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THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION
NEW HAVEN
Rearing Gypsy Moths In the Laboratory

David E. Leonard and Charles C. Doane

The gypsy moth is still a major problem after almost a hundred years in North America. It has now reached the same status here as in the Old World where it originated. The characteristic pattern is violent fluctuation in numbers with periods of scarcity followed by defoliation of large areas of susceptible woodlands. These severe outbreaks have been common in Connecticut since 1950.

A goal of entomologists is the control of pests in forests by some biological means. One handicap to progress toward biological control of the gypsy moth is that the insect is active only a few weeks of the year, prospering on developing foliage of a few species of trees. Young leaves of oaks, which are the most common group of trees in our woodlands, serve as the principal food of gypsy moth larvae. Production of fresh, young leaves of oaks throughout the year in the greenhouse is difficult. An artificial diet is an obvious solution, but another problem—that of disease—immediately arises.

Some diseases that occur sporadically in the field become a major problem when caterpillars are reared in the laboratory. The most serious is the “wilt” disease caused by a polyhedral virus. We found that this virus disease could be greatly suppressed by treating eggs with a dilute solution of sodium hypochlorite, an ingredient in household bleach.

The artificial food study used as a starting point a diet devised for the pink bollworm, a pest of cotton. The first results with this food, when fed to gypsy moth caterpillars, were encouraging. However, it was obvious that some changes were needed, for the survival rate was low and egg production poor. Several modifications of this diet have now provided food which appears nutritionally to be as good or better than fresh oak leaves. Comparisons with larvae in the field show that the laboratory caterpillars are considerably larger, and the adults from them lay over twice as many eggs.

The artificial food contains no leaf constituents and is therefore an alien diet for the gypsy moth. Among the ingredients are wheat germ, casein, salts, sugars, a number of vitamins, fatty acid, and antimicrobial agents. Agar is used to gel the food. The ingredients are blended with a high-speed mixer and poured hot into sterilized 3/4-ounce creamers or petri dishes. In consistency and appearance it somewhat resembles cream cheese.

Sheila Thompson transfers a gypsy caterpillar to a clean petri dish.

In the laboratory, gypsy moth eggs are surface sterilized, and the larvae reared under glass on an artificial food. Many of the fully grown caterpillars are larger than those found in woodlands, and adults from them lay more eggs.

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Eggs, stuck to adhesive tape and surface sterilized in sodium hypochlorite, are placed in creamers with media. After hatching, the small caterpillars are kept in creamers for about a week, then transferred to petri dishes where they are less crowded. Pieces of food are placed in the petri dishes. Maintenance of laboratory cultures is time consuming, for the caterpillars must be fed twice weekly and rearing containers must be changed frequently to prevent diseases.

Large numbers of larvae can now be reared on the artificial diet for experimental studies. Parasites, predators, and pathogens can be studied throughout the year rather than just for a few weeks. Hopefully, new knowledge of the biology, ecology, behavior, and genetics of the gypsy moth will eventually show us how to lessen gypsy moth damage in Connecticut woodlands.

In the News

Raimon L. Beard, entomologist at this Station, left in October for a year in Thailand where he will work with the Atomic Energy Commission for Peace. He expects to investigate biological effects of radioisotopes on insects, and other aspects of insect physiology. . . .

Harley Tomlinson is working with Saul Rich in the Department of Plant Pathology and Botany on air pollution damage to plants. This research is supported by a grant from the U.S. Public Health Service. . . . John S. Boyer has been appointed to the Station staff to study water flow through plants and the rate of water loss from plants. . . . Saul Rich is now president-elect of the Society for Industrial Microbiology. . . . Director James G. Horsfall has been appointed to the Latin American Science Board of the Office of the Foreign Secretary, National Academy of Sciences.

Clayberg and Kuiper Honored

Carl D. Clayberg, Department of Genetics, received the certificate of achievement in horticulture awarded by the Federated Garden Clubs of Connecticut in Hartford on October 13. Dr. Clayberg was honored for his research in inheritance in tomatoes and for his achievements in hybridizing gesneriads. The other certificate of achievement awarded recognized the work of P. J. C. Kuiper for his research on frost- and drought-resistance in plants while on the Station staff in Soils and Climatology.

