VEGETABLE SEED TREATMENT

J. C. WALKER University of Wisconsin

INTRODUCTION

The treatment of agricultural seeds as a control of disease began as early as the seventeenth century. Apparently the first chemical used was sodium chloride. Arsenic and mercuric chloride were suggested in 1756 (5), but neither was adopted. At that time attention was being directed almost entirely to wheat and to control of bunt (*Tilletia tritici* (Bjerk.) Wint.). Lime water was used with some success in 1755 (67). Copper sulfate solution as a seed drench was reported in 1761 (63), and its use increased gradually for a century. Prevost (61) in 1807 demonstrated that copper prevents germination of spores of the bunt fungus. Addition of lime to copper sulfate to reduce toxicity of the latter to seed had come into practice by 1862 (12). By 1889 wheat seed treatment with copper sulfate had become quite common in England (58).

While control of bunt was fairly satisfactory it became recognized that loose smut (Ustilago tritici (Pers.) Rostr.) is not affected by the copper-sulfate treatment (58). Suspecting that the latter fungus was more deep-seated in the seed than the bunt organism, Jensen (42), in Denmark, devised the hot water treatment, described in 1888, which is still used for loose smut of barley (Ustilago nuda (Jens.) K. & S.) and loose smut of wheat. The European work was confirmed and extended in United States (8, 44, 45, 46, 47). In 1895 Guether (33) in Germany was the first to report the treatment of grain seed with formaldehyde solution. Bolley (11) in 1897 reported favorable results with the latter chemical in United States, particularly on oats for the control of loose smut (Ustilago avenae (Pers.) Ien.) and covered smut (Ustilago levis (K. & S.) By the turn of the twentieth century, methods had been Magn.). worked out for the control of several seed-borne pathogens of cereals. Little attention had been given to methods of treatment for vegetable seeds.

TYPES OF SEED TREATMENT

At present there are recognized three distinct types of seed treatment. They are designated herein as seed disinfestation, seed disinfection and seed protection. The objectives of each will be described briefly.

Seed Disinfestation. Seeds are commonly contaminated on the surface by spores or other forms of pathogenic organisms, without being penetrated or infected. A majority of the grain-smut organisms bear such a relation to the seeds of their respective hosts. Chemical dips or soaks are eminently successful as seed disinfestants. The early success of copper sulfate against bunt was as a disinfestant. Numerous pathogens which contaminate vegetable seeds are controlled by chemical means.

Seed Disinfection. The failure of copper sulfate to control smut of wheat not only led to exploration for a better treatment but it also gave a clue to the life cycle of the loose-smut organism. When Jensen reasoned that heat might be more penetrating than chemical ions and less phytotoxic he suggested for the first time the principle of seed disinfection. As used here, then, disinfection refers to ridding the seed of a pathogen which has penetrated, infected and thus has become established within the seed. Hot water is still the standard treatment for loose smut of wheat and loose smut of barley, and it has been adapted to use with a number of vegetable seeds. Some chemicals are seed disinfectants as well as disinfestants.

Seed Protection. Seed protection refers to the treatment of seed, usually with chemicals, neither to kill organisms on the surface nor to kill organisms which have penetrated beneath the surface. Rather, it is designed to protect the seed and the young seedlings from organisms in the soil which might otherwise cause decay of the seed before germination or parasitize the seedling at or immediately following germination. The first successful use of a seed protectant was by Thaxter (66) in 1890 in connection with onion smut (Urocystis cepulae Frost). He had determined that the smut spores infest the soil and that the seedling is susceptible for a short time after germination of the seed. He reasoned that a chemical in close proximity to the seed in the soil might protect the young seedling during this short susceptible period. After trying several materials he found a mixture of sulfur and lime to be the most effective. This treatment was used for a time and was later supplanted by the formaldehyde-drip treatment reported in 1900 (62). Chemicals used with this objective are known as "seed protectants".

MAJOR TRENDS IN VEGETABLE SEED TREATMENT

As already indicated, little attention was paid to vegetable seed treatment before 1900. The use of sulfur and lime and later of formaldehyde as protectants applied to the soil with the seed to control onion smut has been mentioned. In the last decade of the nineteenth century the treatment of potato tubers with mercuric chloride and with formaldehyde came into use (4, 9, 10). The recognition of seed-borne pathogens of vegetables gradually directed attention to seed treatment.

