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EFFECT OF INOCULATION ON SEED AND  
FORAGE YIELD AND OTHER CHARACTERISTICS  
IN LEGUMES

by

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Legume Inoculation

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## ABSTRACT

A two-year study was undertaken to evaluate the effect of inoculation on the seed yield, forage yield, and other characteristics of four species of legumes.

The inoculated and non-inoculated legume plants did not show any significant differences in the seed yield, forage yield and the other characteristics studied. The four species used, however, did vary widely in many of the characteristics that were evaluated. Under a normal amount of rain-fall in 1962 vetch appeared to be the best in the seed yield, followed by lentils, Canadian field peas and Austrian winter peas. In forage yield, lentils and vetch seemed to be the best. Lentils gave the highest total protein yield of the forage in both years and the highest total protein in yield of the seed in 1962.

It is most likely that the untimely rainfall and the probable presence of the proper rhizobial species in the soil of the Bekaa' limited the efficiency of the added inocula. However, no definite conclusion can be made until more work both under irrigation and dry-land conditions is carried out for a longer period.

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## INTRODUCTION

Civilization owes a great debt to legumes (3). The supply of available nitrogen is often a limiting factor in plant growth in many parts of the world. The legumes are capable of obtaining their nitrogen requirements from air through symbiosis with nitrogen fixing bacteria and are also able to benefit the succeeding crop by fixing nitrogen in soil. The legumes are valuable in soil conservation programs because they improve the soil tilth and protect the surface from erosion. They also store in their leaves and seeds an abundance of protein, the basis of the protoplasm of all plants and animals. They also contain desirable amount of bone building minerals and vitamins (3). All these facts have placed the legumes as an important human food, and also have increased their demand in live-stock feeding and as green manures.

The successful growing of legumes is dependent upon the right kinds of rhizobia being present in the soil. Inoculation, the practice of adding the proper species of bacteria to legume seed or soil, may mean the difference between success and failure of the legume crop. The relative importance of leguminous plants for food, for live-stock, for soil improvement, for soil conservation and for industrial products depends primarily upon the effectiveness of nitrogen fixing bacteria living in the nodules on the legume plant roots. The bacteria enables the legumes

to use the nitrogen from the air, building it into protein and to enrich the soil in which they grow. In fact, inoculated legumes play three important roles in the world's food program: (i) they increase the supply of high protein feeds for domestic animals, (ii) they increase the yield of many legumes valuable to man for food and (iii) indirectly they increase the production of other farm crops (8).

In view of the important roles played by inoculated legumes a study was undertaken to determine the effect of inoculation on forage and seed yield and protein contents in four species of legumes.

The experiment was conducted in the Bekaa plain, one of the main agricultural areas of Lebanon, where farmers are generally short of good quality pasture for their animals especially during winter and spring months. In this study four species of legumes, vetch, lentils, Austrian winter peas, and Canadian field peas were grown for two seasons in 1960-61 and 1961-62 under normal rain-fed conditions.

## REVIEW OF LITERATURE

It was Malpighi in 1687 who first recorded the presence of nodules on the roots of leguminous plants. He could not realize the significance of these root nodules and thought them to be root galls (6). In 1866 Woronin, a Russian botanist found root nodules on leguminous plants to be filled with minute bacteria and concluded the 'abnormal' outgrowths to be a pathological condition (6). According to Carroll (6), Hellriegel in 1886 and Hellriegel and Wilfrath in 1888 in Germany proved the real worth of nodules. They grew leguminous and non-leguminous plants in sterilized soil of low nitrogen content until such plants became yellow and their growth and development arrested. Then they added a few drops of leachings from unsterilized soils where nodulated leguminous plants were growing. The legumes soon developed nodules and began to flourish, but the non-legumes were not benefitted and died. The final proof of the relation of bacteria to root nodule formation was put forward by the renowned Dutch bacteriologist Beijerinck, who in 1888 isolated the causative bacteria in pure culture and caused nodules to develop by artificial inoculation (6). Two years after the discovery of Hellriegel and Wilfrath, Salfeld, another German scientist reported that legume growth is benefitted by the transfer of soil from a field which had previously grown the same plant (3). Nobbe and Hiltner in 1890 showed the advantage of artificial inocu-

lation by adding pure bacterial preparations to seeds prior to planting. In fact they could be termed as the pioneers of the present inoculation industry (3). Following this epochal discovery numerous laboratory and field studies were started in U.S.A. and in Europe to know more and more about the activities of the microorganisms known as legume bacteria (30).

The legumes have varied uses. They may be used for hay, silage, seed, winter cover crops, and pasture. The fact that they are rich in high quality protein, well supplied with phosphorus and calcium, and are a good source of vitamins, especially A and D, make them as one of man's best food and as almost indispensable for economic livestock feeding (8). The protein in legumes is directly related to high nitrogen content and in this respect they differ markedly from non-legumes and grasses. For example, when the average protein content of one ton of each of eight legume hays was compared with the protein in eight grasses, the legumes averaged 304 pounds of protein per ton and the grasses 156 pounds (8).

Nearly all legume crops grown today are inoculated with commercial preparations of rhizobia to ensure maximum yield and high quality. For effective inoculation it is necessary to add proper bacteria of the cross-inoculation group concerned. Effective inoculation of legumes is a major factor in improving their yield and quality (3, 8, 9, 10, 20, 24, 25 and 31).

Erdman (8) reported that the more effective strains of legume bacteria can increase the yield or protein content of legumes as much as 20 percent on the average over the natural legume bacteria in the soil.

Allen (3) stated that large increases in crop yields from inoculation were obtained on soils low in available nitrogen. Gains from inoculation were less on fertile soils. On soils of average fertility increases in yield usually vary between 15 and 25 percent (3). The same worker (3) provided data for alfalfa in which the quality as determined by the protein content of alfalfa hay was shown to increase by 3.5 percent on the average as a result of inoculation over the non-inoculated plants.

Fellers (9) working with soybeans on the effect of inoculation, fertilizer treatment and certain minerals on the yield, composition and nodule formation found that inoculation with an efficient culture of Bacillus radicicola (as it was termed then) gave a substantial increase in the yield of total dry matter and of seeds of soybeans. He also observed an average decrease of three percent in the oil content and increase of seven percent in the protein content. No difference in the drying power of the oil extracted from the seeds of inoculated and non-inoculated plants were observed.

Fred and Graul (10), from their results of both pot and field tests at Winconsin, found that in acid soil low in  $\text{CaCO}_3$  both lime, in small amounts, and inoculation pro-

duced the best yield in soybeans. Also, the percentage of nitrogen in the crop and the total amount was increased by such treatment.