The Hungry Nations

DURING the last decade, I have traveled extensively over the globe and have seen some developing nations close-up. Distressed, I come home wondering what agricultural science can do to raise the food production in the hungry nations, as Paddock and Paddock so aptly name them in a recent book.

A nation may be hungry because its land and its crops are hungry. Even farmers may be hungry there. Sometimes hunger is attributed to lack of education, bad health, bad land tenure, bad transportation, or lack of industrialization. These may result from hungry land. They do not produce it.

In Connecticut we knew more than a century ago about insufficient food from the land. The Colonists had "worn out" the land, first in the fertile valleys and later on the marginal hillsides. Then they put their children and chattels in wagons and went west to wear out still other land. Thus, they repeated the pattern that goes at least as far back as Nineveh in the Fertile Crescent of the Near East, where the farmers wore out the land right down to the point of no return. And Nineveh died, and so did Machu Picchu in Peru in later times.

Connecticut did not let this happen here. With characteristic ingenuity, Connecticut introduced a new invention to create inventions. It set up an experiment station "to put science to work for agriculture." As a result Connecticut land is no longer tired and worn out. By now the land yields much above the pristine rate. In 1959 Connecticut grew as many potatoes on 5,500 acres as it did on 22,000 acres a centurty ago, an increase of 4 to 1 in efficiency.

If a hungry nation is to have more food, it must extract more food from each acre of fertile land it has. If a farmer in such a nation moves on, he can move only to marginal land. This is where the hungry soil is. The fertile land is all in use, often growing cities instead of food.

The question is, why is marginal land hungry? The experiment stations have learned some answers to that question, and much marginal land has been converted into good farm land. The tops of the hills in the Catskills are beautifully cool for cauliflower, but cauliflower hungerers there without a few ounces of borax per acre. In Colonial days, cattle died in some New Hampshire intervale because the grass could not find enough cobalt in the soil, and winter crops in Florida depend on a few ounces of copper or zinc per acre. Without cobalt, or copper, or zinc, the land is marginal.

We try hard to export our vaunted "know-how" to the hungry nations. When it falters, we are puzzled. I saw a similarly puzzled Iowa farmer when I was a boy in Arkansas. He spent his money. He tried. He failed. And he returned to Iowa when he learned that he could not inject a corn-hog know-how into a peanut-cotton economy.

(Continued on page 8)
Listing Plant Names and Namers

Punched cards and electronic devices speed publication

Four years ago we noted in Frontiers the progress being made by Sydney W. Gould in compilation of an International Plant Index. The Station has recognized the promise of this undertaking by providing facilities for Mr. Gould and his small staff. The work is sponsored jointly by this Station and the New York Botanical Garden, with the financial support of the National Science Foundation.

Mrs. Dorothy Noyce, a co-author with Mr. Gould of a new volume in the Index series, reports on the current status of this unusual venture of the Station.

Dorothy E. Noyce

In the systematic classification of plants, known to scientists as taxonomy, it is important to know when and by whom a particular kind of plant was first named and described. Indeed, the name of the botanical author, as these taxonomists are called, is properly a part of the full scientific name of the plant itself.

Publication on July 1, 1965, of Authors of Plant Genera places on record in one volume a wealth of information previously scattered throughout the literature of plant taxonomy. This comprehensive work lists the full names of approximately 5,800 botanical authors, with the birth and death dates, and the country in which each man worked. In addition to individual authors, over 1,400 collaborations are noted, and with the name of each single author is a listing of all collaborations in which he participated.

Although many fine author bibliographies have been compiled in the past, the task of keeping any listing up to date in the face of the “information explosion” is extremely difficult. Pritzel's and Jackson's admirable works carry the researcher only to 1881. Many later bibliographies are limited either to a single taxonomic interest or to a particular geographic area. In Authors of Plant Genera an attempt has been made to include authors not only of the flowering plant genera, but also of the bryophytes, algae, fungi, lichens, slime molds, and bacteria.

The punch card technique, which is basic to all of our Plant Index operations, is admirably suited to the compilation of a list of this scope. New and corrected information is interpolated into existing files as soon as it is received so that files are current at all times. Publication of the 336-page book thus became simply a matter of recording some 22,000 cards on a single magnetic tape. This tape then be-
came the basis for a computerized typesetting process.

