which interfere with nuclear division and cause chromosome doubling.

The advantage of a doubled haploid is that both gene copies are identical. Thus, they are "instant inbreds" and breed true when self-pollinated. Diploid mutants can be used for further breeding experiments or may be directly planted without further improvement.

There are clear opportunities for development of all kinds of mutants that affect many biochemical and developmental pathways in plants. Some problems we are working on now include resistance to herbicides, resistance to toxic metals like zinc in the soil, and slower dark respiration. The only limitation is our ability to devise selective techniques or conditions in which only the mutants we want can survive.

The method of culturing plant tissues can also be extended to meristems. These are the organised groups of cells located at shoot tips. They are responsible for shoot growth and development. Meristem fragments of several ornamentals will generate plantlets and the technique is being used to multiply orchids, chrysanthemums and *Freesia* corms.

In the future, other techniques will be used to improve plants. Tobacco cells stripped of their cell walls have been fused together to produce new hybrids, and genetic information from bacteria has been transferred to several higher plants.

For annual crops that are easy to breed, cell fusion provides an opportunity to make hybrids that cannot be made by sexual means. In several laboratories, researchers are attempting to make non-legumes fix nitrogen from the air either by direct gene transfer in extracts or by fusion of cells from different plant families.

While some of these experiments may sound like science fiction, it is clear that plant improvement must be advanced as rapidly as possible to help food production keep up with a growing population.

Sludge Helps Plants Grow But Is It Safe To Use?

By Wendell Norvell and Brij Sawhney

MORE THAN 100,000 tons of sewage sludge are produced each year in Connecticut, creating a major expense and a large disposal problem for treatment plant operators.

Incineration and landfill are common disposal methods, but increasing costs and rising volumes create the need for cheaper and less wasteful ways to dispose of this byproduct of sewage treatment.

For many cities and towns, addition of sewage sludge to soils may be the most economical method of disposal. Use as a soil amendment is particularly attractive since it may become an asset instead of a liability. However, soil application must be done properly to safeguard both health and continued soil productivity.

The major benefits are well known; because sludge is rich in organic material, it improves the physical characteristics of the soil. It also serves as a fertilizer because sludge contains appreciable amounts of nitrogen, phosphorus and micronutrients. These benefits may be outweighed, however, by both short-term and long-term problems.

Temporary difficulties may include disagreeable odors, release of excessive amounts of nitrogen to ground waters, presence of disease-causing organisms, and damage to plants by soluble salts and ammonia. Fortunately, these short-term problems can be overcome by proper sewage treatment and suitable application techniques.

More serious and more permanent hazards may be caused by the heavy metals in sewage sludge. Heavy metals like zinc, copper, cadmium, and nickel tend to accumulate in soils where they may depress plant growth or make crops unfit for consumption. It is imperative that the reactions of these metals in soils and their influence on crop growth be well understood.

Since 1971 we have been studying the uptake of heavy metals by crops grown in soils treated with large amounts of sewage sludge. This work extends the pioneer studies made by Herbert Lunt at the Station during the 1940's and early 1950's.

In a greenhouse experiment, we added sludge to a sandy loam and a silt loam at the high rate of 89 tons per acre. Corn grown on the sludge-treated soil was compared to corn grown on similar soil with the same amounts of heavy metals in inorganic form, and to corn grown on untreated soil.

Conventional fertilizer was added as needed to make sure that all plants had similar amounts of nutrients available. Lime was added to make acid soils less acid and nearly neutral for comparison.

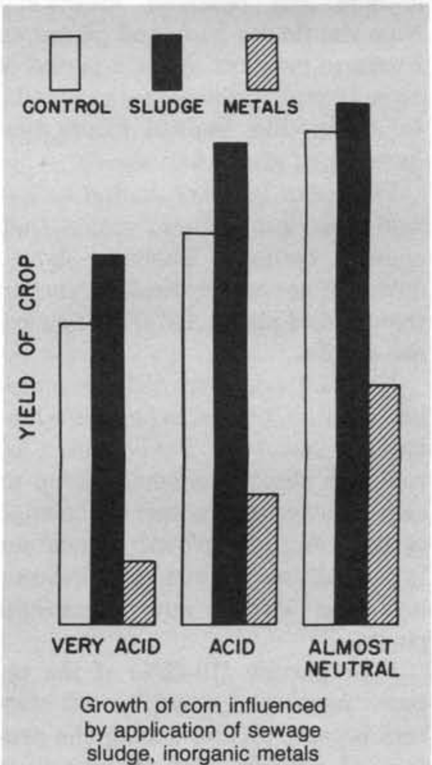
Yield was greater on sludge-treated soils than on those that received only conventional fertilizers. This difference, shown by the bar graph, was undoubtedly due to the improvement of physical properties of the soils. Sludge treatment increased water-holding capacity of the sandy loam by 50 percent and the silt loam by 30 percent. Both are common soil types in Connecticut.

