

Effects of Soybean Mosaic Virus Infection on Nodule Formation in *Glycine max* (L.) Merr.

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大豆모자이크 바이러스 감염이 大豆根瘤形成에 미치는 影響

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ABSTRACT

This investigation was conducted to study the effect of soybean mosaic virus (SMV) on various parameters of nodule formation at different stage of soybean plants. Differences in nodule formation were marked between soybean varieties tested, but nodules were small within soybean varieties infected with SMV. SMV-infection on soybeans were greatly reduced the number, size and weight of nodules, and the earlier the infection of SMV, the greater the reduction of nodules. Maximum reduction (83%) of nodules observed when "Kumkang-Daerip" soybeans were inoculated 2 weeks after seeding, but none occurred 8 weeks or later. Prominent decreases in number of nodules often resulted in an increase in nodule sizes in SMV-infected soybean plants.

INTRODUCTION

Soybean plants infected with soybean mosaic virus (SMV) generally exhibit growth retardation, delayed seed maturation and yield reduction (Bawden 1964; Lee *et al.* 1968). Dwarfing and elongated internodes of the virus infected plants have occasionally been observed in the soybean growing fields (Lee *et al.* 1968). Photosynthesis and chlorophyll is reduced in many soybean mosaic virus infected plants (Bawden 1964; Bawden and Pirie 1952; Esau 1956). This phenomena seems to occur also in SMV-infected soybean plants. Fewer nodules have been observed consistently in SMV-infected soybean

plants in the field of Korea Atomic Energy Research Institute experimental farm, Kum-Kok, Kyunggi-do and in the greenhouse, however the effect of soybean mosaic virus infection on the development of nodule formation has not been determined. Therefore, this investigation was conducted to study the effect of soybean mosaic virus on various parameters of nodule formation at different stage of growth of soybean plants.

MATERIALS AND METHODS

Three local soybeans, "Kumgang-Daerip (K-D)", "Jangdan-Baikmok", "Chungbuk-Baik", and two introduced varieties "Clark" and "S. elby" were

employed in this experiment. The soil taken from the soybean fields which was known to be infected with *Rhizobium* species was used for planting seeds. All greenhouse plantings were made in unsterilized sand: peat: field soil mixture in the ratio of 1:1:2. Two seeds were planted in 6-inch clay pot in the greenhouse temperature range at $26 \pm 3^\circ\text{C}$. Healthy seedlings were mechanically inoculated 2 weeks after planting seeds. Soybean mosaic virus inoculum was prepared by expressing crude sap from SMV-infected leaves of soybean plants and diluting 10 times with 0.01M phosphate buffer solution (pH 7.0) added with small amount of 600 mesh carborundum. The SMV inoculum was then rubbed on leaves of soybean plants previously dusted with carborundum. All control plants were rubbed with neutral phosphate buffer and carborundum. The SMV-inoculated leaves were rinsed with running tap water immediately after inoculation. Nodules formed each pot were collected 10 weeks after seeding. Detailed procedure of specific experiments preface the results described.

RESULTS

Effects of SMV infection on nodules.—Ten pots of each cultivar (Kumkang-Daerip, Jangdan-Baikmok, Chungbuk-Baik, Clark and Shelby) were planted in the greenhouse. Each cultivar group was divided into two groups of 5 plants each. One group was inoculated with SMV and the other group was the noninoculated control. Variation in nodule size, weight and numbers caused by SMV among cultivars are shown in Table 1. The five different cultivars infected with SMV produced nodules that were reduced in weight, size and number were varied in their nodulation ability as compared with healthy control in the greenhouse condition (Table 1). The variance may be due to genetic difference in varietal susceptibility. Nodule reduction appeared correlated with differences in varietal susceptibility. Jangdan-Baikmok was highly susceptible, exhibiting mild leaf rugosity with very indistinct mottling. Clark and Kumkang-Daerip showed intermediate symptom

pattern.

Effect of time of infection on nodule formation.—Two soybean varieties Clark and K-D were used in this experiment. One hundred pots were seeded to each variety and divided into 10 groups. In the first group of 10 pots, soybeans growing in 5 pots were inoculated at the unifoliate, 2 weeks after seeding, with SMV inoculum; the other 5 pots were used control checks. The remaining group of 10 pots were treated identically at 1-week intervals. The effect of time of infection of SMV on the nodule formation of both Clark and K-D variety was similar in their mode of pattern (Fig. 1—B). Fresh weight of nodules reduced 10% in SMV infected K-D variety which showed milder mottlings, when compared with a reduction of 22% in the Clark (Table 1). For example, weight of nodule was least when plants were inoculated with SMV two weeks after planting. Similarly the adverse effect of SMV infection on number of nodules and fresh weight of plants less in Clark than in K-D (Fig. 1—B and D).

Nodule weight (Fig. 1—A) variation in K-D controls occurred in a range of 1.10 to 1.20g/plant. The maximum deviation from the mean (1.15g/plant) in the control was 0.05g, which was roughly $\pm 5\%$. Nodule weight in SMV-infected K-D variety, on the other hand, varied from 0.38 to 1.06g/plant, and was inversely related to growth stage when the soybeans were inoculated. There was little effect of the virus infection during the last two weeks of the experiment on final nodule weight.

