Symptomatology and transmissibility of the mosaic disease of great northern bean
by Bernice Helen Norris

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Montana State University

Abstract:
From the foregoing discussion, it would appear that the viruses of bean are of different types and possibly of different intensities, as their reactions on the plants of the same species of Phaseolus, was quite diversified. Some types proved more virulent than others to the same selections of the Great Northern bean. The bean may have built up a resistance to one type while it is susceptible to all other types, or it may have a resistance to all types.

The sterile pot label and swab method of inoculation of the plant to be infected, is one of the surest methods. The workers' hands do not come in contact with the diseased plant or the healthy plant therefore there is little chance of careless transfer of the disease. The leaves can be gently rubbed or injured with the swab, so a, technique of inoculating can be easily gained.

The mosaic disease of bean may vary with environmental conditions, as the winter inoculations were not as successful as the spring inoculations? Although the yellow veinbanding is a symptom of spray injury, it appears in the field in isolated cases. This symptom could not be transmitted to other plants through inoculation processes. It is probably an abnormality, the cause of which is not determined.

The clearing of the veins is an early symptom of mosaoc of Solan-aceae, sugar beet, and aster, but it is not transmitted from bean plant to bean plant? This clearing of the veins is either the symptom of an abnormal form of bean plant or of a weakened form of virus, that is not in a great enough quantity or in a virulent enough stage to produce mosaic symptoms through inoculation.

The different types of mosaic as demonstrated by the different patterns of the symptoms, can be either of four different pure types or as compound symptoms? The compound symptoms of the disease were more virulent as a group than the pure symptoms of the disease. The virulence of the complex symptoms, were retained longer than the pure symptoms in infected juice.

The virus is not inactivated through drying for five months as dried material is as virulent as the fresh material.

The germination of the seeds was not low due to the presence of virus within the seed, but rather because of the imperviousness of the seed coat to water. A germination test is no indication of the presence of virus within the seed.

The seeds from virus plants, when allowed to remain in storage for five years, produced plants which showed no symptoms of disease.

The environment of the greenhouse tends to bring out a degenerate form of virus in the secondary growth of the diseased plant, characterized by a slight rugosity and fine uniform mottling.

The Ring spot of alfalfa transfered to the bean plants in a slightly different type symptom than any of

the symptoms of the mosaic disease of bean, and very much different from the inoculum, It would not transfer to the tobacco or to the sweet clover.

The tobacco appeared to be susceptible to only one type out of six (6) of the viruses of the bean. It produced yellow lesions at the base of each injured hair for two types but only one multiplied enough in the tobacco plant to be transfered back to the bean. The symptoms gradually disappeared in the tobacco. The disease probably transfered over to the tobacco but was unable to multiply there as it should, and soon died out* The optimum time for successful inoculations for Phaseolus is in the first compound, leaf stage, just as the second compound leaf appears and before it has unfolded. This is true of artificial as well as natural infection. Symptoms of mosaic usually appeared within fifteen (15) days after inoculation.

The disease may be transfered in the earlier stages through rough handling in the field during the processes of roguing. Great care should be exercised in the field especially if the plants are young.

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A THESIS

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INTRODUCTION

"Many of the common diseases both of plants and of animals are attributable to agents called filter passing viruses." Caldwell (8).

The filterability of the principle causing bean mosaic, is an accepted fact. The causative agent is debatable, and little understood. It is know that the disease will transfer from plant to plant in the same species, but the scope of the field of transfer is not known, even within the Leguminosae.

The tobacco viroses have been found to be of more than one kind. This has not been ascertained of the mosaic of Phaseolus.

The investigations reported in this paper were carried on to determine the transmissibility of the mosaic of Phaseolus vulgaris L. to other members of the same species, as well as to other members of the Leguminosae, Solanaceae, Compositae, and Oxalidaceae. Further study was made of the symptoms of the disease in comparing them with the symptoms of a degenerate nature. Symptomatology was studied in an attempt to discover if the mosaic of bean was of more than one kind. An attempt was made to inoculate the ring spot of the alfalfa to the bean, as well as to other plants.

Studies were made of the longevity of the virus in the seed and in the extract from dried mosaic material.

The percentage of the germination of the bean seed was very low. A study was made to determine the factor causing this low germination and its relation to virus contained in the seed.

HISTORICAL

Early History

According to Nelson (35), Mayer in 1886 discovered an infectious disease of tobacco, which he named Mosaikkrankheit; Iwanowski in 1894 by using porcelain bacterial filters demonstrated the presence of the filterable virus as the causative agent, thereby increasing the interest in, as well as the knowledge of the disease, and Beijerinck in 1899, confirmed the filterability of the disease.

Allard (2) demonstrated that the causative organism was not an oxidizing enzyme as alleged by Woods (48) as he could destroy either one without harming the other.

Transmission

C. P. Clinton (10) demonstrated the transmissability of the tomato virus to the tobacco and from the tobacco to the tomato. Jagger (23) demonstrated that infection did not occur through diseased soil. Doolittle (12) incriminated the cucumber beetle with the transmission of the infectious mosaic disease of cucumber. Stewart and Reddick (46) successfully transfered the mosaic to healthy plants by rubbing the leaves of the young bean seedlings with diseased leaves. Jagger (24) successfully demonstrated the non-specificity of the cucumber virus by transmitting it to closely related families of the order Campanula Les.

and to one species each of Compositae and Lobeliaceae. He reinoculated the cucumber from the resulting diseased plants. However, Doolittle (13) was unsuccessful in his attempts to transfer the disease from other plants to the cucumber or with the one exception of Martynia, from a cucumber to other plants. McClintock (30) demonstrated that Lima Bean mosaic was transmitted through the seed, as he obtained diseased plants on soil that had not previously been cropped with beans. Melhus (32) found mosaic to carry over the winter, in the wild perenial Solanum. It was transferred to the healthy commercial solanaceous plants in the spring by insects. Chardon (8) proved the ability of the Aphis maides in transmitting the mosaic of sugar cane to healthy plants. Doolittle (13) found the juice from mosaic cucumber to be infective, for only twenty-four to forty-eight hours; while Fernow (17) found the juice of mosaic tobacco to still be infective after several years when kept in a jar in the greenhouse. Pierce and Hungerford (36) found the mosaic to remain active in bean seed for thirty years, although Duggar (14) through protein tests on the virus pointed out the possibility of the adsorption and inactivation of the virus through storage protein. He found the protein in the bean seed to be poisonous to the virus in concentrated amounts, but not in the amount found in the seed.

Fajardo (16) studied the mode of transmission of the bean virus through the seed. He used surface sterilization of the seeds, both with and without the seed coats and found that the mosaic had not been killed. No infection was obtained when seeds were soaked in diseased

juice. As he found the greatest percentage of spread to be through the seed, he concluded that the virus was concentrated in the embryo of the seed.