But in the soils that received zinc, copper, cadmium, and nickel as inorganic compounds, severe reduction in the growth of corn was evident. Since the toxicity of these metals was most severe in the acid soil, liming reduces the hazard.

The initial toxicity from heavy metals in sewage sludge is low because of the high organic matter content. But, as this organic matter breaks down, more metals will be released, and toxicity may become serious. Thus, the severe toxicity of heavy metals added as inorganic compounds is a clear warning against the use of excessive amounts of metal-contaminated sludge.

Although sewage sludge and inorganic sources of heavy metals differed greatly in their toxicity to corn, heavy metals were taken up in like amounts regardless of the source. In corn grown in soil treated with metals from either source, zinc concentrations increased eight times, nickel concentrations increased two times, copper concentrations increased three times, and cadmium concentrations increased 30 times as compared to corn grown in untreated soil.

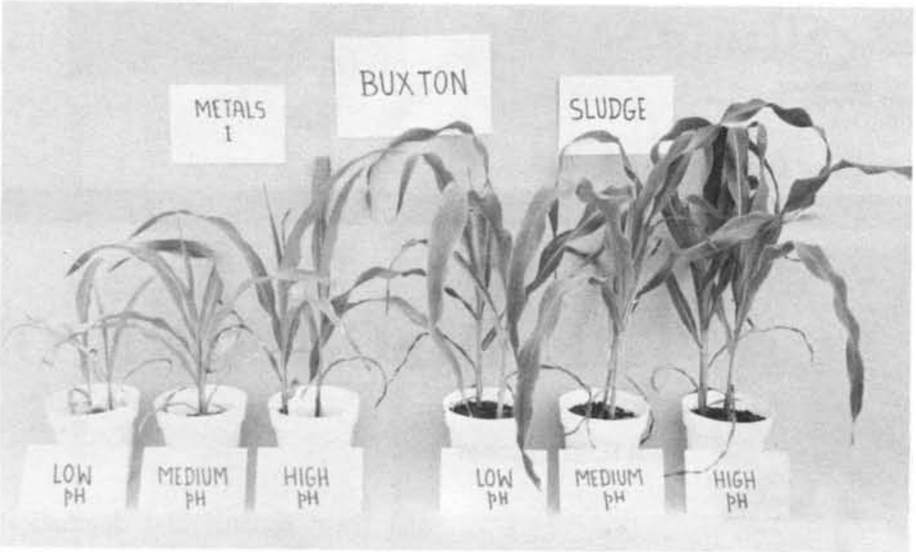
Zinc, copper and nickel, are so toxic that serious plant damage occurs long before they can be accumulated in amounts that would



Growth of corn influenced by application of sewage sludge, inorganic metals and soil acidity.

endanger consumers. Cadmium, however, is harmful to animals at much lower levels and, as we found, this metal readily enters plants. Fortunately, the level of cadmium in sludge is low unless it has been contaminated by industrial wastes.

Sewage sludge is a valuable soil amendment when good quality sludge is applied at moderate rates. However, our studies indicate large or repeated applications of heavy metal-contaminated sludge can harm plants or make them unfit for consumption.



Corn grown in soil amended with sludge containing heavy metals compared to corn grown in soil containing the same quantity of heavy metals as inorganic compounds.

NEW PUBLICATIONS

Three new publications have been issued by the Connecticut Station since the last issue of *Frontiers*. Requests for copies should be addressed to Publications, Box 1106, New Haven, CT. 06504. The results of many other scientific investigations carried out by Station scientists are published in scientific journals. During the past six months, 74 such articles have been submitted for publication.

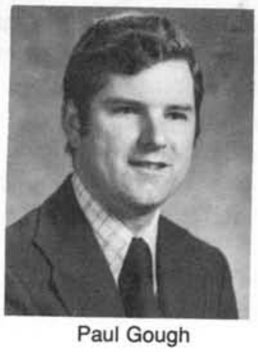
- B 737 77th Report on Food From Connecticut Markets and Farms, 1972. J. Gordon Hanna
- B 738 Commercial Fertilizers Inspection Report for 1972. J. Gordon Hanna
- B 739 Comparative Efficiency of Energy Use in Crop Production. Gary H. Heichel

Jones Medal To Be Awarded Jan. 25

The plant geneticist who developed high-yielding rice will be honored by the Station on January 25 when he is presented with the Donald F. Jones Memorial Medal. This medal is awarded by the Station Board of Control in recognition of outstanding scientific achievement.