The maximum variation in nodulen numbers among controls was an average of ± 2 nodules/plant, a $\pm 3\%$ deviation from the mean of 84 nodules/plant (Fig. 1—B). Soybean plants inoculated with SMV in early stages of growth showed least numbers of nodules, while those inoculated in the later stages were almost unaffected.

Variation in nodule size among the control was small enough to draw a straight line (Fig. 1—C), while size of nodules varied greatly between SMV-infected soybean varieties (Table 1). Plants infected by SMV in 4th to 6th weeks after seeding produced larger nodules than their controls of the

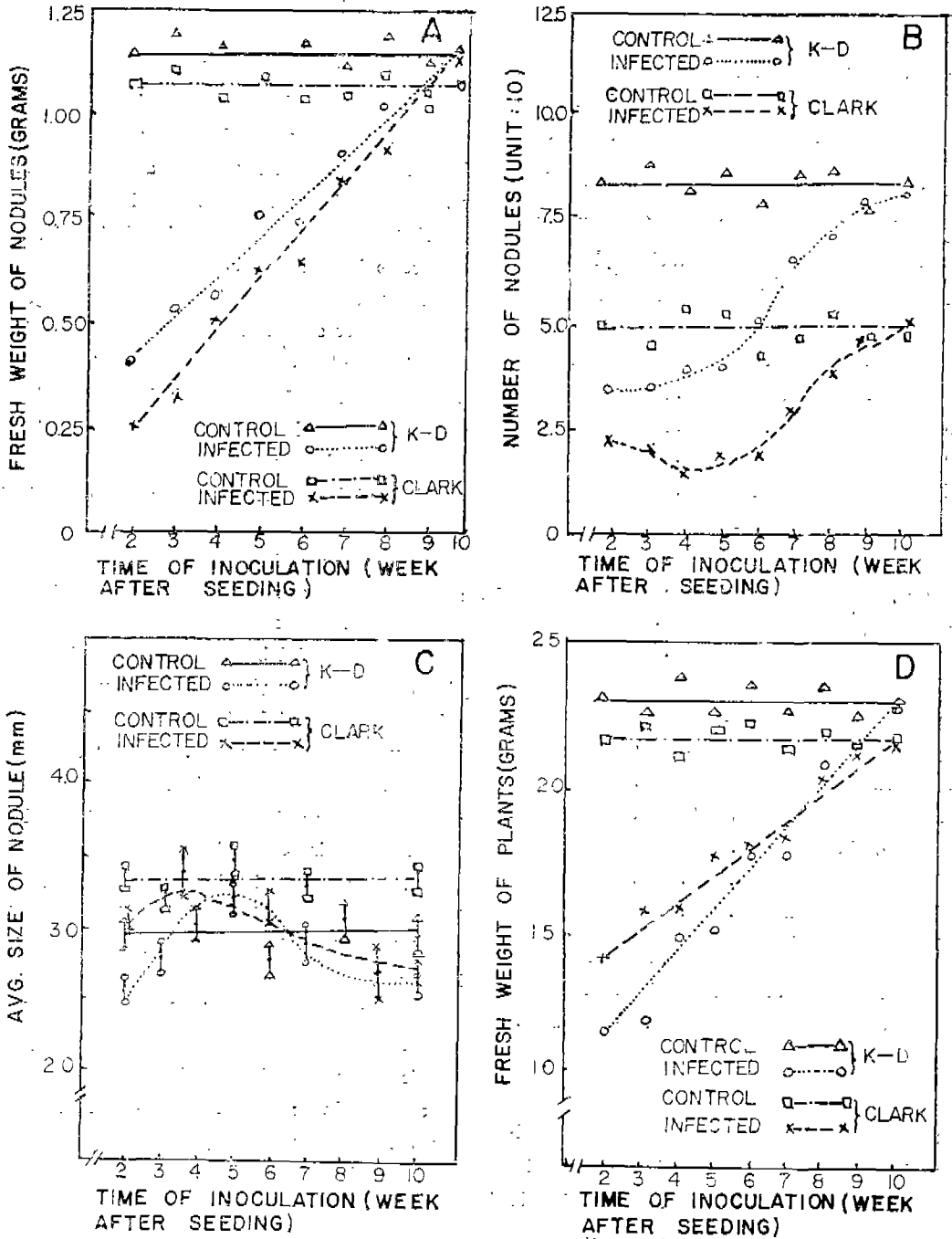


Fig. 1. Effect of soybean mosaic virus (SMV) infection on nodule formation of Kungang-Daerpi(K-D) and Clark soybeans. A) Weight. B) Number. C) Size of nodules per plant(each bar represents the maximum variation of nodule measurements at the dot), D) Fresh weight of plant (root not included).

variety K-D.