Although McClintock (30) demonstrated that Lima Bean mosaic is primarily transmitted through the seed, the disease spreads after it gets into a field. Studies were made on the mode of transmission and on the possibility of the viruses being of different strains. McClintock (30) demonstrated that mosaic is held over in the seed and infection can start from there. Jagger (23) demonstrated that the foliage and not the roots had to be in contact and injured for the transference of the filterable virus of cucumber mosaic. Doolittle (12) attempted several types of inoculations on cucumber. From insect transference he gained one hundred per cent infection, from direct inoculation of torn diseased plant to injured healthy, he gained seventy-nine per cent infection. Stewart and Reddick (46) successfully inoculated plants by rubbing healthy bean seedlings with crushed mosaic diseased leaves. In 1919 they further ascertained that infection is more sure when the plants are in the first leaf stage. Fernow (17) crushed the fresh mosaic material in his hand and rubbed the underside of healthy juvenile leaves. This necessitated the washing of the hands after each inoculation. Rubber gloves were then used, but proved unsatisfactory. Two pairs of forceps were used as they could be flamed between each inoculation. These proved the most successful. Grafting

and splints he also found to be unsuccessful. Pierce and Hungerford (37) pricked through the inoculum, but found this inferior to the method of rubbing the juvenile leaves with the inoculum. Burnett and Jones (6) supported the juvenile leaves with a sterile pot label while they rubbed the top with a piece of sterilized cotton that had been previously dipped in the infected juice. The pot label and absorbent cotton was changed for each inoculation. The hands were thoroughly washed after each inoculation. A modification of the method and the one used in this work was described by Jones (25) in which he replaced the absorbent cotton with small swabs.

Symptoms

Various studies have been made of the virus disease of bean to determine if it is of one strain or of many strains. Fernow (17) found that mosaic symptoms varied with the environmental conditions. Burkholder and muller (7) through disease studies found bean plants to become misshapen and spotted due to hereditary characteristics rather than to disease. This abnormality he found did not reduce the crop yield and was not infectious. McKinnery (31) found three types of mosaic in tobacco, a yellow, a light green, and a mild light green. Through careful inoculation studies, he was able to intensify the three symptoms in different plants.

Causative Agent

The mosaics of plants have only one accepted principle, the infectiousness of the disease. The causative organism of the mosaic diseases

is not known, though four theories have been generally accepted as possibilities. 1. The bacterial theory; 2. The enzymatic theory; 3. The virus theory; and 4. The protozoan theory.

According to Heald (18) Mayer first supported the bacterial theory of the mosaic theory. In (4), Boncquet found a nitrate reducing streptococcus in mosaic infested tobacco. In 1917 (5), he believed that he had found the causative agent of the curly top of sugar beets. This he named Bacillus morulans. Dickson (11) found bacterial like bodies. Nelson (33) isolated a minute coccus form, that usually occurred singly or in pairs or chains, from mosaic plants. Nelson (34) secured negative results from healthy plants, while mosaic plants gave micrococcus bodies. He studied the progeny of mosaic plants and ascertained that the seedlings have an uneven distribution of the mosaic corresponding to the amount contained in the original plants. He believed them to be related to the Rickettsias. He was unsuccessful in producing artificial infection from these cultured organisms. Takahashi and Rawlins (47) used stream double refraction through a microscope for determining the polarization of ultra-microscopic colloidal solutions. They found that the mosaic juice contained rod shaped colloidal particles while the healthy plant juice contained circular particles.

The enzymic theory was first proposed by Woods in 1899 (48). He suggested an abnormal and extensive development of an oxidizing enzyme. Allard (2) demonstrated the impossibility of the oxidizing enzyme being the causative agent when he destroyed the enzyme with hydrogen peroxide,

but did not destroy the virulence of the virus mosaic.

The enzyme could be treated with alcohol and recovered while the infective principle was destroyed.

According to Heald (18), Beijerinck (3) in 1899 used the term "Contagium vivum fluidum" to describe the causative agent, as it would pass through Berkefeld and Chamberlain filters. Allard (2) and Doolittle (13) determined that the infective principle would pass through a Berkefeld filter, but not through a Chamberlain filter.

Holmes (19,20) and Price (38) found a correlation between the concentrations of the virus and the number of lesions produced by tobacco mosaic. Holmes (21) found a relatively definite path of infection of the disease from the focal points of infection. Price (39) found the Nicotiana sylvestris to develop a form of immunity to the disease of ring spot. This immunity is transfered through the cuttings. The virus is contained in the plant, but develops no symptoms. Lachey (27) found that the mosaic of the sugar beet can be attenuated when passed through tomato, squash, Nicotiana rustina, watermelon, resistant varieties of sugar beets, and spinach. It can be reactivated to almost its original virulence when passed through the highly susceptible chickweed.

Sheffield and Smith (42) found bodies in the hairs of mosaic plants which they designated as x bodies. These bodies varied in different plants.

MATERIALS AND METHODS

The studies reported in this paper were carried on in the Experiment Station greenhouse at Montana State College, Bozeman, Montana, during the year 1932-33, under the direction and help of Professor H. E. Morris. The Great Northern bean seed was obtained for the work from the Huntley Branch Station at Huntley, Montana; from Ralph Mercer, County Agent at Forsyth, Montana; and from E. G. Brashear, of Fromberg, Montana. The seeds used in the longevity experiment were from the 1927 Huntley Branch Station selections and were contributed for the work by Dr. P. A. Young, of the Experiment Station of Montana State College. The tobacco and clover were grown in the greenhouse of the Experiment Station.

The mosaic diseased material was obtained from bean plants grown in the Experiment Station greenhouse, from the field run bean selections, and from the Ralph Mercer Idaho Certified bean stock.

The dried material was obtained from plants grown at the Huntley Branch Station at Huntley, during the fall of 1932.

The fresh mosaic leaves were macerated on a sterilized pot label with a small, sterile cotton swab. The macerated material was then transfered to the healthy plants, in the first to the fourth compound leaf stage. A different sterile swab and pot label was used for each inoculation. The healthy leaves were rubbed with the swab containing the inoculum until there was evident injury. The plants used for the inoculum were those having the characteristic mottling. The ring spot

mosaic was obtained from alfalfa plants grown in the Experiment Station greenhouse. Tests were made to determine the optimum stage for the inoculations. Different types of mosaic characterized by mosaic-like patterns were used to determine if they were representative of different strains of the mosaic of bean.

The dried leaves were pulverized in a sterilized mortar with a sterile pestle. An equal quantity of distilled water was added and again ground. This was used as the inoculum. The mortar and pestle were scoured with Peets mechanic soap and water between each pulverization. The hands of the worker were also washed with the soap between each type pulverization.

The juice from healthy leaves was inoculated into healthy plants for check purposes.

The 1927 selections from the Huntley Branch Station of Great Northern and Red Mexican bean seeds were dusted with Cuprous oxide and incubated at 28° C. in sterile damping chambers. As they germinated they were placed in pots in the greenhouse. These plants were watched for signs of mosaic.

Specimen I. Ten Red Mexican bean seeds from normal plants.

Specimen II. Ten Great Northern bean seeds from plants showing a prominent mosaic in the leaves though little stunting of the plant.

Specimen III. Ten Great Northern bean seeds from mosaic plants.

Specimen IV. Ten Red Mexican bean seeds from mosaic plants.

Specimen V. Ten Great Northern bean seeds from mosaic plants.

Specimen VI. Nine Red Mexican bean seeds from plants showing severe mosaic.

Forty seeds of the selection number sixty-nine made in nineteen-hundred and thirty two were tested in sterile damp chambers in the incubator for hard seeds. These seeds were dusted with cuprous oxide. Twenty of the seeds were incubated without pricking, while twenty of the seeds were pricked with a size 10 sewing needle, through the seed coat before incubation. Twenty-five of the ungerminated seeds in the benches were pricked.