Lipman and Blair (20), found in pot culture experiments with soybeans that twice the amount of dry matter per pot was obtained from inoculated ones than from non-inoculated ones and 3.48 percent nitrogen from inoculated ones compared to 1.36 percent nitrogen from non-inoculated ones.

Norman (25), working with soybeans at Iowa, on Marshall silt-loam soil, found that inoculation increased the yield by 31 percent, significantly increased the protein content and decreased the oil content. On an area basis, the production of both protein and oil was substantially greater. The yield of mature straw was not appreciably increased by inoculation.

Moodie and Vandecaveye (24), found that in greenhouse inoculation with a suitable rhizobium produced a marked increase in the nitrogen content of both the straw and grain of mature plants of chick peas. In field trials they found the inoculated plants to be markedly greener and more vigorous than non-inoculated ones. The yield increased markedly as well as the nitrogen content of the tops and the root tissues (about three-fold increase in the nitrogen content of root tissues), on inoculated plots. In the mature grains from inoculated plots the protein content was increased by 5.4 percent.

Whiting (31) at Wisconsin showed that artificial inoculation improved the growth of peas in an acid silt loam soil on which peas were not grown for 11 years. Inoculation also increased the protein content and greater yield of number one and number two grades of peas.

Albrecht (2) working at Alabama on peanut inoculation in new peanut land (Norfolk sand) found both hay and nut yields to be favorably influenced by the application of lime and of fertilizer containing phosphorus and potash when inocula was applied to the seed. Duggar (7) also working at Alabama with Spanish peanut showed that artificial inoculation of unshelled seed resulted in an average increased yield of 30 percent per plant.

Wilson and Leland (34) at Ithaca, New York, conducted experiments to determine the value of supplementing the legume bacteria which the soil naturally supports with an artificial culture for alfalfa, red clover, beans and peas. Limed (pH between 7.42 to 7.68) and unlimed (pH 5.3) field plots representing only one type of soil were used. In case of the pea crop, plots which were limed as well as inoculated yielded 15.1 percent more in total crop and 35.8 percent more in dried peas than similar but non-inoculated plots. Unlimed but inoculated field plots gave an increased yield of 14 percent and 25.7 percent in whole crop and dried peas, than those from similar plots not receiving supplementary bacteria. The total yield from six plots which received supplementary bacteria was 14.3 percent



larger than the total yields from plots not so treated. In dried peas it was 29.3 percent in favor of supplementary bacteria. He concluded that a more general use of artificial cultures to supplement legume bacteria which the soil naturally supports can profitably be used, even though the soil may contain a considerably number of proper legume organisms.

A review of other experiments, however, indicate that inoculation may have little or no influence on the yield and quality of legumes. Moreover, at the same area inoculation in one year may favorably influence yield and quality of legumes, while in another year it may not, depending upon the conditions in which the inoculation is made.

Hofer (14) who carried out inoculation test on fifty pea fields in New York found that 44 percent of the field produced the same yield on the inoculated as on the uninoculated strips. On 20 fields the yield was increased by inoculation. The increases ranged from 10 to 55 percent. On the remaining eight fields the yields were reduced. The increases in yield consistently occurred in fields that had not grown peas for a long time. Yield reductions tended to occur most frequently at pH 6.6-6.8. The author concluded that reduced yields may be related to previous cropping history and reaction of the soil.

Halsey (11) at Florida grew southern peas of three growth-habit types supplied with nitrogen at the rates of

0, 18 and 36 pounds per acre. The seeds were treated immediately before planting with mixed strains of a cowpea group of legume bacteria at the rate of none, 0.3, or 0.6 pounds of inoculum per 100 pounds of seed. The response to the addition of seed inoculum was erratic depending on the rate of nitrogen fertilization. When nitrogen was added to the soil, there was a tendency for depression in yield when the seeds were inoculated at the normal rate of application and a lesser degree of depression at the twice-normal rate of application. When no nitrogen was added to the soil, there was no change in yield upon the addition of normal rate of seed inoculation over the non-inoculation treatment. The effect due to inoculation alone was not significant, nor was there a significant inter-relationship between nitrogen level and inoculation level.

Jones and Wade (16) in California conducted inoculation studies with garden peas over a period of four years at Davis. During the first two years the check plots significantly outyielded the inoculated but in the last two years there was no significant difference. The authors concluded that as peas had not been grown on the land previously, there is possibility that some of the native legumes may be host to the same strain of Rhizobium leguminosarum as the garden peas and may have supplied favorable bacteria in sufficient quantity to provide adequate inoculum.

Mention has been made of effective inoculation. Although the symbiotic association between the plant and the

bacteria in the nodules leads to the fixation of gaseous nitrogen, the actual amount of fixation in any one nodule varies widely (32). Under optimal circumstances nitrogen fixation may be rapid and continuous throughout the growing season in which case the association is effective. Under adverse conditions the period of fixation may be transient and little nitrogen may be fixed and the association is then ineffective. If artificial inoculation leads to effective association it is then termed as effective inoculation. Within one cross-inoculation group strains of bacteria vary in their power to form effective, ineffective, or intermediate associations. Usually this characteristic is the main factor that limits the value of symbiotic association. However, there are limiting plant factors which Nutman (as reported by Whyte et al, 32) terms "responsive" or "unresponsive" and which are determined by genetic factors. Erdman (8) and Wheeler (30) mention the existence of parasitic strains of legume bacteria. The parasitic or ineffective strains enter the root and form numerous small nodules, but fail to fix any nitrogen or otherwise benefit the plant. Low yields and even crop failures may be due to the nodulation of legumes by such poor types of rhizobia.

According to Whyte et al (32) nodulation pattern of an ineffective association has a characteristic appearance. Usually there is a very large number of small, hard, spherical, white nodules scattered over the whole root system

which tends to be long and ramifying with relatively few laterals. McKee (23) made similar observations in the case of birdsfoot trefoil. Allen (3) mentions the formation of numerous small, white nodules scattered over the lateral roots as a result of ineffective nodulation.

Leonard and Dodson (19) reports the appearance of ill effects among legume plants growing under optimum conditions due to another type of parasitism which is inherent in the invading organism. In such cases the organism enters the roots and forms nodules, but these are not beneficial. On the other hand, in effective inoculation the number and distribution of the nodules are relatively restricted (3, 8, 30 and 32).