Unique to this book is a guide to 3,000 name abbreviations, recorded exactly as found in standard botanical sources, together with the names of the authors to which these abbreviations refer. One of the interesting sidelights of such a listing is the revelation that one man may be referred to in four or five different ways and, conversely, that identical abbreviations may be used to designate as many as four different people.

Although ambiguity may be somewhat reduced by the use of an author's entire name in a citation, even this does not guarantee clarity in a science which boasts two Robert Browns, two Annie Smiths, a pair of Ludwig Benjamins, and a small platoon of Karl Muellers. The use of author codes, unique for each authorship (single person or collaboration), makes it possible to identify the author of any given taxon far more speedily and precisely than has heretofore been possible. A code of uniform length also facilitates machine processing of author data. Such a code is necessary if the next logical step in author research is to be completed, namely, the compilation of a complete generic author-publication bibliography.

In his introduction to the first volume of this series, *Family Names of the Plant Kingdom*, project director Sydney W. Gould says, "One of the primary purposes of this project is to achieve both rapid recording and rapid retrieval of botanical information, and to permit botanists to spend less time searching the literature and more time in the field or laboratory."

Correct author identification is but one aspect, albeit an important one, of this goal. Presently work is going forward on a generic index of the entire plant kingdom, fossils as well as living genera. Following the form pioneered in the first volume, each entry is identified by a taxonomic number which tells at a glance the primary group into which the plant falls. In addition, each card records the author who described the plant, gives the date of the description, and shows where it can be found in the literature. The generic file now numbers approximately 30,000 cards. Finally, with synonyms included, we expect to have between 75,000 and 100,000 entries.

Besides taxon and author cards, the Plant Index maintains an ever-growing publications file which makes possible the retrieval of bibliographic data of almost infinite scope and variety. Station Director James G. Horsfall describes the experience of many research workers in his Foreword to *Authors of Plant Genera*: "When I did a small taxonomic thesis in the graduate school, the bookkeeping aspects of the nomenclature took as much time as the research." By utilizing various combinations of data already on file, we can in very short order select and print "bookkeeping" information which would consume weeks or months of routine library searching.

It is possible, for example, to extract very quickly a complete listing of all plants named by a given author, together with date, journal, chapter, and page of each citation. It is an equally simple process to compile, say, a comprehensive bibliography for flowering plants described between the years 1900 and 1910. All taxonomic entries of a given journal, either for a single issue or for a 50-year period, may likewise be indexed readily.

Almost any kind of information can be coded into taxon cards, whether it be global in nature, such as location and distribution, or highly specific, such as pulp content or chromosome count. Toxicity, host-parasite relationships, breeding mechanisms — these are but samples of the areas in which botanists are already considering the possibilities of data processing methods.

In our offices a fledgling geographic code is even now in use as a functional part of the mailing system. This four-digit code, capable of identifying any county-size area in the world, vastly facilitates the sorting of materials for regional mailings and the assignment of correct postage.

The uses and adaptations of a plant index, not only for botanists but for those in any field where plants or plant products are involved, appear to be bounded only by the limits of need and imagination.

### New Publications

The publications below have been issued by the Station since you last received FRONTIERS. Address requests for copies to Publications, The Connecticut Agricultural Experiment Station, Box 1106, New Haven, Connecticut 06504.

**Insect Pests**

C199 (Revised) *Dogwood Borer*. John C. Schread.

**Fungicides**


**Report on Inspections**


**Statistical Relations**

B674 *Statistical Relations in Fertilizer Inspection*. C. I. Bliss.

**Other**


*The Connecticut Station Today.*

A special circular describing some lines of research believed to be of importance to Connecticut citizens now and in the future.
Research Underlies Diagnosis
Of Plant Disorders

Frances W. Meyer

Any laboratory that diagnoses disease, whether in humans or plants, uses techniques based on research and is dependent upon the imagination and soundness of that research.

Diagnosing the plant troubles that find their way into our office is essentially detective work applied to plant pathology. We look for clues, get the story, and make laboratory tests when necessary in order to determine what disease is involved. Then we reach a conclusion or “educated guess” as to what has happened and why.

Any deviation from the normal plant behavior may bring the owner into our office. He may be concerned with the “blight” on rose leaves or his African violets. Often, all we need to do is examine the plant with our microscope. Perhaps this reveals cyclamen mites distorting the growth point of the violet plant, or powdery mildew curling the leaves of his climbing roses. These two causes of sick plants are relatively common, and the controls are well worked out for prevention and cure. But these answers came from someone’s research.