In 1904 Harding, Stewart and Prucha (35) concluded that cabbage seed is commonly contaminated with the black-rot organism (Xanthomonas campestris (Pam.) Dowson). They suggested soaking seed in 1-1,000 mercuric chloride solution for 15 minutes or in formalin (40% formaldehyde), one pound to 30 gallons. The appearance of black leg of cabbage (Phoma lingam (Fr.) Desm.) in United States was recorded in 1910 (52). Henderson (36) demonstrated that the causal organism is seed-borne. While seed treatment with mercuric chloride or formaldehyde served to control the two diseases, growers and investigators became aware that epidemics might arise in spite of treatment. It was shown later that Phoma lingam (69) and Xanthomonas campestris (19) are both borne to some extent within the seed. Mercuric chloride and formaldehyde serve well as seed disinfestants but not as seed disinfectants. Another fact peculiarly patent to vegetable growing, when plants are often started in crowded plant beds, is that a very rare escape of the pathogen from the effect of seed treatment is often sufficient to start an epidemic in the seed bed when favorable environment prevails. This was shown to be a very important phase in the disease cycle of cabbage black leg and black rot, and it applies in much the same way to diseases of many other vegetables. The need for improved seed treatment of cabbage led to exploration of other methods. Dry heat was not practicable (69). Hot water treatment, suggested in 1919 (56), was worked on further for black leg (70). It was reported to be effective for black leg and black rot in 1924 (16, 18). It has since that time remained the standard disinfectant for seeds of cabbage and other crucifers.

The chief seed-borne pathogens of common bean (Thaseolus vulgaris L.) are the bacterial blight organisms (Xanthomonas phaseoli (Smith) Dowson: Pseudomonas phaseolicola (Burkholder) Dowson), the bacterial wilt organism (Corynebacterium flaccumfaciens (Hedges) Dowson) and the anthracnose organism (Colletotrichum lindemuthianum (Sacc. & Magn.) Briosi & Cav.). Extensive experiments were reported (55) in 1917 to devise means of disinfecting bean seeds. The results were entirely negative. Attempts of others since have failed to yield a satisfactory treatment for bean. The same holds true for the bacterial blight of lima bean (Pseudomonas syringae van Hall) and for the important seedborne pathogens of pea (Mycosphaerella pinodes Berk, & Blox.; Ascochyta pinodella L. K. Jones; Ascochyta pisi Lib.; Pseudomonas pisi (Sackett) Dowson). Several of the newer protectant dusts partially disinfest pea seed of surface-borne Ascochyta (51), but for complete control seed disinfection is required, and no suitable means has been forthcoming.

Beet-seed treatment has been the subject of much investigation both in United States and in Europe since early in this century. The most common seed-borne pathogen is Phoma betae (Oud.) Frank ; others are Peronospora schachtii Fckl., Cercospora beticola Sacc. and Pseudomonas aptata (N. A. Brown & Jamieson) Stapp. Coons and Stewart (22) in 1927 showed that hot water at 60°C. for 10 minutes disinfects seed infected with P. betae., but the nature of the protective pulpy tissue surrounding the seeds in the seed ball made liquid treatments unsatisfactory and impractical. Shearing of seed clusters to free individual seeds is a relatively recent process which obviates the difficulty. Leach (49) found that Ceresan (2% ethyl mercury chloride), New Improved Ceresan (5% ethyl mercury phosphate), Arasan (tetramethylthiuramdisulfide) and dichloro-naphthoquinone controlled seedling infection by Phoma but did not completely eliminate the fungus. Ark and Leach (3) found that New Improved Ceresan, used as a dip or dust, and Arasan as a dust, were satisfactory disinfectants for Phytomonas aptata. In most sugar-beet and table-beet experiments the emphasis has shifted in recent years toward protection against damping-off fungi in the soil.