Allen (3), reports that when desirable effective strains of rhizobia are added in inoculation they form large pink nodules on the central tap root system. According to Erdman (8) and Wheeler (30) the clustering of nodules around the tap root at the point where the inoculated seed is planted generally indicates that these nodules were formed by the bacteria added in the inoculant. Nodules scattered over the side roots are usually formed by the legume bacteria naturally present in the soil. Leonard and Dodson (19) found similar results. According to them nodules on effectively inoculated plants of Austrian winter peas tended to branch or become convolute, whereas, on most of the poor plants the nodules were smaller and globular in shape, the former type being placed generally around the

crown, and the latter scattered more or less evenly over the roots. Whyte et al (32) mentions that groups of large, pinkish, fleshy nodules of various shapes are clustered in the vicinity of the crown and the first formed secondary roots. These authors further mention that leghaemoglobin content (responsible for pink coloration) of effective nodules is closely related with high nitrogen fixing activity. Russell (29) mentioning Virtanen's work, reports that there is a good correlation between the amount of haemoglobin in the nodule and the rate of nitrogen fixation. Leghaemoglobin breaks down to green legcholeglobin (32) when nitrogen fixation ceases.

The discovery of ineffective strains of legume bacteria has put less credence in the number of nodules as a measure of the value of a legume inoculant. Numbers of nodules fail to tell the whole story of effectiveness of the bacteria in fixing nitrogen. It is necessary to measure plant growth responses, particularly mass, vigor, color and if possible total nitrogen content (8, 30).

In case of vetch, Henson and Schoth (13) have shown that inoculated plants are easily recognized by their greener color and more vigorous growth and nodules on their roots. Moodie and Vandecaveye (24) in field trials with chick peas found inoculated plants to be markedly greener and more vigorous than non-inoculated plants. Leonard and Dodson (19) found plants grown from seeds treated with beneficial cultures to be greener than those not so treated

in soils known to contain inefficient nodule producing organisms. Jordan and Gerrard (17) found that highest concentration of leghaemoglobin in field legume occur just prior to blossoming and determination of haemoglobin made at this critical period would enable one to make accurate comparison between various legumes and efficiency of nitrogen fixation.

Although the number of nodules is not a good measure of the value of a legume inoculant, in many instances effective nodulation have been found to be favorably influenced by inoculation, and inoculation and fertilization together. Albrecht (2) reports in case of peanuts (in new peanut lands in Alabama) the nodulation of inoculated plants were more abundant on fertilized plots than on unfertilized plots. Duggar (7) working with Spanish peanuts found increase in the number of both total and large nodules as a result of artificial inoculation. Moodie and Vandecaveye (24) found nodulation to be heavy in the inoculated plots of chick peas and not present at all in the non-inoculated plots. Norman (25) working on the effect of inoculation and nitrogen fertilization in soybeans found that 87 percent of all plants in the inoculated plots were nodulated; whereas no nodules were present on plants taken from uninoculated plots. Wilson and Leland (34) found that artificial inoculation supplementing natural bacteria in the soil produced a larger quantity of nodules in peas, than in those of the non-inoculated plots where there were

numerous small nodules.

Various factors may affect nodulation and nitrogen fixation. Conditions essential to the satisfactory growth of legumes must be fulfilled before maximum effective nodulation and high nitrogen fixation can be expected (8, 18). The carbohydrate/nitrogen ratio is important in controlling nodule activity (32). Any sudden change which may markedly restrict carbohydrate supply may induce changes in the nodules. According to Whyte et al (32) under conditions of a steady slow rate of photosynthesis the carbohydrate supply to the nodule is assured and if an effective symbiotic association is present, nitrogen is fixed at a rate which maintains the carbohydrate/nitrogen equilibrium. Any factor which increases the rate of photosynthesis may increase nitrogen fixation. Leonard (18), from his greenhouse test with certain varieties of soybeans, found that nodule formation is intimately connected with the carbohydrate-forming function of the plant so much so that nodules will form sparingly or not at all when insufficient light, carbon dioxide, or chlorophyll excessively diminishes the capacity of the plant to produce the proper carbohydrate.

Pate (27, 28) in nodulation studies on Pisum arvense and Vicia sativa found maximum nitrogen in red nodules just before flowering. Nodule losses in vetch was temporarily arrested by regular removal of flower buds. According to the author it may be that the nutritional demands of flowering may promote withdrawal of protein and

carbohydrate from vegetative organs with consequent starvation of underground portions of the plant which arrests root growth and promote rapid nodule destruction. However, he maintains that detailed analyses of the carbohydrate and nitrogen nutrition of flowering plants is necessary for a greater appreciation of the actual physiological processes involved in nodule destruction. If combined nitrogen is applied to legume as a fertilizer, the uptake of nitrogen by the plant may sustain such a narrow carbohydrate/nitrogen ratio that fixation is depressed; if this continues for long the nodules begin to degenerate and under such circumstances the presence of the nodules on the host is more deleterious than useful (32). The carbohydrate/nitrogen ratio also affects the development of nodules by the plant. A high level of combined nitrogen in the soil prevents formation of root hairs and hence entry of bacteria is precluded and no nodules formed (32).

Allison and Ludwig (4) working with alfalfa also expressed similar view that decreased nodulation in the presence of soluble nitrogenous salts is due to inadequate carbohydrate supply in the roots. Where nitrogen is abundant the carbohydrate synthesized is used for top growth and little is available for growth of roots or nodules. Halsey (11) working with southern peas in Florida found a depression in yield when the seeds were inoculated at the normal rate of application prior to planting and when nitrogen was added to the soil. Albrecht (1), however, in a study of ni-



trogen fixation as influenced by the nitrogen content of the soil through pot culture experiment, found that nitrate did not prohibit fixation. In case of cowpeas there was as much fixation in soils containing varying amounts of total nitrogen, as in one with 300 pounds of total nitrogen. Halsey (11), on the other hand reports Wilson's work who showed that some readily available nitrogen in cowpea may be beneficial to the plant as they allow the seedling to make a good start before fixation has a chance to occur.

Nodules can only fix nitrogen actively if the plant is adequately supplied with all the mineral elements essential for active growth (29). Phosphorus is important in symbiotic nitrogen fixation as well as in relation to earlier infection stages of nodulation (32). Molybdenum is also required in small quantities, for in soils very low in molybdenum the roots of legumes may be well nodulated but they fail to fix nitrogen (29). Also some species may have higher boron and sulphur requirement and a few have a higher calcium requirement (29).

Although legumes may grow successfully at a pH 4.5, nodulation tends to be greatly reduced below pH 5.5 (32). Brayn (5) found most favorable reaction for inoculation of soybean to be pH 6.5, the limit in which nodulation may occur being from pH 4.6 to 8. According to Wilson (33) soils may become unfavorable habitat for various groups of legume bacteria and that the bacteria may entirely disappear from the soil with increasing acidity. McKee (23) in

a study with birdsfoot trefoil seedlings found them to nodulate at pH values between 4.5 and 7.9, but satisfactory nodulation was only in the pH range of 6.2 to 7.5 Allen (3) mentioned that nodules disappear rapidly in very acid soils (below pH 6.0) and lack of aeration, water-logged conditions and shortages of necessary fertilizer elements such as potash, calcium and phosphorus and other necessary plant foods also account for their absence or fewer numbers.