Many diseases such as the powdery mildews or black spot of roses can be recognized on sight. Fungi often leave marks of their identity somewhere on the plant in the form of fruiting bodies. An azalea leaf gall or plum black knot is actually an enlargement containing fruiting bodies, but the swelling is distinctive for the disease and needs no further investigation. However, many fruiting bodies are so tiny they can be seen only by using a microscope. We carefully pick out their minute bodies or slice them thinly with a razor blade so they can be examined under the high power microscope. We may use stains which color the fungus tissue and not the plant.

If fruiting bodies are not present, the fungus can be made to produce them by placing the diseased plant in a moist chamber. This can be a large or small petri dish or a closed plastic bag or box. Water or moist blotting paper is enclosed with the diseased material to maintain the high humidity necessary to encourage the production of fruiting bodies.

A similar idea is involved in culturing, except that the diseased material is surface sterilized and a small piece of it placed on a sterile medium such as agar, or in sterile water. Culturing may be necessary either to determine a pathogenic agent or to distinguish between two possible pathogens. Sometimes special conditions of light or temperature or both may be needed to bring out the fruiting bodies. Much recent research has been done on this process, particularly on the relation of different enzymes to spore production — a very basic consideration.

This forcing of the reproductive phase of a fungus is often necessary for identification. The numbers of the fungi range in the millions, and they are grouped taxonomically according to the character of their fruiting structures. We use this information to identify a fungus much as the stranger needs a guide of some kind to find a certain house in a large city.

Diagnosis of plant ills goes beyond knowledge of plant pathology as limited to disease-producing organisms and their control — it also requires some knowledge of the conditions around the plant. A gardener may start the trouble unwittingly by overfertilizing, overwatering, or making plantings where light and moisture conditions are unfavorable. We need all the information we can get about the plant, its history and care, to make an accurate diagnosis.

A vexing problem of laurel leaves that have turned yellow and green, perhaps with brown tips, illustrates this need for all available information about a plant. I have seen just such leaves on laurel infected with Rhizoctonia root rot, and the same symptoms on a bush infected with another fungus called shoestring rot. Black vine weevils can cause similar symptoms, as can the excessive use of fertilizer or use of the wrong kind. In such cases we need samples of soil, roots, and information about care. It is a waste of time and material to add anything to the soil without first determining the reason for the chlorosis.
and discoloration. Home diagnosis can easily be corroborated by laboratory analysis before remedies are applied.

Overtreating frequently results from not realizing that plants differ in their needs for food. A plant can vary in its need for nutrients depending on the conditions under which it is growing. Plants, unlike people, do not have to be fed often, and one fertilizer application can last for quite some time, particularly with house plants. A favorite but commonly harmful remedy is to feed a plant if it looks “sick” — when the basic cause can be something quite different.

For accurate diagnosis a complete story is essential and often provides the clue to the trouble. For instance, branches from a jade plant were sent to us because they were shriveling badly and falling off. It was a huge old specimen with a history — and the owner was frantic because it was such a valuable plant. No disease or insects appeared on the samples of branches or on the roots. Care had the plant not changed in 15 years, and analysis showed a good soil for this particular type of plant. No gas fumes were present, nor could have been. Digging for facts revealed that the wood was too big, in which the wood was potted had recently been crossed — this explained why the leaves were drooping and branches withering.

We also identify plants for various reasons — sometimes connected with police cases — usually just because some citizen wants to know “what is it?” It may be a weed in the lawn or it may be a piece of a plant from the garden of a friend. We also identify mushrooms in case of emergency — a child in a playpen will reach out and grab a toadstool — the mother finds a piece in the baby’s mouth. She is frightened and wants to know if this is a poisonous mushroom. Usually it is not, but there are poisonous ones that grow in the lawn. And some gatherers of wild mushrooms are careless and end up in the hospital. Knowing what kind of mushroom is involved may be essential to proper treatment. Identifications extend to poisonous plants (which children and animals may eat) as well as poisonous mushrooms.

Problems of florists, nurserymen, and vegetable growers furnish many basic questions from which research is evolved. The results of this research are part of the world’s body of knowledge accumulated at this and other institutions to help solve plant problems.

How Do Fungi Overcome Disease Resistance In Plants?