The indefinite growth of all plants was trimmed back at frequent intervals with scissors, flamed with an alcohol lamp, between each cutting. This guarded against any transfer of the disease from plant to plant due to the cutting. It produced bushier plants and so concentrated the disease and brought forth symptoms whenever the plant was diseased. It enabled a closer study of the plants as they were not allowed to entwine together. The possibility of the transference of the disease through accidental injury in the individual study of the plants, was in this way avoided.

The possibility of the transfer of the disease in the field through injury and contact was studied by injuring adjoining healthy and diseased plants and allowing them to rub together.

The sterile swab method was used for the plant inoculations in this work.

Following is a classification of the mosaic symptoms observed.

Symptoms:- Following is a classification of the mosaic symptoms observed:

Types of Bean Mosaic used - classified

Symptoms that appeared to be single.

1. Stunting.

Type A. The plant was stunted and of a light green color. The cotyledonous leaves developed small, short, light yellow zig-zag lines.

2. Vein banding. A darkened banding of the main veins of the leaves.

Type B. The leaves displayed a typical vein banding of the principle veins, throughout the whole leaf. The leaves were very unsymmetrical.

Type C. The vein banding displayed a yellow banding, starting with a yellow tip mottling and working inward toward the base.

Type D. The leaves were slightly elongated and very misshapen. The vein banding of dark green worked back from the tip.

Type E. The leaves were broader than they were long, with a darkened vein banding throughout.

3. Rugose. A necrotic spotting of dark and light green throughout the leaf, with a hypertrophy of the leaves between the veins, giving a very rough appearance.

a. Small rugose spotting.

Type F. The leaves were elongated with small angular darker green patches on a lighter green background.

Type G. Dried leaves obtained from the Huntley Experiment Station, 1932, contained very small rounded spotting of darker green on light green.

Type H. The cross veins were all of a light transparent green while the small darkened areas between were very hypertrophied.

b. Large rugose spotting.

Type I. Dried leaves from the Huntley Branch Station, 1932, displayed an enlarged spotting of dark green on light green. The leaves were normal in shape.

Type J. The leaves showed a large dark green spotting on a light background with a hypertrophy of the leaf between the veins. The leaves were of a normal shape.

Type K. The leaves were extremely elongated with the dark green large rugose spotting. A high degree of hypertrophy between the leaves.

Type L. The leaves were broadened with larger dark green spotting.

4. Curling. The leaves were curled, not cupped.

Type M. The leaves were a very uniform dark green throughout.

The symptoms of some of the types appeared to be complex expressions of more than one virus. These types have combined symptoms as shown below.

5. Vein banding and cupping. The leaves had the characteristic darker green vein banding, while the edges of the leaves were cupped downward.

Type N. was of this type.

6. Vein banding and small rugose combined. The leaves were veinbanded in dark green with a very small spotting of dark green with the hypertrophy of the leaf between the veining.

Type O. The leaves were alongated, with dark veinbanding. There was a small dark rugose spotting in the light between the darkened veining with a hypertrophy of the leaf between the veins.

7. Vein banding and large rugose. There was a darkened vein banding with enlarged darkened mosaic.

Type P. Dried leaves from the Huntley Branch Station, 1932. The leaves were elongated with the veinbanding containing enlarged darkened rugose spotting in the enlightened areas.

Type R. The leaves were light green with darker green rugose mottling, combined with a darkened vein banding.

Type S. The leaves were of a normal shape with veinbanding and with an enlarged darkening in the lightened areas.

The Ring spot. The alfalfa leaves of the plants effected, showed a veinbanding of very small, alternating lines of yellow green and normal green, combined with concentric rings of the two colors of green.

EXPERIMENTAL DATA

By "successful", the author means that the plant developed the disease, while by "unsuccessful" the author implies that there was no change in the natural development of the plant due to the inoculum used.

The following results are summarized in Table I and II.

Type A was inoculated into thirteen (13) plants, all of which proved unsuccessful for this inoculum.

Type B was inoculated into four (4) plants. The two (2) resulting successful inoculations were used as inoculating material for four (4) plants, one (1) of which proved successful and it was used as the inoculum for twenty three (23) inoculations, of which eighteen (18) were successful. This type was also used as the inoculum for three (3) tobacco plants and three (3) clover plants--all unsuccessful.

Type C was the inoculum used for nineteen (19) bean plants, one (1) clover plant, and one (1) tobacco plant. Twelve (12) of the bean plants became diseased, while neither the tobacco nor the clover showed any disease symptoms.

Type D was used as the inoculum for seventy (70) bean plants, sixty one (61) of which became diseased.

Type E was the inoculum for seventeen (17) bean plants, seven (7) of which were successful. This inoculum was again used as the infective juice for one (1) tomato, two (2) lambsquarters, and six (6) fan weeds.

TABLE I.

Total inoculations made on Great Northern Bean and other host plants.

Plant Inoculated	Inoculum		Results		
	Type	No.	Postive	Percent	Negative
Great Northern Bean	A	13	-	-	13
	B	31	21	67%	10
	C	19	12	63%	7
	D	70	61	86%	9
	E	17	7	41%	10
	F	274	172	62%	102
	G	29	20	69%	9
	H	15	-	-	15
	I	29	13	45%	16
	J	10	9	90%	1
	K	134	91	67%	43
	L	56	47	83%	9
	M	14	8	57%	6
	N	42	29	69%	13
	O	76	45	59%	31
	P	29	20	68%	9
	R	10	9	90%	1
	S	30	22	73%	8
	F-T-B	15	2	13%	13
	K-T-B	15	10	66%	5
	Ring	29	22	75%	7
Total		957	620	64%	337
Chili	K	10	1	10%	9
Kentucky Wonder	1	5	-	0	5
	2	5	5	100%	-
	3	5	4	80%	1

TABLE II. (Continued)

Plant Inoculated	Inoculum Type	Inoculum No.	Results Postive	Results Percent	Results Negative
Improved Golden	1	5	5	100%	-C
	2	5	-	0	-5
	3	5	4	80%	1
Burpee Stringless	1	5	4	80%	1
	2	5	4	80%	1
	3	5	4	80%	1
Pencil Pod	1	5	5	100%	-
	2	5	5	100%	-
	3	5	3	60%	2
Burpee Brittle Wax	1	5	3	60%	2
	2	5	5	100%	-
	3	5	4	80%	1
Scarlet Wonder	1	5	1	20%	4
	2	5	-	0	5
Sweet Pea	K	3	-	0	3
Clover White Annual	B	3	-	-	3
"	C	3	-	-	3
"	F	3	-	-	3
"	K	3	-	-	3
"	N	1	-	-	1
5 Common - 5 Red	Ring	10	1	10%	9
Tobacco	B	3	-	0	3
	F	3	3	100%	-
	K	3	3	100%	-
	N	1	-	0	1
	Ring	3	-	-	3

TABLE I. (Continued)

Plant Inoculated	Inoculum		Results		
	Type	No.	Postive	Percent	Negative
Tomato	E	1	1	100%	-
	D	1	-	0	1
	Ring	1	1	100%	-
Dahlia	E	1	-	0	1
Oxalis sp.	Ring	6	1	20%	5
Thaspi arvense L.	E	6	-	0	6
	L	6	-	0	6
	Ring	6	-	0	6
Chenopodium album L.	E	2	-	0	2
	Ring	1	-	0	1

TABLE II

Results of inoculation with six types of mosaic in Great Northern Bean plants

Type	Plants Inoc.	Results		Percentage Positive
		Positive	Negative	
Stunting	13	-	13	0
Veinbanding	137	101	36	73%
Rugose Small	318	192	126	60%
Rugose Large	229	160	69	69%
Curling	14	8	6	57%
Complex	187	125	62	69%

Type F was the inoculum for two hundred and forty-four (244) bean plants, three (3) tobacco plants, and three (3) clover plants. Of the bean plants inoculated, one hundred and forty-eight (148) were infected, the tobacco appeared slightly infected, and the clover remained unchanged.