McKee (23) mentioned the importance of moisture. Within limits as the moisture content of the soil increases so does the growth of the plant and adequacy of nodulation increases to reach a maximum at or near field capacity. Like soil pH and calcium availability, soil moisture affect the survival of rhizobia and subsequent nodulation. In this experiment (23) nodulation in soil of low average moisture content averaged poor to very poor (nodules mostly small, white and nonfunctional) with up to 55 percent of the plants still not nodulated 77 days after seeding. Check plants produced on soil kept at or close to field capacity were well nodulated at 37 days with numerous, large, pink, functional nodules. In a second test, seedlings produced on a fertile soil kept at or close to field capacity had large, pink, functional nodules 24-30 days after seeding. In contrast seedlings grown on the same soil at low average moisture content had not yet nodulated at 137 days after seeding, but subsequently did so when the soil moisture content was increased. The same worker (23) found that in-

oculated seed of birdsfoot trefoil planted in the same soil and subjected to cycles of drought may not nodulate adequately. Erdman (8) pointed out that nodules come and go with varying moisture levels in soils.

Russell (29) mentioned that nodules remain on the roots of many leguminous crops if the soil was kept moist and the first effect of the onset of drought was for the crop to shed its nodules. Wilson (35) presented results which indicate clearly that a reduction in moisture of a few percent for 24 hours from that which was present when nodulation occurred resulted in a shedding of nodules. Shedding of nodules occurred more readily in the small and fibrous roots than in the tap root. The importance of moisture is further explained by experiments of Masefield (22) on a clay soil at Oxford. He showed that irrigation increases the weight of plants in a dry season and number of nodules as well as size. The weights of nodules in the plants also increased. The same author (21) from a field survey of the nodulation of leguminous crops in Malaya found that in general nodulation was greatest where soils were wettest. Hofer (14) from experiments with canning peas in New York concluded that under neutral or alkaline conditions where manure is lacking may favor some agent (e.g. bacteriophage) which is detrimental to nodule bacteria. Russell (29) also mentioned that nodules can be short-lived through being parasitised by larvae of insects.

## MATERIALS AND METHODS

The experiment was conducted for two years during 1960-61 and 1961-62 seasons on the American University farm located in the Bekaa plain about 80 km. east of Beirut under dry land conditions. The soil is clayey, low in organic matter, nitrogen and phosphorus content, high in potassium, and is calcareous with a pH of 8.0. An area of land that had not been planted to legumes for several years was used. Austrian winter peas (Pisum sativum var. arvense.), Canadian field peas (Pisum arvense.), vetch (Vicia sativa.), and lentils (Lens esculenta), were used in this experiment.

Rainfall data of the farm revealed that the 1960-61 crop year was comparatively dry one for the Bekaa plain. The total rainfall for the year beginning from September 1, 1960 to August 31, 1961 was 284.9 mm. as compared to 219.0 mm. during 1959-60, 316.4 mm. during 1958-59 and 409.6 mm. during 1957-58..In 1961-62 the rainfall was 469.7 mm.

The soil was fertilized only with  $P_2O_5$  (in the form of superphosphate) at the rate of four kg. per dunum. The fertilizer was broadcasted and worked into the soil before planting. All the species of legumes were inoculated before planting with "Nodogen's", a commercial compound containing the appropriate rhizobium for the peas and vetch group. The seeds were inoculated just before planting by moistening with a little water and then shaking them in a container with a small amount of inoculum. Plantings were done with

a seed drill (Planet Jr.) with the hole so set as to get closest to the standard seed rates for the species concerned. The seeding rates used were: vetch 10 kg. per dunum, lentil four kg. per dunum, Austrian winter peas and Canadian field peas 10 kg. per dunum each. Planting was made on Nov. 12 and 13 in 1960 and on Nov. 11 and 12 in 1961.

The experiment was laid out on a simple split plot design involving four replications. In each replication, the main plot factor was inoculation vs. non-inoculation and species of legumes were the sub-plot factors. The sub-plot factors were randomized within the main plot. Non-inoculated strip in each replication served as check. A distance of 0.5 meter was left out between an inoculated and non-inoculated strip. On the outer side of each replication a distance of two meters was available for the purpose of examining legume root-nodules and also to allow for border. Each replication covered an area of 14 x 12.5 meters. In each replication six meters were available for forage, six meters for seed and the remaining two meters for nodule observation. Each of the four species in a main plot had six rows, each row 25 centimeters apart. Two rows on each side of a plot were discarded from experimental plot yield to reduce border effects.

Regular care was taken to reduce weed population so as to minimize competition between the legume species and weeds. Weeding was done with the regular nursery equipment. During the first week of May peas were sprayed with para-

thion to control aphids.

Number of plants per square meter, plant height at harvest time, percentage of moisture in forage, seed yield and kernel size were obtained and recorded. However, it was found that Canadian field peas were susceptible to winter-killing, hence percentage winter-killing was also recorded for this species. Because Austrian winter peas are very late maturing, in the first season they suffered from lack of moisture in the soil during later part of May. Consequently, there was very little or no seed formation in this species in 1961. A few seeds were formed which were harvested for nitrogen determination. For both forage and seed yields two samples from each plot, each from an area equivalent to two square meters, were harvested excepting in the case of seed yield in Austrian winter peas. The forage samples, which were harvested in the flowering stage were placed in cloth bags, labelled and allowed to dry in sunshine for about a month. They were then weighed and a representative sample taken for moisture determination and chemical analysis.

Observation for nodule incidence was made just at the initiation of flowering of the species concerned. A trench about 40 centimeters deep was dug in the appropriate place and then a few bucketfuls of water were added to the ground to moisten it so as to be able to lift the plants with the help of a fork with minimum of damaged roots and rootlets. In most cases the main roots were obtained intact

and the nodules were mostly concentrated on the main root, with a few and small nodules on the secondary roots. After lifting the plants the roots were carefully separated from adherent soil particles by a forceful stream of water. The tops were then severed and the roots kept in bottles containing five percent formalin solution for further study in the laboratory.

Nodules' size was measured under a compound binocular microscope fitted with a calibrated micrometer. On average, twenty roots were observed from each plot for counting the number of nodules and measuring their size.

For protein determination, a representative sample of each plot (forage or seed) was dried in an oven for six hours at a constant temperature of 70° C and then cooled before grinding. Oven dried samples were ground in a Wiley-mill with 40 mesh sieve and the ground material collected and stored in bottles. Before weighing (in the determination of protein), the ground samples were put in the oven at 70° C for several hours to remove the moisture, cooled in a dessicator and then weighed on the electrical analytical balance to the nearest tenth of a milligram. Protein determinations were made according to the modified Kjeldahl method as specified in Association of Official Agricultural Chemists' Official Methods of Analysis (15), to determine nitrogen percentage. The nitrogen values so obtained were multiplied by 6.25 to get the percentage of protein. Analyses were carried out in duplicates. Results

of duplicates differing from the sample mean by six percent or more were rejected and the determination carried out again.