The work of Gardner (28) and that of Carsner (15) established the seed-borne nature of the organism of cucurbit anthracnose (Colletotrichum lagenarium (Pass.) Ell. & Halst.) and of cucumber angular leaf spot (Pseudomonas lachrymans (E. F. Sm. & Bryan) Farraris), respectively. Gilbert and Gardner (29, 31) developed seed disinfestation control by the use of mercuric-chloride solution, a practice still in vogue. The celery-blight organism (Septoria apiigraveolentis Dorogin) was shown to infect celery seeds, and the hot-water treatment devised in 1921 (48) has been used widely. Gardner and Kendrick (30) showed that the causal organism of bacterial spot of tomato (Xanthomonas vesicatoria (Doidge) Dowson) is seed-borne, and they worked out a mercuric-chloride treatment reported in 1921. Formaldehyde solution with or without previous immersion of the seed in alcohol was also effective as a seed disinfestant. More recent work has brought forth New Improved Ceresan (5% ethyl mercury phosphate), applied as a dust or a drench as a seed disinfestant (24, 68). Development of bacterial canker as a major disease in many areas in the United States following 1926 (13) indicated that the organism (Corynebacterium michiganense (E. F. Sm.) Jensen) is commonly seed-borne. Chemical treatments failed to rid the seed of the organism. Blood (6) in 1933 reported that if seed was allowed to remain in fermenting pulp for 72 hours it became disinfected. He attributed this to the increasing acidity of the pulp. He worked out an acetic-acid treatment which is effective if applied to freshly extracted seed (7). At the moment inspection and certification of seed fields together with application of New Improved Ceresan (5% ethyl mercury phosphate) as a dip or as a dust disinfestant are the chief measures applied to tomato (24, 28). Verticillium spp. and Fusarium oxysporum f. lycopersici (Sacc.) Snyder & Hansen and Pellicularia filamentosa (Pat.) Rogers are also occasionally seed-borne. Hot water (30 minutes at 50° C.) is the only disinfectant so far devised (5a.) Pepper and eggplant seed may be considered in the same category as that of tomato.

From about 1900 to 1925 major attention was directed toward treatment of vegetable seeds to disinfest or disinfect them. Fairly satisfactory but usually cumbersome methods were devoloped for cabbage, tomato, celery and cucurbits. Little or no progress was made with fleshy seeds, such as pea, lima bean and bean. Up to 1925 seed protection received only minor attention, the notable example being the formaldehyde-drip control of onion smut. It was from 1925 to 1932 that the field of vegetable seed protectants was opened up (17, 22, 34, 37, 43, 57).

Coons and Stewart (22) reported in 1927 on an extensive series of treatment experiments with sugar-beet seeds. They emphasized that the major damage to seedlings in the period immediately following germination is not usually due to Phoma betae, which commonly infects beet seeds, but to soil-inhabiting fungi. They pointed out that complete disinfection of the seed by hot water at 60° C. for 10 minutes has no effect upon soil-inhabiting damping-off fungi. They found that mercury and copper compounds applied as dusts has value as seed protectants. Clayton (17) in 1928 showed marked improvement in stands with a number of vegetables when seed was treated with organic mercury compounds. Semesan, Uspulun and Bayer Dipdust, applied in either liquid or dust form. Jones (43) reported in 1931 that organic mercury dusts (Uspulun and Semesan) are effective protectants of pea seed and are particularly useful in wet soil in which soil inhabitants cause pre-emergence damping off. Horsfall began to explore the value of other seed protectants on various vegetables shortly before 1930. Poor adherence of copper sulfate powder was a decided handicap. Cuprous oxide was found to be a cheap, effective protectant for peas and tomatoes. First used as red cuprous oxide and later as the more finely divided yellow oxide, it came into extensive use on peas. Zinc oxide was also found to be an effective protectant and was used extensively on spinach (20, 21, 40). Several disadvantages of cuprous oxide and zinc oxide soon appeared. Both increased the friction between seeds in the planting operation, as was found earlier with copper carbonate as a wheat-seed-treatment dust, and it became necessary to add graphite to lubricate the seed in machine planting. Cuprous oxide was found to cause some injury, and it was particularly injurious to crucifer seeds. Zinc oxide caused injury on peas (38, 39, 41, 59).

The search for less phytotoxic materials continued and was stimulated by the shortage of mercury and copper which developed with the onset of World War II. Non-metallic organics were explored, and several new seed protectants were uncovered. The first of these was tetrachloro-para-benzoquinone, later marketed as Spergon. It was reported as an effective protective on lima beans in 1940 (23) and on peas in 1942 (26), and was rapidly adopted as a dust treatment for both. In 1943 tetramethylthiuramdisulfide (Arasan) and ferric dimethyldithiocarbamate (Fermate) were reported as effective protectants in several parts of the United States (25, 32, 53, 60, 65).