Type G was the inoculum for twenty nine (29) bean plants, twenty (20) of which became diseased.

Type H was inoculated into fifteen (15) bean plants. None became diseased.

Type I was the inoculum for twenty nine (29) plants of which thirteen (13) plants were successful.

Type J was the inoculum for ten (10) bean plants, nine (9) of which became successfully diseased.

Type K was the inoculum for one hundred and thirty-four (134) bean plants, three (3) tobacco plants, and three (3) clover plants. Ninty-one (91) bean plants became diseased. The tobacco appeared to receive a slight infection, while the clover was normal.

Type L was the inoculum for fifty-five (55) bean plants and six (6) fan weeds. Forty-seven (47) bean plants became diseased and none of the fan weeds became affected.

Type M was inoculated into fourteen (14) bean plants, eight (8) of which were successful.

Type N was the inoculum for forty-two (42) plants. Twenty nine (29) of these inoculations were successful. This type was the inoculum

for three (3) tobacco and three (3) clover plants, all of which remained normal.

Type O was the inoculum for seventy-six (76) bean plants, of which forty-five (45) became successfully diseased.

Type P was the inoculum for twenty nine (29) bean plants, twenty (20) of the inoculations being successful.

Type R was the inoculum for ten (10) bean plant inoculations, of which nine (9) produced diseased plants.

Type S was the inoculum for thirty (30) bean plants, twenty two (22) of which became diseased.

Types F and K were reinoculated from the tobacco into fifteen (15) bean plants each. Of the fifteen (15) plants inoculated by Type F, two (2) were diseased, while of the fifteen (15) inoculated with Type K from tobacco to bean, ten (10) were diseased.

The Ring spot was inoculated into twenty nine (29) bean plants, twenty two (22) of which became diseased. It was the inoculum for three (3) tobacco seedlings, ten (10) clover plants, six (6) fan weeds, one (1) lambsquarters, six (6) oxalis, and one (1) tomato plant.

Type F was injured and allowed to rub against three (3) injured healthy bean plants in the second leaf stage only. One (1) became diseased. Type N was allowed to rub against three (3) injured healthy bean plamts in their fifth compound stage only. These all proved negative. Type J was injured and allowed to rub against four (4) injured healthy bean plants in the third leaf stage only. One (1) of the four (4) plants

became diseased.

The tobacco was inoculated with types B, N, F, K, and L of the mosaic disease of bean, and with the ring spot from the alfalfa. Types B and N produced no symptoms on the tobacco plant. Type F showed a slightly lightened mottling on the leaves immediately above the leaves inoculated, after the yellowing at the base of each injured hair had faded. Type K showed a definite yellowing of the leaf at the base of each broken hair infected. This reached its greatest development within five days and then gradually faded. Three (3) tobacco plants were injured without inoculum and three (3) were inoculated with healthy bean plant leaves. The leaves on these plants remained clear. The ring spot of alfalfa produced no disease symptoms on the tobacco plant.

The clovers remained unaffected from all of the inoculations from the diseased bean plants. One (1) clover, the red sweet clover, produced the characteristic symptoms of the ring spot disease from the inoculation from the alfalfa.

The tomato was inoculated with types O and R of bean and with the ring spot of the alfalfa. Type O produced no mottling of the leaves, although a slight curling of the leaves was noticeable. Type R produced a clearing of the veins. The ring spot produced no symptoms.

The three (3) sweet peas inoculated with Type K remained normal, although slightly stunted.

Six (6) varieties of beans were inoculated with five each of the following types.

1. Veinbanding

2. Rugose

3. Curling

The results of these inoculations are recorded in Table I.

The symptoms of the disease are not distinguishable until after the compound leaves appear, as they may be masked in the simple leaves and not appear until this later stage is reached. To inoculate in the cotyledonous stage is therefore, very uncertain, as the results are never definitely positive. When the first compound leaves have unfolded there is a better chance of determining the diseased from the healthy plants because the symptoms are becoming evident. The symptoms appeared plainly on the plants when inoculated in the primary compound leaf stage. The second compound leaves show more definitely the symptoms of the disease from the seed, than from the primary leaf stage. Although the difference is slight, the symptoms from inoculation are not quite as definite as in the compound leaf stage. The third leaf stage symptoms of the disease from the seed were more pronounced than in the earlier stages. The results of these inoculations were not as certain of success as the others. The fourth compound leaf stage proved to be too old for inoculation purposes, since the percentage of successful results was very low.

The plants inoculated with the different types of mosaic developed the characteristic symptoms on the first leaves becoming infected. These symptoms continued throughout the primary growth

in the plant. By primary growth the author refers to the compound leaves arising from the main stem, while the secondary growth arises in the axils of the compound leaves. As the secondary growth started the symptoms became the small or medium rugose type, while the leaves were greatly stunted. These secondary symptoms developed from any type, varying in intensity, comparably with the intensity of the primary infection in the plant. The secondary symptoms did not develop until after the fourth week.

One plant developed all four types distinctly on four different leaves. Each leaf was a distinct type in itself, while two other leaves on the plant were a blending of the four. Of the three types, one leaf was curled, one veinbanded, one small mosaic, and one large rugose. Of the complex symptoms, one leaf was a combination of veinbanding and curling, and one of veinbanding and rugose. In most of the other plants the types were true throughout all of the leaves for the primary symptoms. This was true of the complex symptoms, as well as the pure strains.

Two of the dried specimens were of the true symptoms of Type G, small rugose, and Type I, large rugose. Type P was a complex of veinbanding and large rugose. The true types were much more virulent the first day but lost their virulence more quickly than the complex type. Types G and I produced 80% small rugose and 86% large rugose mosaic from the inoculations made the first day. (Table III). The leaves of Type P, displaying the complex symptoms of leaf banding and large rugose produced seventy three per cent (73%) mosaic from the first inoculations.

TABLE III.

Results of inoculations with extract from dried leaves of Great Northern beans, showing typical mosaic symptoms.

Type	1/31/33				2/1/33				2/4/33			
	No.	+	-	%	No.	+	-	%	No.	+	-	%
G	15	12	3	80%	9	6	3	66%	5	2	3	40%
I	15	13	2	86%	9	-	9	0	5	1	4	20%
P	15	11	4	73%	9	4	4	44%	5	2	3	40%

The second day, Type I produced no mosaic while it produced twenty per cent (20%) the third day. The other two types went gradually down to forty per cent (40%) the third day. The decline of the curve of the Type P was not as great as the curve of Type G. Type I may have been inoculated into a plant in which the symptoms had been masked until after the inoculations. Although Type I was very virulent the first day, its virulence was quickly lost on standing in sterile test tubes, while the virulence of the other two was lost more slowly. The complex type was not as virulent as the other two at the start, but it retained its virulence better.