All results relating to forage yield were reported on a 12.5 percent moisture basis.

The results of the experiments were analyzed using the analysis of variance method and the 't' test (26).



## EXPERIMENTAL RESULTS

A two-year experimental study was undertaken to determine the effect of inoculation and non-inoculation on the seed yield, forage yield and other characteristics of four leguminous crops. The year 1962 was more favorable than the year 1961 so far as total rainfall was concerned and a better crop was harvested. Canadian field peas, however, were more susceptible to winter-killing in 1962 (15 percent) than in 1961 (7 percent).

The data obtained from this study will be reported in Tables 1 to 11 under the following headings: seed yield, 1000 kernel weight, forage yield, percentage protein in the seed, total protein yield in the seed, percentage of protein in the forage, total protein yield in the forage, plant height, size and number of nodules, and number of plants per square meter. Also in Tables 1 to 11 means and LSD values have been given for the treatment inoculation vs. non-inoculation, and only for other significant treatments.

Analysis of variance values for the characteristics studied have been reported in the appendix in Tables 12 to 22. Table 23 gives the rainfall and average temperature at the University farm in the Bekaa' during the 1960-61 and 1961-62 seasons.

### Seed Yield

In both of the years under study inoculation re-

sulted in little, if any, beneficial effect on the seed yields as shown in Table 1. Yields from the inoculated and non-inoculated plots were 76.3 and 59.8 kg. per dunum for 1961 and 83.9 and 81.8 kg. per dunum for 1962, respectively.

In 1961 Canadian field peas yielded the highest amount of seed followed by vetch and lentils. In 1962 these three species did not differ significantly among themselves but each of them significantly out yielded the Austrian winter peas.

Only a few seeds were produced by the Austrian winter peas in 1961 as the species is very late in maturity and the rainfall was much below normal.

#### 1000 Kernel Weight

The weight of 1000 kernels was not significantly influenced by the inoculation treatment, in either 1961 or 1962 (Table 2). The kernel weight was lower in most of the legume species in 1961, except in the non-inoculated lentils, than it was in the more favorable year of 1962. Seeds of the Austrian winter peas were larger in 1961 than those in 1962, but both crops represent low and abnormal seed yields that may have influenced the 1000 kernel weight. The increase in seed weight in the other three species is evidently due to increased rainfall in the second year as shown in Table 23.

In both of the years the four legume species differed significantly among themselves as to the 1000 kernel

Table 1. Effect of inoculation on seed yield in kg. per dunum, of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	77.2	114.5	61.9	109.9
peas † (S2) Canadian field	----	16.4	----	15.8
peas (S3)	107.4	96.6	88.6	88.3
Lentils (S4)	44.4	110.9	29.0	113.0

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	23.2	43.3	37.3	68.4
Species	16.0	22.5	28.5	39.0

	1961			1962			
	I	Io		I	Io		
Mean	<u>76.3</u>	<u>59.8</u>		<u>83.9</u>	<u>81.8</u>		††
	S3	S1	S4	S1	S4	S3	S2
Mean	98.0	69.5	36.7	<u>112.2</u>	<u>112.0</u>	<u>91.0</u>	16.8

† Because of the lateness of the maturity of this species a few seeds were produced. The data was not included in the means of 1961.

†† Treatment means underlined by the same line do not differ significantly at 5% level.

Table 2. Effect of inoculation on 1000 kernel weight in grammes, of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	43.5	48.2	42.8	46.8
peas (S2) Canadian field	110.8	91.9	110.7	93.4
peas (S3)	144.1	165.5	141.9	162.8
Lentils (S4)	29.7	34.1	30.9	28.9

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	4.1	7.3	12.3	22.7
Species	6.0	8.2	8.0	10.9

	1961				1962			
	I		Io		I		Io	
Mean	<u>82.0</u>		<u>81.6</u>		<u>84.9</u>		<u>83.0</u> //	
Mean	S3	S2	S1	S4	S3	S2	S1	S4
	143.0	110.7	43.1	30.3	164.1	92.6	47.5	31.5

// Treatment means underlined by the same line do not differ significantly from each other at 5% level.

weight. Canadian field peas showed the highest weight per 1000 kernels, followed by Austrian winter peas, vetch and lentils.

### Forage Yield

The data for the forage yields of the four species of legumes studied are shown in Table 3. It will be noted that in any single year the yields are about the same for both the inoculated and the non-inoculated trials. In the cases of lentils, vetch and Austrian winter peas, the forage yields were almost double in 1962 of that in 1961. Also, there was an increase in yield in the Canadian field peas in 1962, but the increase was about 24 percent of that obtained in the previous year. The great increase in forage yields in 1962 was due to the higher rainfall that occurred. In 1962 a total of 469.7 mm. of rain was recorded at the University farm and 284.9 mm. in 1961, as shown in Table 23.

The two species, lentils and vetch, produced the greatest amount of forage in both years while Canadian field peas and Austrian winter peas yielded significantly less during the two seasons.

### Percentage of Protein in the Seed

The results of the protein analysis reported in Table 4 show that the percentage of protein in the legume seeds was not affected by the inoculation treatment. The

Table 3. Effect of inoculation on forage yield in kg. per dunum, of four species of legumes. (12.5% moisture basis)

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1)	255.3	579.4	282.1	533.3
Austrian winter peas (S2)	179.1	418.7	170.4	424.2
Canadian field peas (S3)	197.2	268.7	226.1	254.3
Lentils (S4)	292.2	580.8	264.8	570.4

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	49.3	90.1	59.3	108.8
Species	39.8	54.5	76.4	104.6

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>230.9</u>	<u>235.8</u>	<u>461.9</u>	<u>445.5</u>	<u>461.9</u>	<u>445.5</u>	<u>461.9</u>	<u>445.5</u>
Mean	S4 <u>278.5</u>	S1 <u>268.7</u>	S3 <u>211.6</u>	S2 <u>174.7</u>	S4 <u>575.6</u>	S1 <u>556.3</u>	S2 <u>421.4</u>	S3 <u>261.5</u>

// Treatment means underlined by the same line do not differ significantly at 5% level.