Peter R. Day

A British plant pathologist, William Brierly, once remarked that “fungi are a mutable and treacherous tribe. He may have had in mind the heavy toll they take of food and fiber. Throughout the United States every year nearly one-fifth of the field crops, fruits, and vegetables we grow are destroyed by plant diseases, most of them caused by fungi.

On the credit side, however, we have to remember that fungi are also useful. The yeasts we use in brewing and baking and the molds which yield antibiotics are but three instances.

To the geneticist, fungi offer unique opportunities to bridge the gap between the simplest and the most complex living organisms. They are in some ways like bacteria and in other ways like higher plants. John Puhalla and I are using two plant disease fungi and a toadstool to try to gain some insight into one of the many perplexing details of plant disease caused by fungi. We are trying to find out more about the nature of resistance to disease in plants and about the ways in which fungal pathogens overcome this resistance.

Much can be discovered from a chemical approach. What chemicals are formed when a disease resistant host is invaded by a pathogen which it cannot successfully repel? Work of this kind is being actively pursued at this Station already. We, however, are interested in finding out how the disease organism itself behaves. Disease resistant plants often prove susceptible to new, mutant, forms of disease fungi. We want to find out how the mutant pathogen avoids the resistance encountered by the non-mutant form. We say "avoids" rather than "is insensitive to," because of evidence from several sources that suggests our mutants simply fail to trigger the host plant’s resistant reaction.

A first step is to obtain a number of mutants of this kind for our study. Although these already exist in our greenhouses and on our farms, they vary too much. We must use mutants derived from one source and Puhalla and I are therefore working on this. Even though these are rare, they can be found by using a resistant plant to select them. Non-mutant, or avirulent, disease spores inoculated to a resistant plant will not give rise to symptoms. However, we treat the spores before inoculation with an agent that causes gene mutation. As a result of this, perhaps one spore in 100,000 now has the virulent mutant character we are looking for. Only these spores give rise to disease lesions from which they may later be recovered.

We include markers as a precaution against contamination. That is, the recovered mutant is examined to see if it has a marker character that the original non-mutant had. Mutations resulting in a difference in color may be used as markers. Another kind of marker is a requirement for a chemical which the fungus is normally able to make for itself. A fungus carrying such a marker can only be grown on a synthetic culture medium to which this chemical has been added. Of course if the host plant cannot also supply the chemical, the marker will be of no use because the fungus carrying it will be non-pathogenic. Not surprisingly, scientists have speculated that such chemical requirements, and the failure of certain plants to satisfy them, could account for resistance. It seems that most examples cannot be explained in this way.

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Fungi Overcome (Continued from page 7)

Experiments are presently underway with the tomato leaf mold disease (Cladosporium fulvum) and corn smut (Ustilago maydis). In both of these fungi markers have now been obtained and are being tested in the two controlled environment chambers recently installed in our Slate Laboratory (see photo). In these chambers standardized plants and inoculation conditions may be obtained at any time of the year.

Our work with the ink-cap toadstool (Coprinus lagopus), which is not a pathogen, avoids the complications of growth on the host plant. We hope it will answer some questions bearing on the problem of why many important plant disease organisms, for example the cereal rusts and some smuts, can only be grown on their host plants and never on artificial culture media.

Coprinus, like the rusts and smuts, exists in two forms. One is made up of cells each containing a single nucleus. The other form is made up of binucleate cells, the two nuclei in each cell are usually different from each other. In the pathogens there are important and significant differences between the two forms. They may, for example, attack entirely different plants. Often one is a weed, the other a crop plant. In Coprinus we are trying to find out what simple features of metabolism vary in one form from the other. We are also studying the formation of certain enzymes in specially produced mutant strains and hope to progress from these simple beginnings to the pathogens themselves.

Mutable and treacherous the fungi may be, but also useful in the search for new knowledge of disease resistance.

To Study Farm Policies

The White House announced on November 3 that Director James G. Horsfall is one of the 30 “Americans of broad experience and great talent” appointed to the National Advisory Commission on Food and Fiber.

A Note To Readers

We have made many corrections in the stencils used to address FRONTIERS to regular readers. We thank all those who replied to our card of inquiry, particularly those who commented on the publication. Others are invited to notify the editor if they wish to continue receiving FRONTIERS. Change of address requests should include both the old and the new address. Your ZIP code is part of your address if you live in the United States.