The inoculations made on the Great Northern bean plants, were divided into two groups; the one, on plants grown from the Ralph Mercer stock of Certified Idaho seed, and the other plants grown from seed from the Huntley Branch Station at Huntley, Montana. One set of inoculations was made in the winter months on these two groups, while another set of inoculations was made during the spring months. Table IV gives the results from the winter inoculations, while Table V gives the results from the spring inoculation in these two groups. Two of the plants used as inoculum for fifteen (15) plants each of the Certified Idaho Stock were, possibly, pseudomosaic as the results were unsuccessful. Type F from tobacco to bean had two (2) diseased plants out of the fifteen (15) inoculated. The disease in these two was probably from the seed, rather than from the inoculations. Type H showed a clearing of the veins in the original plant used for the inoculum.

TABLE IV.

Results of inoculations on Great Northern bean from the Idaho Certified Stock and the Huntley Field Run Stock. These inoculations were made during the winter months.

Inoculum Type	Certified Idaho Stock				Huntley Field Stock			
	No. Inoc.	Positive	%	Negative	No. Inoc.	Positive	%	Negative
A	4	-	0	4	9	-	0	9
B	23	18	78%	5	8	3	37%	5
C	19	12	63%	7	-	-	-	-
F	15	12	80%	3	144	86	60%	58
G	20	14	70%	6	9	6	66%	3
I	22	13	65%	7	9	-	0	9
J	10	9	90%	1	-	-	-	-
K	-	-	-	-	33	29	87%	4
N	25	20	80%	5	17	9	53%	8
P	20	16	80%	4	9	4	44%	5
R	10	9	90%	1	-	-	-	-
* Culture	4	2	50%	2	-	-	-	-
TOTAL	169	124	73%	45	238	139	57%	101

Types and where they were obtained:

A - Huntley Field Run
B - " " "
C - Idaho Certified
D - Huntley Field Run
E - " " "
F - Idaho Certified
G - Huntley Field Run
H - Idaho Certified
I - Huntley Field Run
J - Huntley Field Run
K - Idaho Certified
L - Huntley Field Run
M - Experiment Station Greenhouse
N - Huntley Field Run
O - Huntley Field Run
P - " " "
R - " " "
S - " " "

*Explanation on Page 33.

This type produced no symptoms in the fifteen (15) plants inoculated. Excluding these two types F and B, and H, the percentage of successful inoculations in the spring inoculations is higher than the percentage of the winter inoculations. In the Certified Idaho Stock, the spring inoculations were eighty per cent (80%) successfully diseased, while the winter inoculations were seventy-three per cent (73%) successful. In the Huntley Field Stock the winter percentage of successful inoculations was fifty-seven per cent (57%) as compared to sixty-eight (68%) per cent in the spring inoculations.

Clearing of the veins has been reported as an early symptom of curly top of sugar beet by Ralph Smith (45). It was also reported by K. M. Smight (44) as a symptom of the mosaic virus of Solanaceae, and by Kunkel (26) as a symptom of aster yellows. Type H (Table V) showed the symptoms of clearing of the veins. Fifteen (15) bean plants were inoculated with this type and all proved unsuccessful.

Of the total inoculations, (Table VI) the Certified Idaho Stock was more susceptible to the virus disease of mosaic of bean as the percentage of successfully diseased plants is higher in the Certified Idaho Stock than in the Huntley Field Stock.

Type A was unsuccessful in the Huntley Stock and in the Certified Stock, Table VI. Following thirteen (13) unsuccessful experiments, Type A was discarded for inoculation purposes, as a degenerate or pseudo-mosaic plant. The pods were allowed to ripen and the seeds were germinated in a 30° C. incubator. They were potted in the greenhouse as they

TABLE V.

Results of inoculations on Great Northern Bean from the Certified Idaho Stock, the Huntley Field Stock, and the Brashear Stock. These inoculations were made in the spring months.

Inoculum	Certified Idaho Stock				Huntley Field Stock				Brashear Stock			
Type	No. Inoc.	Positive	%	Negative	No. Inoc.	Positive	%	Negative	No. Inoc.	Positive	%	Negative
D	15	13	86%	2	58	50	86%	8	-	-	-	-
E	-	-	-	-	-	-	-	-	17	7	41%	10
F	15	12	80%	3	85	49	57%	36	-	-	-	-
H	15	0	0	15	-	-	-	-	-	-	-	-
K	15	14	93%	1	76	47	60%	29	10	1	10%	9
L	-	-	-	-	56	47	83%	9	-	-	-	-
M	-	-	-	-	-	-	-	-	14	8	56%	6
O	-	-	-	-	62	33	56%	29	-	-	-	-
S	15	11	73%	4	15	11	73%	4	-	-	-	-
F-T-B	15	2	13%	13	-	-	-	-	-	-	-	-
K-T-B	15	10	66%	5	-	-	-	-	-	-	-	-
Ring spot	-	-	-	-	15	11	73%	4	14	11	78%	3
TOTAL	105	62	59%	43	367	248	68%	119	55	27	49%	28

TOTAL VI.

Total inoculations of Great Northern Bean from the Certified Idaho Stock and the Huntley Field Stock.

Inoculum	Certified Idaho Stock				Huntley Field Stock			
Type	No. Inoc.	Positive	%	Negative	No. Inoc.	Positive	%	Negative
A	4	-	0	4	9	-	0	9
B	23	18	78%	5	8	3	37%	5
C	19	12	63%	7	-	-	-	-
D	15	13	86%	2	58	50	86%	8
F	30	24	80%	6	244	148		96
G	20	14	70%	6	9	6	66%	3
H	15	0	0	15	-	-	-	-
I	20	13	63%	7	9	-	0	9
J	10	9	90%	1	-	-	-	-
K	15	14	93%	1	109	76	60%	33
L	-	-	-	-	56	47	83%	9
N	25	20	80%	5	17	9	53%	8
O	15	12	80%	3	62	33	56%	29
P	20	16	80%	4	9	4	44%	5
R	10	9	90%	1	-	-	-	-
S	15	11	73%	4	15	11	73%	4
F-T-B	15	2	13%	13	-	-	-	-
K-T-B	15	10	66%	5	-	-	-	-
Ring spot	-	-	-	-	15	11	73%	4
* Culture	4	2	50%	2	-	-	-	-
TOTAL	290	199	69%	91	620	400	64%	220

*Explanation on Page 33.

germinated. These plants were watched for signs of the transfer of the mosaic through the seed in an attempt to discover if the plant was diseased in a weakened form, or if it was degenerate.

The pure symptoms in the total inoculations in the Certified Idaho Stock, produced sixty-two per cent (62%) infection, while the inoculum showing complex symptoms, produced eighty per cent (80%) infection.

The Certified Idaho Stock was 1.26 times more susceptible in the winter inoculations, generally, to the mosaic diseases, than the Huntley Field stock. The Idaho stock was quite susceptible to the Types B and N, while the Huntley stock was not as susceptible to these types. As the types were from the Huntley mosaic stock, a slight amount of immunity may have been built up. The Idaho stock being naturally more susceptible may have lacked this degree of immunity, as the Type F was from the Idaho stock and it was 1.36 times more susceptible in the Idaho stock.

Type C produced 98% darker green rugose mosaic/though no typical plants yellow veinbanding symptoms appeared at first. The plants were sprayed with Black Leaf 40 (8 in 100) and molasses for Thrips. Following this the same yellow symptoms appeared throughout the greenhouse. This same yellowing appears in the fields without any previous spraying, but there it is not as prevalent.

Types J and R were predominant in the 1932 Huntley field selections. The Idaho stock proved 90% susceptible to both types. These two types appeared to be of a very virulent nature in both stocks.

Type K of the Mercer plants produced 88% successful inoculations in

the Huntley field stock. The plants inoculated with this type showed very prominent symptoms.