Arasan has been adopted widely as a protectant on sweet corn, peas, onion (pelleted), lima beans and other vegetables. To a large extent Spergon and Arasan have now replaced cuprous oxide and mercury dusts on vegetable seeds. Spergon has been found, however, to be inferior on beet and spinach, and not as consistently effective on sweet corn as Arasan. Other organic materials coming into use as seed protectants are 2, 3 dichlor 1, 4 naphthoquihone (Phygon) and zinc trichlorophenate (Dow 9B).

In the development and application of seed protectants certain problems arise which are of less importance with seed disinfestants and seed disinfectants. In the last two types of treatment the environment at the time of treatment can be controlled, and the chief variable is the physiological status of the seed sample concerned. In the case of seed protectants the environment is usually beyond control and may be expected to vary widely as to soil type, soil reaction, soil moisture, soil temperature and soil flora. Buchholtz (14) has shown, for instance, that on sugar beets in Iowa, *Pythium debaryanum* Hesse is the major soil fungus which attacks seeds and young seedlings. It is quite successfully controlled by seed protectants. However, another damping-off fungus, *Aphanomyces cochlioides* Drechsler, begins to attack seedlings about the time when Pythium injury ceases, and this fungus is not controlled by seed treatment.

Since treatment may be highly successful in one instance and of much less value in another, uniform procedures for general use as to material and dosage are more difficult to define for protectants than for disinfestants and disinfectants. In recognition of this fact, a special committee of the American Phytopathological Society, under the leadership of Dr. H. T. Cook, undertook on a national scale a series of seed-protectant-treatment tests on standard vegetables with promising fungicide dosage rates. These trials were extended over a period of five years beginning in 1941. In this plan the same lot of seed was treated and distributed to various locations in the United States and Canada for trial. Thus the seed variable was eliminated and attention was focused upon variables in the soil environment which affected the usefulness of the fungicide concerned. In this way some fungicides were eliminated or placed in secondary positions for certain seeds, and better evaluation of dosages was secured. Certain of these results are now available in published form (1, 2). They have comprised a base upon which recommendations were made during the World War Period.

It is indeed surprising that such general agreement was reached in these extensive cooperative experiments with seed protectants, when trials were conducted under such a wide range of conditions. While they leave some important questions unanswered, they provide a tentative basis for application and a point of departure for further exploration and research. Crucifer seeds are sensitive to copper compounds, while beets and spinach are commonly injured by Spergon. The latter is eminently successful as a protectant on peas and lima beans. Under certain conditions, Semesan Jr. (1% ethyl mercury phosphate) and Spergon are eminently successful protectants for sweet corn. The former has the additional advantage of being a disinfectant when Diplodia spp. are concerned. To injured seed it is, however, phytotoxic under some conditions. Since Arasan has been shown to be a safer protectant and a fairly efficient disinfectant, it is rapidly becoming the most widely used fungicide for corn seed. Leach and Smith (50), in a critical study of seed protectants on peas, secured beneficial results with soil infested by Pythium ultimum Trow. but not with soil infested with Pellicularia filimentosa (Pat.) Rogers (Rhizoctonia solani Kühn). Semesan, cuprous oxide, Spergon, New Improved Ceresan and Arasan were very effective, the first two being the most so. However, when germination was delayed by low soil temperature cuprous oxide and New Improved Ceresan (5% ethyl mercury phosphate) were injurious to the extent that germination was retarded and depression in yield resulted. Taylor and Rupert (64) showed that Arasan is distinctly superior to Spergon and cuprous oxide in the duration of the protective period, and thus is most effective in reducing post-emergence damping off. Their tests were made with spinach planted in soil inoculated with Pythium ultimum.

Foster (27) offered an explanation for the fact that copper compounds tend to be injurious to seeds of crucifers and peas and sometimes to cucumbers, while they are not so to seeds of beet, spinach and solanaceous vegetables. In fact, he showed that copper sulfate and cuprous oxide in certain concentrations actually stimulate the latter group. He suggested that the differential effect of copper on seeds may be due in part to its effect upon the enzyme system in the seed. One of the systems present in pea and cucumber seeds requires the presence of sulfhydryl compounds, and enzymes of this type are inhibited by copper. In beets and spinach, on the contrary, the enzyme system contains tyrosinase which requires the presence of copper ions for its activity.

CURRENT RECOMMENDATIONS

In the table on page 597 are listed some of the currently accepted methods of seed treatment which are worked out for disinfection, disinfestation and protection, respectively.

