DUTCH ELM DISEASE

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I want to thank the NEFTIC for inviting me to come back to New Jersey. I grew up in New Jersey, in the town of Plainsboro, just outside of Princeton, in a community called "the Aqueduct." Actually, I am here today sort of illegally. New Jersey is not the only place with budget problems, and all out-of-state travel has been forbidden to people who are working for the Commonwealth of Massachusetts, so I was turned down on my request for funds to come here to keep my promise to speak to you. But I am glad to be with you anyway (without my university's financial support). I am also glad to have learned what "Cook College" is (now I know that it does not mean a College of Home Economics).

I can't tell you all about Dutch Elm Disease in a 20-minute speech, of course, because it is a very complex problem and has involved the time and efforts of many people for half a century.

We have done a number of different "DED" studies at the Shade Tree Laboratories, mostly under a (USDA-CSRS) Regional Research Project NE-25, which began in 19515 and just ended. This has been supplanted now by project NE-99, which began as of July 1 and will continue for five years. There was one person paid to give 100% time to this project at the Shade Tree Laboratories, Joe Demaradzki, and most of the experiments on DED were carried out by him. It is my regret to report to you that five weeks ago he died of a heart attack, so he doesn't have a chance to hear me say, here today, "It was all possible because of his help." Joe was Senior Technical Assistant at the Laboratories.

Only a small part of our overall program involved the work on seeking mutations in American elm, but when Brookhaven offered to irradiate things for people in the late 1950's and early 1960's, we leaped to the chance to irradiate elm seed. I was told, "No, this won't do any good, because the elm seed is too big. You really should irradiate a single cell, like a pollen grain, if you must, because in seed irradiation what you will get will be just a change in one cell in the embryo, and it won't become a feature of the whole plant." But seed is what we had, so we had it irradiated to see what would happen.

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The stories we had been told were not true, because when we saw an obvious morphological difference in the plant, it carried through the plant. If there was a notch missing in one corner of the leaf, every leaf would have that notch missing on that same side of the leaf. If there was a white area down the center of the leaf, every leaf on the plant would have a white area down the center. (Unfortunately, the few "variegated" elms we created were stolen from the greenhouse, so I was unable to market an elm with a white center on the leaf. I've always wanted to find those trees in someone's front yard, so I would know who took them.)

I visited Dr. Seymour Shapiro, at Brookhaven National Laboratories, Upton, Long Island, and he kindly agreed to treat any elm seed we sent them, and so we started sending. We tried different levels of irradiation to find a level that would leave from 0.1 percent to 1.0 percent living. We successfully killed 99.0 to 99.9% of the seed with the irradiation, and with that level I was told you would get the largest percentage of mutations in the remaining seed.

We calculated the number of treated seed needed to end up with the number of seedlings we had space to plant. Since we were assigned only a few acres, we sent a half million seed each year and of this amount five thousand grew in the greenhouse when they came back from being irradiated. We also sent one bag of elm seed each year that was not to be irradiated, but merely to be kept and sent back. Thus we could see what would happen to the seed along the way, because it was going through the post. One year none of the seed of either lot germinated! Apparently, they must have been set on a radiator, or something equivalent, and killed en route.

Generally, the process was very successful, and of our half million we would get about 5,000 seed to germinate, from which 3,000 seedlings would reach the point of being transplanted into the nursery. Some of the other 2,000 were lost in transplanting and others were lost through a post-emergence death that was not related to damping-off fungi but was assumed to be a delayed effect of irradiation.

The seed were collected each May and sent in as soon as collected and cleaned of trash. They usually came back in September and were put into cold storage until greenhouse space was available. This had an advantage. Although most elm seed will germinate as soon as it ripens, some of it has a dormancy requirement that we were able to deal with by placing it in cold storage. The seed were taken out of storage about the first of January. They were put in the greenhouse and as soon as the seedlings came up we added artificial light to extend the day length. By May or June we were transplanting out to the field. Sometimes in the field it was so hot and dry that, even though we watered, all of the plants lost all of their leaves. Yet they all would live, and in the end we would have many long rows of beautiful elm trees.