The culture juice was sterilized through a Chamberlain filter, and plated on agar. A minute coccus was found in the juice. On the plated agar, numerous minute translucent colonies were formed. This micrococcus was a gram negative. When inoculated into the plants, it produced negative results as two were diseased and two were not. This micrococcus may have entered from some other source regardless of the care in handling the apparatus, or it may have been a saprophyte originally from the bean plants. The two successful inoculations may have been originally diseased plants. Tables IV and VI.

The 1932 and 1931 selections from the Huntley Branch Station were tested for resistant individuals. (Tables VII and VIII). Selections No. 45, 67, and 70, were resistant to the virus of Type F, a very virulent type. Selections 38, 39, 55, 56, and 69 were 100% successful inoculations and contained no disease in the uninoculated checks. Selection 36 was susceptible to Type F, but was unsusceptible to Type K. Selection 48 was susceptible to virus B while it was unsusceptible to Type K. Contrasted to this, selections 37, 42, 43, 47, and 60, were resistant to F, although susceptible to K, while Selection 4 was resistant to B and susceptible to K, and Selection 50 was resistant to Type N, while susceptible to K. Selection 47 was 5% diseased, susceptible to only one Type K, while Selection 53 had 33% susceptible inoculated plants.

Selections No. 45, 67, and 70 were germinated in a 30° C. incubator and were potted in the greenhouse. They were tested with Type K for

TABLE VII

Results from the inoculations of the seventy-four selections of Great Northern Bean made in 1932 from the Huntley Field Stock

Selection	% Germ. 3/31/33	% Germ. 5/9/33	% Hard	No. Inoc.	Types	Results +	Results -	%	Un-inoc. +	Un-inoc. -	Mosaic
1	66%	66%	0	5	K	1	4	20%	1	4	20%
2	60%	80%	20%	-	-	-	-	-	12	-	100%
3	93%	73%	20%	5	K	4	1	80%	-	6	0
4	60%	60%	0	3	K	3	-	100%	1	5	16%
5	60%	86%	26%	3	K	3	-	100%	10	-	100%
6	73%	80%	13%	4	K	3	1	75%	-	8	0
7	60%	80%	20%	4	K	-	4	0	-	8	0
8	73%	80%	7%	4	K	3	1	75%	4	4	50%
9	46%	73%	27%	4	K	3	1	75%	-	7	0
10	73%	86%	13%	5	K	-	5	0	-	8	0
11	46%	86%	40%	1	K	1	-	100%	12	-	100%
12	73%	73%	0	5	K	5	-	100%	-	6	0
13	46%	60%	14%	4	K	4	-	100%	1	4	20%
14	73%	73%	0	3	K	3	-	100%	7	1	87%
15	26%	26%	0	2	F	-	2	0	2	-	100%
16	46%	53%	7%	2	F	2	-	100%	6	-	100%
17	66%	66%	0	5	F	5	-	100%	1	4	20%
18	73%	73%	0	-	-	-	-	-0	11	-	100%
19	73%	73%	0	1	F	1	-	100%	10	-	100%
20	66%	80%	14%	5	F	5	-	100%	-	7	0
21	86%	73%	13%	5	D	3	2	60%	2	4	33%
22	73%	73%	0	5	D	2	3	40%	-	6	0
23	40%	60%	20%	3	D	3	-	100%	2	4	33%
24	46%	46%	0	4	D	4	-	100%	1	2	33%
25	56%	80%	14%	5	D	5	-	100%	2	5	26%
26	33%	60%	27%	3	D	3	-	100%	-	6	0
27	53%	66%	13%	3	D	3	-	100%	7	-	100%
28	66%	73%	7%	5	D	1	4	20%	5	1	83%
29	80%	80%	0	6	D	6	-	100%	2	4	33%
30	86%	86%	0	6	F	6	-	100%	4	3	33%
31	80%	80%	0	6	F	6	-	100%	-	6	0
32	73%	73%	0	5	F	2	3	40%	1	5	16%
33	93%	86%	7%	6	F	4	2	66%	1	6	14%

TABLE VII (Continued)

Selection	% Germ. 1/28/33	% Germ. 2/25/33	% Hard	No. Inoc.	Types	Results +	Results -	%	Un-inoc. +	Un-inoc. -	% Mosaic
34	43%	63%	20%	7	4F	4	-	100%	1	11	8%
					1K	1	-				
					2P	2	-				
35	30%	53%	23%	7	4F	4	-	57%	-	10	0
					2K	-	2				
					1P	-	1				
36	46%	80%	34%	7	3F	3	-	57%	1	15	6%
					1K	1	-				
					3P	-	3				
37	26%	70%	44%	7	2F	-	2	71%	2	12	14%
					2K	2	-				
					3P	3	-				
38	36%	53%	17%	7	3F	3	-	100%	-	9	0
					4G	4	-				
39	60%	86%	26%	8	4F	4	-	100%	-	17	0
					3G	3	-				
					1K	1	-				
40	30%	56%	26%	7	2B	2	-	100%	11	2	84%
					1G	1	-				
					4K	4	-				
41	33%	63%	30%	7	3B	-	3	42%	-	11	0
					1G	-	1				
					3K	3	-				
42	30%	80%	50%	7	2F	-	2	71%	1	16	5%
					5K	5	-				
43	33%	76%	43%	7	4F	-	4	42%	1	16	5%
					3K	3	-				
44	40%	53%	13%	7	1I	-	1	71%	1	9	10%
					6N	5	1				
45	56%	73%	17%	7	6F	-	6	0	-	15	0
					1I	-	1				
46	56%	80%	24%	7	6F	3	3	42%	-	16	0
					1I	-	1				
47	53%	83%	30%	7	3F	-	3	5%	-	17	0
					3I	-	3				
					1K	1	-				
48	36%	56%	20%	7	3B	3	-	42%	-	10	0
					3I	-	3				
					1K	-	1				
49	56%	70%	14%	6	N	4	2	66%	2	13	13%
50	63%	90%	27%	6	1K	1	-	16%	2	19	9%
					5N	1	5				

TABLE VII (Continued)

Selection	% Germ. 1/28/33	% Germ. 2/25/33	% Hard	No. Inoc.	Types	Results +	Results -	%	Un-inoc. +	Un-inoc. -	% Mosaic
51	66%	100%	34%	6	F	6		100%	12	12	50%
52	30%	83%	53%	6	4F 2K	4 2		100%	5	14	35%
53	50%	70%	20%	6	F	2	4	33%	-	15	0
54	66%	76%	10%	6	F	4	2	66%	1	16	5%
55	70%	86%	16%	6	F	6	-	100%	-	21	0
56	63%	73%	13%	6	F	6	-	100%	-	16	0
57	50%	70%	20%	6	F	5	1	83%	3	12	20%
58	70%	83%	13%	7	F	7	-	100%	3	15	16%
59	53%	63%	10%	8	F	5	3	61%	1	10	9%
60	33%	63%	30%	6	5F 1K	- 1	5 -	16%	2	9	18%
61	66%	93%	27%	0	-	-	-	-	-	-	100%
62	53%	73%	20%	2	F	2	-	100%	16	4	80%
63	50%	70%	20%	6	F	1	5	16%	1	14	6%
64	53%	80%	27%	4	F	2	2	50%	2	18	10%
65	50%	93%	43%	6	F	4	2	66%	2	20	9%
66	43%	66%	23%	6	F	6	-	100%	1	13	7%
67	83%	96%	13%	6	F	-	6	0	0	23	0
68	50%	70%	20%	5	F	4	1	40%	-	16	0
69	10%	83%	73%	7	2F 5K	2 5	- -	100%	-	18	0
70	60%	70%	10%	7	F	-	7	0	-	15	0
	3/31/33	5/9/33									
71	53%	66%	13%	-	-	-	-	-	9	-	100%
72	46%	66%	20%	3		3	-	100%	7	-	100%
73	60%	60%	0	4		4	-	100%	-	5	0
74	73%	73%	0	5		4	1	80%	1	5	16%

*Percentage of plants that died.