Table 4. Effect of inoculation on the percentage of protein in the seed, of four legume species.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	24.79	19.47	23.61	19.56
peas (S2) Canadian field	28.88	27.82	28.95	28.19
peas (S3)	27.57	22.31	27.70	22.89
Lentils (S4)	29.15	24.45	29.40	23.82

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	1.58	2.96	0.56	1.03
Species	0.93	1.27	0.68	0.93

	1961				1962			
	I		Io		I		Io	
Mean	<u>27.60</u>		<u>27.41</u>		<u>23.51</u>		<u>23.61</u> //	
Mean	S4	S2	S3	S1	S2	S4	S3	S1
	29.28	28.92	27.64	24.20	28.00	24.13	22.60	19.51

// Treatment means underlined by the same line do not differ significantly at 5% level.

mean protein percentages were 27.60 and 27.41 for 1961 and 23.51 and 23.61 for 1962 for inoculated and non-inoculated, respectively. It will be noted that during the drier year of 1961 the percentage of protein in the seeds was almost four percent higher than that obtained for the same legumes in the high moisture year of 1962. Russell (29) reported similar results in cases of wheat and barley, and indicated that such decreases are reflected in both the seed and in the straw.

The four legume species studied varied widely in the percentage of protein in the seeds. Only in 1961 the protein content was about the same in both the lentils and in the Austrian winter peas on the basis of mean protein percentage. The other comparisons indicate that the legumes studied vary significantly in this seed characteristic.

From the results shown in Table 4 the species can be arranged as follows in order of improved grain-quality as determined by protein percent: Austrian winter peas, lentils, Canadian field peas and vetch.

#### Total Protein Yield in the Seed

The total protein yield of the seeds was not influenced by the inoculation treatments in either of the two years under study as seen in Tables 5 and 16. In the first year the species differed significantly from each other in total seed protein per dunum. Canadian field peas yielded the greatest amount of protein (27.2 kg. per dunum) fol-



Table 5. Effect of inoculation on total protein yield of the seed in kg. per dunum, of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1)	19.2	22.3	14.6	21.4
Austrian winter peas / (S2)	----	4.0	----	4.5
Canadian field peas (S3)	29.6	20.9	24.7	20.4
Lentils (S4)	13.1	27.1	8.6	26.9

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	6.4	11.6	8.1	14.8
Species	4.8	6.7	6.1	8.3

	1961			1962			
	I	Io		I	Io		
Mean	<u>20.6</u>	<u>16.0</u>		<u>18.6</u>	<u>18.3</u>	//	
Mean	S3 27.2	S1 16.9	S4 10.8	S4 27.0	<u>20.7</u>	S1 21.8	S2 4.2

/ Deleted as explained in Table 1.

// Treatment means underlined by the same line do not differ significantly at 5% level.

lowed by vetch (16.9 kg. per dunum) and lentils (10.8 kg. per dunum). In 1962 lentils yielded the greater amount of total protein in the seed followed by Canadian field peas and vetch. Austrian winter peas were too late maturing to produce a satisfactory seed crop so the total seed protein was very low per dunum.

#### Protein Percentage in the Forage

The percentage of protein in the forage was not influenced by the inoculation and non-inoculation treatments as indicated in Tables 6 and 17. In the comparatively dry year of 1961 Austrian winter peas showed the highest percentage of protein in the forage with 23.85, and vetch the lowest, or 17.44 percent. Canadian field peas and lentils were intermediate in protein percentage with means of 20.52 and 19.63 percent, respectively.

In the second year, with a rainfall of 469.7 mm. as compared with 284.9 mm. in 1960-61 (Table 23), the forage yields were greatly increased, but the protein percentage in the forage was decreased (Table 6). It is well known that in seasons of high rainfall which produce high yields, the protein percentage in forage will be lower than in dry years and lower yields. The extent of decrease varied from species to species (Table 6). Thus, while in the first year Austrian winter peas ranked first in protein percentage, in the wet year of 1962, it was the lowest. In the 1962 season, Canadian field peas were the highest in protein percentage

Table 6. Effect of inoculation on the percentage of protein in the forage, of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	17.67	16.53	17.20	16.68
peas (S2) Canadian field	23.99	15.54	23.70	15.60
peas (S3)	20.85	18.10	20.19	17.79
Lentils (S4)	19.85	17.38	19.41	17.05

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	1.70	3.11	0.59	1.07
Species	0.94	1.29	0.64	0.87

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>20.59</u>	<u>20.13</u>	<u>16.89</u>	<u>16.78</u>	//			
Mean	S2 23.85	S3 <u>20.52</u>	S4 <u>19.63</u>	S1 17.44	S3 17.95	S4 <u>17.22</u>	S1 <u>16.60</u>	S2 15.57

// Treatment means underlined by the same line do not differ significantly at 5% level.

in the forage. Lentils and vetch were intermediate in this character and did not differ significantly from each other.

#### Total Protein Yield in the Forage

In both of the years inoculation did not affect the total protein yield of the forage in any of the four legumes (Table 7). Although the protein percentage in the forage was less in the 1962 crop than in 1961 the total protein yield per dunum was higher in 1962 for each of the four legumes. This was due to increased forage yield obtained during the more favorable growing season in 1962. In both years lentils yielded the highest amount of forage protein per dunum when compared with that obtained in the other species. In 1961 the three other species did not differ significantly from each other in total protein yield of forage. In the second season lentils did not differ significantly from vetch in their total protein content per dunum. The fact that winter injury reduced the yield of Canadian field peas is probably the reason that this species produced the least total protein per dunum in 1962.

From the two-year data, it seems that so far as the greatest amount of protein from forage per dunum is concerned the four legume species can be ranked as follows: lentils, vetch, Austrian winter peas and Canadian field peas.

#### Plant Height

Several authors (8, 13, 24 and 30) have mentioned

Table 7. Effect of inoculation on total protein yield of forage in kg. per dunum, of four legume species.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	45.2	95.0	48.6	89.3
peas (S2) Canadian field	43.0	65.0	40.5	66.2
peas (S3)	41.0	48.6	45.5	45.2
Lentils (S4)	58.2	100.8	53.6	97.1

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	10.8	19.8	10.7	19.7
Species	8.3	11.3	14.2	19.4

	1961				1962			
	I		Io		I		Io	
Mean	<u>46.8</u>		<u>47.1</u>		<u>77.4</u>		<u>74.5</u> //	
Mean	S4	S1	S3	S2	S4	S1	S2	S3
	55.9	<u>46.9</u>	<u>43.3</u>	<u>41.7</u>	<u>99.0</u>	<u>92.2</u>	65.6	46.9

// Treatment means underlined by the same line do not differ significantly at 5% level.

that one way to evaluate a legume inoculant is to measure plant growth responses particularly mass, vigor and greener color of the inoculated plants. In the present study no significant differences were found between inoculated and non-inoculated plants in mass (measured by forage yield, Table 3), or in vigor (measured by plant height, Table 8). Neither was there any difference observed in the green color of the inoculated and the non-inoculated plants.