About three or four years later, depending on which field we had used, we would inoculate with Ceratocystis ulmi through

There were oddities from the irradiation, visible effects that were not necessarily related to Dutch Elm Disease. We assumed that these were entirely separate effects and more a matter of curiosity than usefulness. We found leaves that looked like dandelion leaves, we had instances of leaves being covered with speckles and we found a number of other such abnormalties you find with other plants.

Most of the trees died of Dutch Elm Disease, of course. From 2 to 15 trees each year, from each half million seed, lived through the Dutch Elm Disease inoculation and they lived through in different ways. Sometimes the tree would die back, i,e, the branches would have dead twigs, the leaves would fall off and we would have a smaller tree.., which is not a very desirable way of living through DED. Occasionally, however, we saw something else, something that was very encouraging: this was a situation where the tree lost leaves here and there all over the twiggy structure but none of the twigs died! That was much nicer. We wouldn't mind a tree that would shed a few leaves when it got Dutch Elm Disease, if the twigs didn't die and the tree would come into full leaf the following year.

A letter from Dr. Harold Smith, who has taken the place in Brookhaven of Dr. Seymour Shapiro, stated that, as of 1972, Brookhaven would no longer irradiate things for biologists. There was apparently some pressure from other groups suggesting they shouldn't do this anymore - or perhaps we just "wore out our welcome." The trees from the last irradiation treatments will be inoculated sometime in 1975 and 1976.

As a result of this program, we now have 192 little trees set aside in a nursery in Belchertown, Mass.(This is far enough from the campus, I hope, so they can't think of building anything else there. Our nursery has already had to be moved three times!) Last week I marked the ten of these trees that seem to be best and also arranged for cuttings of them to be rooted, We want to have cuttings so that we can make valid comparisons, You can't compare clones by comparing individual trees, as you all know.If you have many cuttings from the tree, you will expect to have some variation among the cuttings; and you can't see just what is the range of this variation without inoculating 10 to 20 cuttings of each clone.

We tried hybridizing some of our earliest selection that did not come from this program, ones which we had set aside in the 1950's. We crossed them with Cornell's R-182 in both directions. I knew what it was like to be up in a young tree which was not able to support my weight, so I had to be on the end of a very long ladder, I tried to hold the tree, the paintbrush, the pollen, the bag that had been over the flowers, the wire by which the bag was tied closed, and everything else. I had only two hands, and I wished I had six. As usually, we had a good, cold March wind blowing all the while and chilling our fingers. And, after all this had been endured, the hybrids didn't have the resistance that was in <u>either</u> of the parents!

Oddly enough, the cuttings that we grew from some of the Cornell trees which Dr. Sinclair has spoken and written about, and from ours too, didn't seem to carry the resistance any more than the hybrids did! Maybe we are trailing a "will-o'-the-wisp," but people want the American Elm for its vase-like form so much that we are struggling to bring resistance to these trees.

I was surprised that I was asked to speak to you here today, because others have gotten so much farther ahead towards releasing resistant elms. Some of these are Frank Santamour at the National Arboretum in Washington, D.C., Larry Schreiber from Delaware, Ohio, Gene Smalley in Madison, Wisconsin, and Hans Heybroek (not with American Elm) in Wageningen, The Netherlands. All of them are more deeply into it than we are.

You see we had only about 5 acres to devote to this work, and so we couldn't do an experiment where half of the land was given to seedlings not irradiated. Since an experiment usually involves a comparison of some sort, if there is no comparison, there is no experiment. So I can't really claim our effort was even an experiment. All we were doing was trying to get as many mutants as we could, in as short a time as we could, and not to wait ten thousand years...because we were impatient. As a result we now have some trees growing in our small field that at least have not lost their twig or branch structure, although they have been inoculated three times with the Dutch Elm Disease fungus.

I can hope that in the future we will not only be able to use these resistant elms for themselves, propagated as cuttings, but also to hybridize between them. All of these young trees should begin to flower in a few years, which will provide a lot of natural crossing between these trees as well as controlled crosses. Of course, we will collect enough seed to plant the whole surface of Massachusetts and maybe the National Guard will come and help us do the planting! They ought to be doing something, and military things are not what people should be doing.