TABLE VIII

Results from inoculations made on the 1931 selections of Great Northern Bean from the Huntley Field Stock

Selection	% Germ. 3/31/33	% Germ. 4/19/32	% Hard	No. Inoc.	Types	Results +	Results -	%	Un-inoc. +	Un-inoc. -	% Mosaic
1	33%	33%	0	2	F	-	2	0	-	3	0
3	60%	80%	20%	4	F	-	4	0	1	7	12%
6	60%	66%	6%	4	F	-	4	0	1	5	16%
8	0	83%	83%						1	12	7%
9	13%	40%	27%	1	F	1	-	100%	1	4	20%
13	66%	73%	77%	5	F	3	2	60%	-	6	0
14	60%	73%	13%	4	F	3	1	33%	4	3	57%
16	73%	83%	10%	5	F	-	5	0	-	8	0
18	60%	66%	6%	5	F	5	-	100%	-	5	0
19	20%	26%	6%	1	F	-	1	0	1	2	33%
25	20%	all died									
26	46%	53%	7%	3	F	-	3	0	1	4	20%
28	33%	33%	0	3	F	3	-	100%	-	2	0
29	60%	60%	0	5	F	-	5	0	-	4	0
31	60%	73%	13%	5	K	-	5	0	-	6	-
32	93%	80%	*13%	6	K	1	5	16%	-	6	0
33	46%	60%	14%	4	O	-	4	0	1	3	25%
34	53%	53%	0	3	O	-	3	0	-	5	0
36	73%	73%	0	6	O	6	-	100%	-	5	0
37	73%	86%	13%	6	O	-	6	0	-	7	0
38	86%	93%	7%	6	O	-	6	0	-	8	0
39	73%	73%	0	6	O	5	1	86%	1	4	20%
42	66%	80%	14%	6	O	6	-	100%	1	5	16%
44	80%	86%	6%	5	O	2	3	40%	-	8	0
47	66%	80%	14%	5	O	-	5	0	-	7	0
51	20%	40%	20%	-	-	-	-	-	6	-	100%
52	93%	86%	* 7%	7	L	7	-	100%	-	6	0
53	80%	80%	0	5	L	3	2	60%	-	7	0
54	33%	33%	0	2	L	2	-	100%	-	3	0
55	40%	53%	13%	3	L	3	-	100%	1	4	20%
56	46%	46%	0	3	L	3	-	100%	-	4	0
58	26%	26%	0	1	L	-	1	0	-	3	0
59	40%	46%	6%	3	L	2	1	66%	3	1	75%
61	53%	53%	0	4	L	3	1	75%	3	1	75%
62	40%	33%	* 7%	2	L	2	-	100%	3	-	100%
66	13%	33%	20%	1	L	1	-	100%	3	1	75%
67	66%	66%	0	4	L	3	1	75%	1	6	14%
69	56%	26%	*30%	4	L	3	1	75%	-	-	-
70	73%	80%	7%	5	L	4	1	80%	1	6	14%

*Percentage of plants that died.

further tests for susceptibility. No mosaic symptoms resulted.

The Huntley Branch Station 1927 selections of Great Northern and Red Mexican beans were germinated in the incubator and were transfered to the greenhouse to pots. No mosaic symptoms were observed. (Table IX).

TABLE IX.

Results of the germination and presence of mosaic in the 1927 selections from the Huntley Branch Station

Selection No.	Germ.	No. Grew	Did not Grow	Results
1	100%	80%	2 rotted	No mosaic
2	100%	60%	4 molded	No mosaic
3	90%	90%	-	No mosaic
4	100%	90%	1 wormy	No mosaic
5	70%	50%	1 rotted 1 wormy	No mosaic
6	100%	100%	-	No mosaic

W. H. Pierce (36) found seed to contain mosaic after thirty years storage. Duggar (14) found that the seed protein in sufficient quantities, would inactivate the virus, although the amount of protein in each seed was insufficient to inactivate the virus. He states that the amounts contained in the seed, might produce this inactivation when the seeds were held in storage. The seeds used from the 1927 selections were six years old. No mosaic was observed on the fifty nine plants.

Five of the twenty unpricked seeds of selection sixty-nine of the Huntley Branch Station 1932 selections germinated in incubation at 30° C. The twenty pricked seeds all germinated in the incubator. The unpricked seeds required five days to germinate, while the pricked seeds required three days. In the bench, twenty-four of the twenty-five pricked seeds came up. These seeds had failed to germinate. The seeds were fertile, but the seed coats were imprevious to water and were, therefore, hard seeds.

The first three sets of seeds placed in the incubator, were dusted with Cuprous oxide. The last set was disinfected with Uspulun. The sets dusted with Cuprous oxide, were free from mold for two weeks, while the ones sterilized with Uspulun were moldy within five days. These seeds were then dusted with Cuprous oxide and placed in the incubator again. They did not regain their moldy state for a week.

The mosaic Types B and N produced no symptoms on the tobacco (Table I). Type K produced a small ring of yellow around each injured hair, within three days. This strengthened in color for two days and then

slowly faded. The check plants that were incubated at the same time from healthy leaves produced no yellowing at the base of the injured hairs. Type F displayed the same yellow circle at the base of each injured hair. The leaves immediately above each inoculated leaf became fainly mottled one week after the inoculation. This mottling slowly faded out. Type C produced no symptoms of mosaic.

Trichomes of healthy and diseased beans were mounted and observed. The cytoplasm of the healthy trichomes was no less granular than the cytoplasm of the diseased trichomes.

DISCUSSION

The mosaic in Phaseolus vulgaris is transmitted through the seed from mosaic plants, as the diseased plants appeared in new soil in benches, (that had not contained bean plants before). It is possible the protein in the seed will inactivate the virus in time, though it may take a few years to accomplish this. Artifical transmission of the virus has proven successful although there are three possibilities. First, that the mosaic may have been in the seed, although masked or weakened, and appeared later in the plant. The inoculation may have strengthened the virus within the plant. Second, the plant may be a resistant individual that would not become infected. Third, the mosaic stock used for the inoculum may have been a degenerate plant rather than a truly mosaic plant, or it may have been of a very weak individual. The suspiciously appearing plants may be caused by either weakened virus or by a degeneration of the plant.

Yellowing, which appeared in large quantities in the greenhouse, would seem to be due to the spray injury, but when in the fields and not subjected to spray it may be due to degeneration of the chloroplasts of the plant. McKinney (31) found a yellow symptom in tobacco that he was able to concentrate to a pure form through artificial systematic inoculations. The yellowing in the beans does not appear to be comparable to the yellowing in the tobacco. The yellowing in the bean can not be transfered to other beans or other plants by artificial inoculations.