Variation in fresh weight of plants, not including roots, ranged from 21 to 24g/plant in the controls (Fig1—D). In general, the earlier the infection of SMV, the greater reduction in fresh weight. SMV-infected soybean plants were slightly taller than

Table 1. The effect of soybean mosaic virus infection on nodulation of soybean plants

Varieties	Average Weight/plant		
	Diseased	Healthy	Reduction
	g	g	%
Jangdan-Baikmok	1.12	1.36	18
Kumkang-Daerip	1.37	1.52	10
Chungbuk-Baik	0.74	0.95	23
Clark	1.24	1.59	22
Shelby	1.20	1.37	13
	Average no. Nodules/plant		
Jangdan-Baikmok	57.6	69.2	17
Kumkang-Daerip	72.4	84.3	15
Chungbuk-Baik	58.8	63.7	8
Clark	38.6	43.2	11
Shelby	50.1	57.8	13
	Average Size Nodules/plant		
	mm	mm	
Jangdan-Baikmok	2.1×2.4	2.6×3.0	
Kumkang-Daerip	2.5×2.9	2.9×3.2	
Chungbuk-Baik	1.9×2.2	2.2×2.4	
Clark	3.0×3.2	3.3×3.5	
Shelby	2.4×2.7	2.8×3.0	

healthy plants due to stem elongation. Nodule weight was relatively more affected by SMV infection than was fresh weight of plants (Fig. 1—A and D).

DISCUSSION

Numerous environmental factors such as soil acidity, nutrition, C-compound availability, and rhizobial population, etc., affect the nodule formation in soybeans (Van Schreven 1958). Joshi *et al.* (1967) reported that the effect of a clover phyllody virus on nodulation of white clover probably involved a mycoplasma rather than a virus. Soybean plants infected with SMV reduced the size, number, and

weight of nodules, and most pronounced reductions of these occurred in early infections. Reduction of nodules are probably caused by the virus multiplication causing physiological changes of reduced photosynthesis or increased respiration (Bawden 1964; Bawden and Pirie 1952; Diener 1963), imbalance of enzyme level and auxins (Smith *et al.* 1968), which may directly or indirectly affect the *Rhizobium*-soybean symbiotic relationship.

Since soybean mosaic virus reduced both size and number of nodules on soybean plants, the process is probably more complicated physiology than a simple reduction of photosynthesis or increased respiration. Van Schreven (1958, 1959) insisted that limiting CO₂ and light levels affect photosynthesis that influence nodule formation and function. SMV infection had little effect on preventing or delaying nodule formation. SMV affected nodule growth immediately, according to our data, and possibly had a adverse effect on rhizobial establishment in newly formed roots. Greater soybean nodule numbers were always accompanied by smaller nodule size. According to Nutman (1958), a similar inverse relationship of average nodule size and abundance occurs in red clover and other legumes. The most prominent relationship between sequential infection of the virus and nodule formation was reflected in the linear nodule weight increases with delayed inoculated (Fig. 1—A). Soybean mosaic virus has a more complex influence on the symbiotic relationship and its nitrogen-fixing ability. Nodulation was reduced by SMV infection in all soybean varieties tested, although there was variations in varietal reactions (Table 1). Likewise, there may have varietal and specific genetic differences in symbiotic effectiveness of *Rhizobium* (Bowen and Kennedy 1961). Differences of nodule formation are perhaps due to varietal differences in disease severity.

REFERENCES

- Bawden, F.C. 1954. Plant virus and virus diseases. Ronald Press, N.Y. 361p.
 Bawden, F.C. and N.W. Pirie, 1952. Physiology of virus diseases. Annu. Rev. Plant Physiol. 3: 171—188.

- Bowen, G.D. and M.K. Kennedy 1961. Heritable variation in nodulation of *Centrosema pubescens* Benth. Ad. J. Agr. Sci 18 : 161—170.
- Diener, T.O. 1963. Physiology of virus-infected plants. Ann. Rev. Phytopath. 1 : 197—218.
- Esau, K. 1956. An anatomists view of virus disease. Amer. J. Bot. 43 : 739—748.
- Joshi, H.U., A.J.H. Carr and D.G. Johnes 1967. Effect of clover phyllody virus on nodulation of white clover (*Trifolium repens*) by *Rhizobium trifoli*. J. Gen. Microbiol. 47 : 139—159.
- Lee, J.H., Oh, J.H., Chang, B.U. and C.K. Park 1968. A study of a bean mosaic virus. J. of Nuclear Sci., Office of Atomic Energy 8(1), Ser. 2 : 155—161.
- Nutman, P.S. 1958. The physiology of nodule formation, p.87—107. In Nutrition of the legumes. Butterworths Sci. Publ. London.
- Smith, S.H., McCall, S.R. and J.H. Harris 1968. Auxin transport in curly top virus-infected tomato. Phytopath. 58 : 1669—1670.
- Van Schreven, D.A. 1958. Some factors affecting the uptake of nitrogen by legumes, p. 137—163. In Nutrition of the legumes. Butterworths Sci. Publ. London.
- Van Schreven, D. A. 1959. Effects of added sugars and nitrogen on nodulation of legumes. Plant Soil. 11 : 93—112.

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