Some of the slides point out the problems we encountered and the results we obtained. It apparently was rather difficult for the Brookhaven Lab accurately to control the amount of irradiation that reached the seed so they were never sure of the exact dosage our seeds received. We did specify two levels and some of the strange appearing American Elms we obtained may be due to the higher levels of irradiation used. In some cases, you see that our trees had very deep notches in the leaves and some of the double-notched types looked like the cut-leaf varieties of <u>Crategous</u> and <u>Acer,</u> or even like dandelions! The growth rate was also variable, with some plants reaching a height of more than a meter in the same period of time that others grew only 10 centimeters. I don't know whether we could market trees that only grow as small shrubs, but we saved some of them anyway. These were

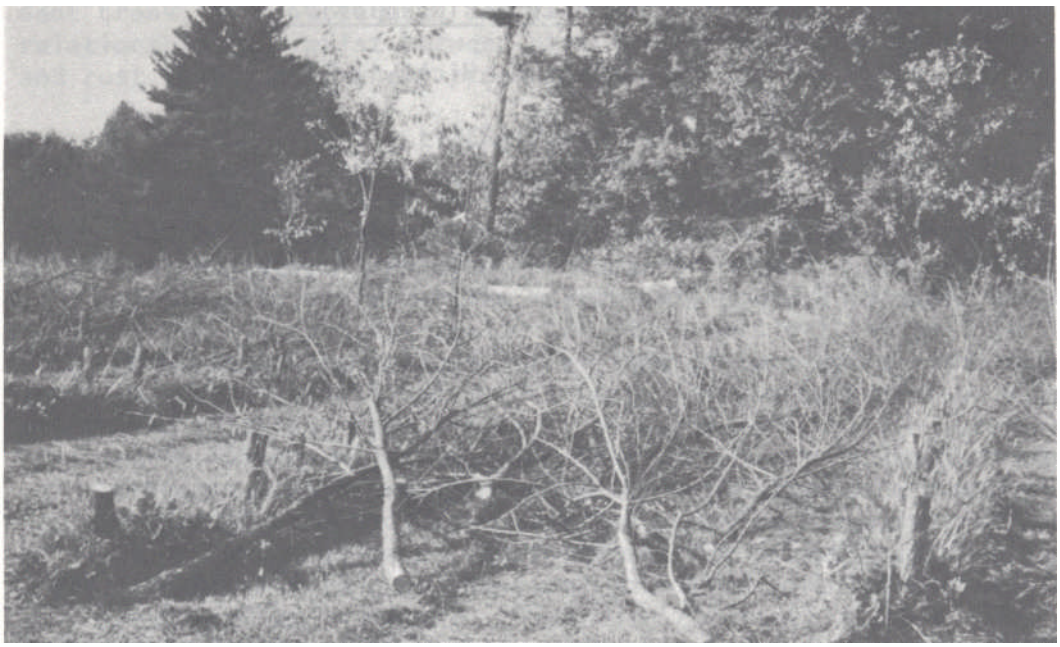

Figure 1.--"Many are called but few are chosen."

All of our trees were inoculated for Dutch Elm Disease with a small chisel invented and made by Joe Demaradzki. The chisel makes a small v-shaped notch in the stem. This chisel conveniently holds the inoculum in a trough in its upper surface. With this tool you can move quickly from tree to tree, inoculating each by just striking the handle-end of the chisel with the heel of one hand, while guiding the chisel tip with the other hand.

We always wait at least one full year after transplanting before we start to do our inoculation work, because it is extremely difficult to get the Dutch Elm Disease to "take" right after transplanting of the elm.

What we do not know is what we might have found if we had worked simultaneously with 35,000 nursery-grown, non-irradiated elm seedlings; 35,000 is approximately the number of trees we inoculated in this program from our 6½ million irradiated elm seed. Would we have ended up with several hundred resistant elms <u>without</u> irradiation treatments? That is why I say this is not a bonafide experiment; it was an attempt to obtain resistant material. Thank you, friends.