FUTURE NEEDS IN SEED TREATMENT RESEARCH

Exploration for new treatment materials since 1933 has yielded a small number of compounds vastly superior to those previously available as protectants for vegetable seeds. In this period we have seen the fungicides containing the heavy metals (*e.g.*, cuprous oxide and zinc oxide) rise and recede in favor largely because of their phytotoxicity, while organic compounds of sulfur and chlorine have come to the front, *e.g.*, Spergon (tetrachloro-para-benzoquinone), Arasan (tetramethylthiuramdisulfide), Phygon (dichlornapthoquinone). None of the last group is without limitation. Research in the nature of the interaction of each with host, pathogen and environment is the greatest present need. Exploration for better compounds may thereby become less empirical.

In the same period of nearly two decades methods of seed disinfection have changed little. Only in the case of sweet corn do seed protectants serve also as seed disinfectants. In the case of seeds of crucifers, celery, tomato, pepper, eggplant and beet we have nothing to supplant hot-water treatment. Its effectiveness is offset by its clumsiness and by a narrow margin of safety. Most seed producers and seed merchants, consequently, avoid it. McWhorter and Miller (54) have explored the possibility of heated vapor as a substitute for the hot-water soak. With fleshy seeds (beans, lima bean, and pea) no advance has been made. Here, certainly, plant pathology has been at a standstill for a quarter century and shows little sign of moving off dead center. In the meantime the pea

Vegetable	Disinfectant	Disinfectant ^a	Protectanta
Beet	Hot water, 60 % 10 min.	New Improved Ceresan dust, ^b 0.5%	Arasan, ^c 0.5%
Cabbage, cauli- flower and other cruci- fers	Hot water, 50° C., 30 min.	None	Arasan, 0.5%
Carrot	Hot water 52° C., 10 min.	None	Arasan, 0.75%
Celery	Hot water, 48° C., 30 min	None	None
Cucumber, and other cucur- bits	None	Mercuric chloride, 1–1000, 5 min.,	Spergon, ^d 0.3% or Arasan, 0.3%
Lettuce Lima bean Onion	None None None	rinse None None None	Spergon, 2.0% Spergon, 0.1% For damping off: Arasan, 0.5% For smut: 1-64 for- maldehyde, 1 gal. per 350 feet of row; or Arasan, 25% pelleted on seed with Metho- cel sticker
Pea	None	None	Spergon, 0.2%;
Spinach Sweet corn Tomato, pepper, eggplant	None None Hot water, 50° C., 30 min.	None Arasan, 0.3% New Improved Ceresan dust, 0.5%	0.2% Arasan, 1% Arasan, 0.3% New Improved Ceresan, dust, 0.5%

The dosages of dusts are given as percentages of the weight of seed.
5% ethyl mercury phosphate.
50% tetramethylthiuramdisulfide.

^d Tetrachloroparabenzoquinone.

crop is at the mercy of Ascochyta blights and bacterial blight, and the bean crop is continually pest-ridden with bacterial blights. Production of disease-free seed of these crops in any sizeable amounts is far from reality.

In fact, the great emphasis on seed protectants has, if anything, tended to deemphasize seed disinfection and its importance. With the wholesale application of dust protectants (usually worthless as

disinfectants) to seeds at the source of distribution, there is a growing tendency for layman consumers to assume that all seedborne pathogens have been cared for. Moreover, there is a striking lack of warning in current extension literature against this illusion. Seed salesmen are known to advise customers that hotwater treatment is not necessary for cabbage if it is dusted with Semesan, and that cucumber seed need not be treated with mercuric chloride for angular leaf spot if dusted with Semesan, etc. What is the result? Nearly every year in the last several the writer has encountered epidemics causing losses running into thousands or tens of thousands of dollars due to pathogens traceable directly to introduction on infected seed.

What is the answer? If plant pathology is to meet this important gap in disease control, one or both of two developments must come about. First, adequately supported research must be directed toward the improvement of methods and materials for seed disinfection which are sufficiently safe and effective so that seed producers and distributors can be expected to use them at the source. Second, research must be directed toward the development of bio-assay methods whereby presence of important plant pathogens can be detected with certainty. The latter is a tool much needed by seed producers and examiners and is one which is essential to an adequate system of regulation and certification. Until research shows definite progress in these directions vegetable crop losses from pathogens which infect the seeds may be expected to continue unabated.

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