The species varied widely in plant height at the time of forage harvest as shown in Table 8. Canadian field peas were the tallest in height followed by Austrian winter peas. Vetch and lentils were the shorter species in this study. It will be noted, also, that even the two seasons varied widely in total rainfall, the plant height was not affected greatly by the different growing seasons.

#### Size, Number and Coloration of Nodules

The size of the nodules and the average number of nodules on the tap roots of the legumes studied were not influenced by the inoculation and non-inoculation treatments as shown in Tables 9 and 10. In both of the years significant differences in size and number of nodules existed only between the species. Austrian winter peas and Canadian field peas produced the largest nodules on the tap roots followed by vetch and lentils (Table 9). Canadian field peas produced the largest number of nodules on the central tap roots, whereas the fewest nodules were

Table 8. Effect of inoculation on plant height in centimeters, of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	25.1	26.0	26.6	25.2
peas (S2) Canadian field	32.4	33.2	32.8	34.0
peas (S3)	43.8	45.3	42.4	45.4
Lentils (S4)	26.7	26.0	26.5	25.0

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	2.6	4.7	1.9	3.5
Species	2.5	3.5	2.3	3.2

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>32.0</u>	<u>32.1</u>	<u>32.6</u>	<u>32.4</u> //				
Mean	S3 43.1	S2 32.6	S4 <u>26.6</u>	S1 <u>25.8</u>	S3 45.3	S2 33.6	S1 <u>25.6</u>	S4 <u>25.5</u>

// Treatment means underlined by the same line do not differ significantly at 5% level.

Table 9. Effect of inoculation on size of nodules in millimeters, on tap roots of four species of legumes.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter peas	1.05	1.22	0.98	1.30
(S2) Canadian field peas	1.34	1.67	1.26	1.58
(S3) Lentils	1.25	1.42	1.27	1.43
(S4)	0.88	1.19	0.87	1.13

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	0.28	0.51	0.19	0.35
Species	0.11	0.15	0.12	0.16

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>1.13</u>	<u>1.09</u>	<u>1.37</u>	<u>1.36</u>				
Mean	S2 <u>1.29</u>	S3 <u>1.26</u>	S1 1.01	S4 0.88	S2 1.62	S3 1.42	S1 <u>1.26</u>	S4 <u>1.16</u>

// Treatment means underlined by the same line do not differ significantly at 5% level.



Table 10. Effect of inoculation on number of nodules on the central tap roots of four legume species.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	12	15	11	14
peas (S2) Canadian field	14	17	13	17
peas (S3)	18	21	18	20
Lentils (S4)	14	16	14	16

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	2	4	2	5
Species	2	3	2	2

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>14</u>	<u>14</u>	<u>17</u>	<u>17</u>	<u>17</u>	<u>17</u>	<u>17</u>	<u>17</u>
Mean	S3 18	S4 <u>14</u>	S2 <u>14</u>	S1 11	S3 21	S2 <u>17</u>	S4 <u>16</u>	S1 15

// Treatment means underlined by the same line do not significantly differ at 5% level.

found in vetch (Table 10).

Effective nodulation as a result of inoculation is determined by the distribution of the nodules on the central tap root system of legumes and the pink coloration of the nodules (3, 23 and 32). In the present study, in neither of the two years, was there found any difference in the manner of distribution of the nodules on the central tap root system or in their coloration.

Data in Tables 9 and 10 provide an excellent example of the effect of differences in soil moisture on rhizobial activity. The year 1961, which was comparatively dry, showed a lesser number of nodules per plant and a smaller size of nodules than those produced in 1962 when there was 184.8 mm. more rainfall. This is in close agreement with the results obtained by Masefield (21 and 22).

#### Number of Plants per square meter

The data in Table 11 reveals that the number of plants per square meter was the same in the inoculated and in the non-inoculated plants. By moistening the seed to coat them with the inoculant immediately prior to seeding did not affect the rate of planting. The increased number of plants per square meter in the second year for some of the species may be attributed to the more favorable rainfall during the germination period of the seeds in 1961-62 season.

Table 11. Effect of inoculation on number of plants per square meter of four legume species.

Species	Inoculated (I)		Non-inoculated (Io)	
	1961	1962	1961	1962
Vetch (S1) Austrian winter	286	317	286	306
peas (S2) Canadian field	128	145	116	142
peas (S3)	89	86	89	102
Lentils (S4)	408	392	401	410

	LSD 1961		LSD 1962	
	5%	1%	5%	1%
Inoculation vs. non-inoculation	26	47	20	37
Species	21	29	26	36

	1961				1962			
	I	Io	I	Io	I	Io	I	Io
Mean	<u>228</u>	<u>223</u>	<u>235</u>	<u>240</u>				
Mean	S4 405	S1 286	S2 122	S3 89	S4 401	S1 312	S2 144	S3 94

// Treatment means underlined by the same line do not differ significantly at 5% level.

## DISCUSSION OF RESULTS

The results obtained in this two-year study warrant some explanation. Although there was more rainfall in the second year, the rainfall was unevenly distributed throughout the whole growing season (Table 23). There were periods during March when the species of legumes suffered greatly from lack of moisture in the soil. Cycles of drouth seriously affect nodulation (23), with consequent improper functioning of the legume bacteria. This could be one reason why the inoculation treatment did not influence the characteristics that were studied.

According to Vavilov's concept of the origin of a plant species, the Mediterranean region (which includes Lebanon) is the center of origin of lentils, peas and vetch, (12). It is not unlikely that Rhizobium legumimosarum, the rhizobia specific to the cross-inoculation group which includes peas, vetch and lentils, is also present in this region. Moreover, the Mediterranean region is a very old civilized area which was cultivated for many years and the cultivation most probably included these legumes. Even though legumes were not grown on the experimental plot for eight years, the possibility of the existence of these bacteria in the soil of the experimental plots, may also be the reason why inoculation did not show any response. Jones and Wade (16) working in California with garden peas found no improvement in yield over non-inoculation, where they

suspected the presence of this bacteria in the soil. However, there are some experiments reporting good yields where inoculation was made to supplement the natural soil bacteria which, presumably, were insufficient.

Whether in the present experiment natural rhizobial population in the soil was sufficient or not, cannot be said with only two years of data. It would be desirable to conduct further experiments both under irrigation and dry-land conditions over a larger number of years to determine the effect of inoculation on the forage and grain yields and other characteristics in legumes.

## SUMMARY AND CONCLUSIONS

Four species of legumes were tried in a two-year study at the University farm in the Bekaa' to evaluate the effect of inoculation on seed yield, forage yield, 1000 kernel weight, percentage of protein in seed and forage, total protein yield in kg. per dunum from seed as well as from forage, heights of plants, size and number of nodules, and number of plants per square meter.