The recipient will be Dr. Peter Jennings, leader of the Inter-American Rice Program in Colombia, South America. While working with the Rockefeller Foundation in the Philippines, Dr. Jennings employed genetic engineering techniques to develop IR-8 rice. This variety, which grows shorter but yields more grain than previously used varieties, has made a substantial contribution to the Green Revolution in Asia.

A tea will be held in the Donald F. Jones Auditorium at the Station at 3 p.m., with the award ceremony and a lecture by Dr. Jennings following. The public is invited to attend.


Paul Gough

Bruce Miner, who was Station editor for the past 18 years, retired Sept. 1. He accumulated journalistic honors for the Station and left *Frontiers* a fine, well-known publication. It is appropriate to note the change here rather than simply change the name on the masthead.

Editing a publication like *Frontiers* is a distinct challenge because its readers are a healthy mixture of scientists and citizens, researchers and consumers. Each is bound to the Station by the printed word and the desire to learn more about how plant scientists serve Connecticut and the world. It is an honor to continue reporting on the work of Station scientists.

P.G.

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THE DISCIPLINE OF DISCOVERY

Paul E. Waggoner

THE ROLE of the Experiment Station in increasing the amount of food available was discussed in the last editorial. Now, it's time to tell how we strive to do effective research on this subject.

Although we use flasks, instruments and experimental plots, I shall concentrate on another essential: a triple discipline like that of an acrobat who is juggling two balls while watching for a third. On the one hand, we must reason how plants and insects grow, while on the other, we must observe how they really grow, and at all times, watch for ways to make our discoveries useful.

I learned this was nothing new when my son brought home a paper about Roger Bacon, the man who described the discipline of a fruitful science after a millenium of fruitless theories spun in the cloisters while practical men were left to travel their hard and hungry way.

Although Bacon wrote in 1620, as the Pilgrims made their way to the new world, it is still his kind of discipline that we strive to keep at the Station. There is no better way to describe his thoughts than through liberal use of his words.

Observation and experimentation are checks on reason, and knowing all about medieval reasoners and their rational and dogmatical sciences, Bacon realized a check was needed.

"When the rational and dogmatical sciences began, the discovery of useful works came to an end," Bacon wrote. The result, he said, is endless scholarly repetition and a product "fruitful of controversies but barren of works."

Since men, as Bacon noted, tend to think there is more order and regularity in the world than there really is, the fondness for generality is still with us, 350 years later.

"The mind longs to spring up to positions of higher generality, that it may rest there; and so after a while, wearies of experiment," Bacon said. Since this is no less true today, we must discipline ourselves to leave our desks, go to the field, and settle questions "not by argument, but by trying," as he suggested.

Although the Baconian discipline checks reason by observation, it does not neglect reason, but uses it to guide observation.

Bacon noted that although "it may happen that a man shall stumble on a thing . . . far better things, and more of them, and at shorter intervals, are to be expected from man's reason and industry and direction and application than from accident."

The third part of the discipline we try to keep is a watchfulness for utility, avoiding the natural tendency to leave earthy experience and matter and fly to what Bacon described as "the serene tranquility of abstract wisdom.

We must instead stick to things as they are. Even banal and ordinary things such as corn and compost must be admitted to natural history. "Nor is natural history polluted thereby; for the sun enters the sewer no less than the palace, yet takes no pollution." "For whatever deserves to exist deserves also to be known."

The disciplinary trinity is recapitulated in the philosopher's own words: "The men of experiment are like the ant; they only collect and use; the reasoners resemble spiders, who make cobwebs out of their own substance. But the bee takes a middle course, it gathers its material from flowers of the garden and of the field, but transforms it by a power of its own."

Finally, being no Brahmin, Bacon gave an admonition to all: "That they consider what are the true ends of knowledge, and that they seek it not either for pleasure of the mind, or for contention, or for superiority to others, or for profit or fame, or power, or any of these inferior things; but for the benefit and use of life."

This is the way to bring home the bacon for a hungry world.

Frontiers of Plant Science published in May and November, is a report on research of The Connecticut Agricultural Experiment Station. Available to Connecticut citizens upon request.
Vol. 26 No. 1 November, 1973 ISSN 0016-2167 Paul Gough, *Editor*