The mosaic diseases of Phaseolus appear to be of different types. Some plants have a resistance to one virus while they may be susceptible to all others, or they may be resistant to all but one. The pure forms of the virus are very prevalent and usually remain pure throughout. The complex forms of the disease appear to be mixtures of the virus. The two forms combined as one, are not as virulent at first as the pure type, but they appear to act in conjunction in their ability to retain the virulence of the disease.

In the field the symptoms remain of the pure type to the end of the season and do not degenerate into the small rugose, as they do in the greenhouse. By bringing out the secondary growth in the plant, any virus that might be masked, either from the seed or from later infection, would be brought out. This may account for the degeneration of the type. The shortening of the growing period and the forcing of the plants may bring forth the virus in the plant. Fernow (17) found mosaic symptoms to

change with the environmental conditions. This would account for the change between the symptoms in the field and greenhouse.

The clearing of the veins in the bean plant can not be considered a symptom of the disease as it can in the sugar beet, Solanaceae, or aster. Inoculations made from leaves showing clearly the property of clearing of veins, produced no infection.

The ring spot of the alfalfa does transfer to the bean, but not as the typical ring spot mosaic. It transfers as a faint mottling of light yellow green on darker green, without the accompaning symptoms of rugosity. The ring spot of alfalfa does not transfer over to the tobacco, oxalis, sweet clover, or fanweed. It only transfered to the red clover. The fanweeds inoculated with this type died.

The dark green curling of the leaves, transfered over a darkened curling. The leaves were slightly elongated, but not as elongated or as dark as the original inoculum.

The presence of the virus in the seed did not influence the germination of the seed as rows having the least number or the greatest number of seeds germinate, contained no virus. Similar rows contained a great amount of virus. The failure to germinate was caused by the imperviousness of the seed coat rather than the unfertility of the embryo or the influence of the virus. A higher percentage of germination could be obtained if the seed coats were slightly scratched, without injuring the embryo, before planting. Cuprous oxide proved the most effective fungicide in controlling the fungi on the seeds.

The virus of Phaseolus appeared to transfer to the tobacco as the

light yellowish rings were formed around each injured hair in the inoculated plant. As the check plants did not show this yellowing at the base of each injured hair, there must have been a transfer of the mosaic in the inoculated plants. The symptoms gradually disappeared. Bean plants inoculated from the tobacco did become sixty per cent diseased in one type only. The virus may have transfered over but it was unable to increase greatly in the tobacco plant. The clover showed no signs of any transference of the mosaic from the bean. It did show a faint transference of the ring spot from the alfalfa. The tobacco did not demonstrate any symptoms from the alfalfa ring spot.

The mosaic disease of the Phaseolus appears to be specific to the variety of the bean, and is not readily transfered to other plants.

The Huntley field run bean is less susceptible to the disease than the Idaho Certified, possibly because of the change in environment and the inoculations of other viruses and possibly because the Idaho Stock is naturally less resistant to the virus as a group. They were more susceptible to the virus from their own stock as well as the virus from the Huntley Stock.

Drying does not inactivate the virulence of the disease. Inoculations made from dried material were as successful as the inoculations made from the fresh material. The virus loses its virulence gradually upon staning as an extract.

The virus in the seed does not remain active over a period of a few years as there was no mosaic obtained from the seeds that were six years old. They were from plants that were mosaic as well as one set

from healthy plants. None of the plants showed any signs of the symptoms of mosaic. The mosaic did not live over in the seed. The seeds were originally from plants that had become mosaic through their seed. The first and second compound leaf stage was the optimum time for the transference of the disease artificially or through injury. Before that time the mosaic may not have shown up, while after that time the percentage of diseased plants is not as great. The second compound leaf should be just unfolding to be used. The swab and pot label method of artifical transference, if performed at this time, is of the best methods. The swab and pot label in being used for the inoculations, eliminate all possible transference of the disease by the hands of the worker. Through changing the label after each inoculation, there is no possibility of careless transfer.

The plants that were injured and allowed to rub together were too old in the field. There was a transfer, so there is a possibility of a transference of the disease in the field through careless handling of the plants while they are still young.

SUMMARY AND CONCLUSIONS

From the foregoing discussion, it would appear that the viruses of bean are of different types and possibly of different intensities, as their reactions on the plants of the same species of Phaseolus, was quite diversified. Some types proved more virulent than others to the same selections of the Great Northern bean. The bean may have built up a resistance to one type while it is susceptible to all other types,

or it may have a resistance to all types.

The sterile pot label and swab method of inoculation of the plant to be infected, is one of the surest methods. The workers' hands do not come in contact with the diseased plant or the healthy plant therefore there is little chance of careless transfer of the disease. The leaves can be gently rubbed or injured with the swab, so a technique of inoculating can be easily gained.

The mosaic disease of bean may vary with environmental conditions as the winter inoculations were not as successful as the spring inoculations.

Although the yellow veinbanding is a symptom of spray injury, it appears in the field in isolated cases. This symptom could not be transmitted to other plants through inoculation processes. It is probably an abnormality, the cause of which is not determined.

The clearing of the veins is an early symptom of mosaoc of Solanaceae, sugar beet, and aster, but it is not transmitted from bean plant to bean plant. This clearing of the veins is either the symptom of an abnormal form of bean plant or of a weakened form of virus, that is not in a great enough quantity or in a virulent enough stage to produce mosaic symptoms through inoculation.

The different types of mosaic as demonstrated by the different patterns of the symptoms, can be either of four different pure types or as compound symptoms. The compound symptoms of the disease were more virulent as a group than the pure symptoms of the disease. The virulence of the complex symptoms were retained longer than the pure symptoms

in infected juice.

The virus is not inactivated through drying for five months as dried material is as virulent as the fresh material.

The germination of the seeds was not low due to the presence of virus within the seed, but rather because of the imperviousness of the seed coat to water. A germination test is no indication of the presence of virus within the seed.

The seeds from virus plants, when allowed to remain in storage for five years, produced plants which showed no symptoms of disease.

The environment of the greenhouse tends to bring out a degenerate form of virus in the secondary growth of the diseased plant, characterized by a slight rugosity and fine uniform mottling.

The Ring spot of alfalfa transfered to the bean plants in a slightly different type symptom than any of the symptoms of the mosaic disease of bean, and very much different from the inoculum. It would not transfer to the tobacco or to the sweet clover.

The tobacco appeared to be susceptible to only one type out of six (6) of the viruses of the bean. It produced yellow lesions at the base of each injured hair for two types but only one multiplied enough in the tobacco plant to be transfered back to the bean. The symptoms gradually disappeared in the tobacco. The disease probably transfered over to the tobacco but was unable to multiply there as it should, and soon died out.

The optimum time for successful inoculations for Phaseolus is in the first compound leaf stage, just as the second compound leaf appears and before it has unfolded. This is true of artificial as well as natural infection. Symptoms of mosaic usually appeared within fifteen (15) days after inoculation.

The disease may be transfered in the earlier stages through rough handling in the field during the processes of roguing. Great care should be exercised in the field especially if the plants are young.

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EXPLANATION OF PLATES

Plate I. A leaf of Phaseolus vulgaris demonstrating the symptoms of veinbanding. (3/4 natural size)

Plate II. A leaf of Phaseolus vulgaris demonstrating the symptoms of small rugose. (Natural size)

Plate III. A leaf of Phaseolus vulgaris demonstrating the symptoms of large rugose. (3/4 natural size)

Plate IV. A leaf of Phaseolus vulgaris demonstrating the symptoms of veinbanding and curling. (Natural size)

Plate I

Plate II

Plate III

Plate IV