In neither of the two years there appeared to be any significant difference between the inoculated and non-inoculated plants in any of the characteristics studied. Species of legumes varied in most of these characteristics. Under the more favorable growing condition of 1962 with about normal amount of rainfall, vetch appeared to be the best in seed yield followed by lentil, Canadian field peas and Austrian winter peas. In forage yield lentils and vetch seemed to be the best, Austrian winter peas and Canadian field peas coming next.

The percentage of protein in the seeds was the highest in lentils and Austrian winter peas in the drier year of 1961. In the wetter year of 1962, however, Austrian winter peas gave the highest protein content in the seed with lentils coming second. In terms of total protein yield of the seed, lentils gave the highest yield in 1962, Austrian winter peas the lowest, Canadian field peas and vetch were intermediate.

In percentage of protein in the forage, Austrian winter peas were the highest in the drier year of 1961, but in the high moisture year of 1962 Canadian field peas were the highest with lentils coming second. However, in both the years lentils gave the highest total protein yield of forage with vetch being next best.

In the wetter year of 1962 the protein content in the forage and seed in all of the legumes was lower than that obtained in 1961, a drier year.

Seed yield, forage yield, total protein yield from forage as well as from seed, number of nodules and their size were greatly increased in each of the species by the greater amount of rainfall obtained in the second year than in the first year.

Untimely rainfall and the possibility of the presence of rhizobial species in the soil of the Bekaa, probably limited the efficiency of the added inocula. Additional work, both under irrigation and dry-land conditions for a longer period is necessary before any definite conclusions can be made.

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APPENDIX

Table 12. Analysis of variance for seed yield  
of four species of legumes. *///*

Source	1961		1962	
	D.F.	M.S.	D.F.	M.S.
Replication	3	1000.38	3	1867.56
Inoculation vs. non-inoculation	1	1630.20	1	34.65
Error (a)	3	329.80	3	1097.98
Species	2	7531.28 <i>///</i>	3	16623.49 <i>///</i>
Inoculation x species	2	7.71	3	24.47
Error (b)	12	216.32	18	733.56

*///* Denotes F values significant at 1% level.

*///* Only three species included in 1961.

Table 13. Analysis of variance for 1000 kernel weight of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	50.72	64.31
Inoculation vs. non-inoculation	1	1.70	29.38
Error (a)	3	13.51	120.29
Species	3	23269.19 //	28227.20 //
Inoculation x species	3	2.69	16.03
Error (b)	18	32.28	57.01

// Denotes F values significant at 1% level.

Table 14. Analysis of variance for forage yield of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	332.06	13627.85
Inoculation vs. non-inoculation	1	193.35	2137.57
Error (a)	3	1919.53	2773.47
Species	3	19196.24 //	169004.94 //
Inoculation x species	3	1521.79	932.73
Error (b)	18	1432.22	5282.36

// Denotes F values significant at 1% level.

Table 15. Analysis of variance for percentage of protein in the seeds of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	0.8833	0.9100
Inoculation vs. non-inoculation	1	0.2700	0.0800
Error (a)	3	2.0467	0.2467
Species	3	42.7767 //	99.6667 //
Inoculation x species	3	0.8967	0.5567
Error (b)	18	0.7767	0.4172

// Denotes F values significant at 1% level.

Table 16. Analysis of variance for total protein yield in the seed of four species of legumes. *///*

Source	1961		1962	
	D.F.	M.S.	D.F.	M.S.
Replication	3	69.30	3	110.73
Inoculation vs. non-inoculation	1	130.76	1	0.63
Error (a)	3	24.24	3	51.24
Species	2	546.94 <i>//</i>	3	777.62 <i>///</i>
Inoculation x species	2	0.12	3	0.68
Error (b)	12	19.49	18	33.35

*//* Denotes F values significant at 1% level.

*///* Only three species included in 1961.



Table 17. Analysis of variance for the percentage of protein the forage of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	4.62	0.7733
Inoculation vs. non-inoculation	1	1.76	0.0900
Error (a)	3	2.27	0.2733
Species	3	56.65 //	8.0733 //
Inoculation x species	3	0.04	0.1267
Error (b)	18	0.80	0.3667

// Denotes F values significant at 1% level.

Table 18. Analysis of variance for total protein yield of forage of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	20.35	281.93
Inoculation vs. non-inoculation	1	0.42	67.00
Error (a)	3	92.13	91.15
Species	3	322.61 //	4642.23 //
Inoculation x species	3	39.73	16.71
Error (b)	18	61.88	181.52

// Denotes F values significant at 1% level.

Table 19. Analysis of variance for plant height of four species of legumes.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	7.60	4.73
Inoculation vs. non-inoculation	1	0.02	0.40
Error (a)	3	5.23	2.92
Species	3	508.55 //	701.72 //
Inoculation x species	3	3.07	1.29
Error (b)	18	5.76	5.00

// Denotes F values significant at 1% level.

Table 20. Analysis of variance for size of nodules on tap roots of four legume species.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	0.3873	0.0298
Inoculation vs. non-inoculation	1	0.0131	0.0014
Error (a)	3	0.0062	0.0291
Species	3	0.3112 //	0.3310 //
Inoculation x species	3	0.0055	0.0117
Error (b)	18	0.0115	0.0121

// Denotes F values significant at 1% level.

Table 21. Analysis of variance for number of nodules on the central tap roots of four legume species.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	8.37	4.87
Inoculation vs. non-inoculation	1	2.00	3.78
Error (a)	3	3.08	4.86
Species	3	62.79 //	52.03 //
Inoculation x species	3	0.33	0.36
Error (b)	18	3.40	2.48

// Denotes F values significant at 1% level.

Table 22. Analysis of variance for number of plants per square meter of four legume species.

Source	D.F.	M.S. 1961	M.S. 1962
Replication	3	674.12	206.73
Inoculation vs. non-inoculation	1	171.12	200.00
Error (a)	3	519.46	321.27
Species	3	173273.46 //	164364.00 //
Inoculation x species	3	75.46	406.67
Error (b)	18	392.90	623.56

// Denotes F values significant at 1% level.

Table 23. Rainfall and average temperature at the University farm in the Bekaa during 1960-61 and 1961-62. †††

Month	Rainfall (mm)		Temperature(°C) Mean	
	1960-61	1961-62	1960-61	1961-62
September	0.0	3.0	21.2	18.7
October	9.0	5.9	18.7	16.9
November	33.3	41.4	12.7	11.1
December	37.1	138.5	9.3	8.0
January	76.1	93.1	5.6	6.3
February	58.9	130.6	5.1	5.1
March	43.1	10.5	7.5	10.9
April	25.6	43.3	12.9	11.3
May	1.8	3.7	18.1	17.0
Total	284.9	469.7		

††† Source: Bu-Shakra, S.S. and Malouf, F.M. American University farm meteorological data (Mimeographed).