SEED IMPERMEABILITY IN SPECIES OF LEGUMINOSAE GROWN IN LEBANON

Ву

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Hard seeds in Legumes

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ABSTRACT

From June, 1957 to October, 1958 a study was made of seed impermeability among 90 seed lots of leguminous plants normally considered as suitable for winter and 27 seed lots which may be planted in the spring months or during winter along the sea coast of Lebanon. The list included 38 species among 14 genera, all of which are in the subfamily Papilionoideae.

Among the lots which are normally grown in winter, seed impermeability remained pronounced for more than 25 days after harvest in samples of <u>Medicago tribuloides</u>, <u>Mosp.</u>, and <u>Melilotus indica</u>. Species with intermediate impermeability of seeds included <u>Lathyrus tingitanus</u>, <u>Lospo</u>, <u>Vicia ervilia</u>, one lot of <u>Vocativa</u> and 6 lots of <u>Vocativa</u>.

Among the lots normally planted in the spring, seed impermeability was persistent in lots of Lotus corniculatus, Melilotus indica, M. alba, Trifolium fragiferum, T. hirtum, T. incarnatum, T. pratense, T. repens, T. resupinatum, and 5 species of Medicago. Seeds of Pueraria thunbergiana also retained impermeability for a long period.

Seed impermeability was greatly reduced in several species by impaction using a 2-liter glass bottle in which the seeds were shaken for 10 minutes (90 bumps per minute). The most pronounced reduction was achieved with seeds of Medicago arabica, M. sp., Melilotus alba, Trifolium hirtum,

T. incarnatum and T. pratense.

Soaking in osmic acid (0.1 percent aqueous solution) for 12 to 36 hours caused blackening of permeable seeds, thus indicating permeability which correlated closely with tests in blotters to determine the percentage of germinable and permeable seeds.

The moisture content of mature seeds when tested for impermeability varied from 4.32 to 17 percent. The small-seeded species had a lower moisture content, on the average, than large-seeded species and likewise contained a higher percentage of impermeable seeds.

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INTRODUCTION

The <u>Leguminosae</u> is one of the larger families of seed plants ranking second only to the <u>Gramineae</u> in supplying many of the basic needs of man and herbivorous animals. As a source of portein in the diet of man and the nutrition of animals this family holds first place and in addition is the only important group of plants capable of nitrogen-fixation in soils in combination with symbiotic bacteria. Members of this family also provide raw materials for paints, industrial oils, varnishes, fibers, and tannins.

One of the distinguishing characteristics of leguminous plants is seed impermeability which in some species persists for long periods of time in the soil or in dry storage. This characteristic is of value in several ways. It aids in the maintenance of species of plants in a given area by the gradual loss of seed impermeability accompanied by annual appearance of new plants. Also impermeable seeds remain viable longer in storage than permeable ones. One disadvantage is that seeding rates must be adjusted on the basis of estimated germination of the impermeable seeds in any given year, a variable which cannot be controlled, hence it is often necessary to treat seeds so that they will germinate whenever soil moisture and temperature conditions are favourable.

Impermeability of seeds in the Leguminoseae family

has been known and mentioned in the literature since the time of Theophrastus (Third century B.C.). It is a condition characterized by the inability of certain seeds to absorb water even when soaked for short to long periods of time. Many investigators have studied seed impermeability in different countries using a large number of plant species but there are no published records of studies in Lebanon.

It has long been known that climatic conditions as well as the storage environment contribute to the development or loss of impermeability. Furthermore, plant species vary in the degree and duration of the condition.

Lebanon is a small country with a considerable variation in climatic environment. Along the seacoast, in the viscinity of Beirut, rainfall is heavy averaging about 98 centimeters annually for six months of the year extending from October, November to April or May. The relative humidity during the summer in spite of no rain averages about 70 percent. In the interior between the Lebanon and Anti-Lebanon mountain ranges the annual rainfall varies from 30 cm in the north to 95 cm in the south. At Rayak, near the American University Farm, the annual rainfall for 1952-57 was 66 cm and the relative humidity for the summer was 45 percent.

The Bekaa plain located in the intermountain area is only 60 kilometers from Beirut and at a distance of 80 kilometers from Beirut the American University has an experiments are conducted in the various phases of Agriculture.

The location of two areas with such climatic differences so close together provided a good opportunity to make a study of seed impermeability among indigenous, established and recently introduced species of the <u>Leguminoseae</u>.

The object of this study was to determine

- 1 The relative impermeability of seeds of indigenous or introduced species of leguminous plants in Lebanon.
- 2 The effect of environment on the degree and persistence of impermeability.
- 3 The period of maturation at which impermeability develops as measured by moisture content of the seed.
- 4 Impaction as a means of making impermeable seeds readily germinable.

REVIEW OF THE LITERATURE

Seed impermeability is best known among members of the <u>Leguminosae</u> but it is also prevalent in the <u>Malvaceae</u>, <u>Cannaceae</u>, <u>Geraniaceae</u>, <u>Chenopodiaceae</u>, <u>Convallariaceae</u>, <u>Convolvulaceae</u>, and <u>Solanaceae</u> families (9).

Interest in the behaviour of hard seeds of legumes in the laboratory and in the field is a continuing one. As early as 1894, Michalowski, (51) in Germany, perfected a machine which was designed to scratch the surface of the seeds in bulk lots in such a way as to cause the hard seeds to germinate as readily as others. Some three years later, Rostrup (73) in Denmark, treated seeds with sulfuric acid for the same purpose. By 1912, hard seeds were receiving attention in America. Love and Leighty (41), in New York experimented with acid treatment and Hughes (29) in Iowa, invented a scratching machine which he called a scarifier. It was soon followed by many other similar machines, some of which are in use at the present time.

The structure of the seed coats of legumes was studied by Pammel (63) as early as 1899, and some twenty years later by Coe and Martin (8) and independently by Lute (43). These later studies were for the purpose of learning why some seeds are hard while others are not. Hard-coatedness has been shown in some instances to be determined by a genetic factor, but environmental conditions also influence

the percentage of hard seeds which appear in any one crop. Collections of white sweet clover (Melilotus alba) vary as much as 98 percent in the number of hard seeds produced in different years from the same plants. In this plant hard or soft seed strains can be produced by selection and inbreeding (88). The same can be done for hairy vetch (Vicia villosa) (28). However, James (32) was unable to obtain any evidence of the inheritance of impermeability in crimson clover (Trifolium incarnatum) seed. In the first generation, selfed parents with a difference of 60.3 percent in seed coat permeability produced offspring with a difference of only 0.3 percent. James (33) then turned to a study of environmental factors as affecting the production of hard seeds in crimson clover, and found a correlation between sequence of flowering and amount of small seed produced, also between seed size and permeability. When conditions favored the production of small seeds they were hard. Middleton (52) used ten lots of seeds of Lespedeza stipulacea which were divided into first and second grade seeds on a commercial grading machine. The large seed fraction had an average germination of 77.3 percent and a hard seed content of 15.9 percent while the small seed fraction had an average germination of 54.9 percent and a hard seed content of 37.4 percent. When the seeds were dehulled and a finer separation made by screens in the laboratory the samples showed an even more marked difference.

Seed lots of <u>Trifolium pratense</u>, <u>T. hybridum</u> and <u>T. repens</u> were each separated into four color classes (78). In

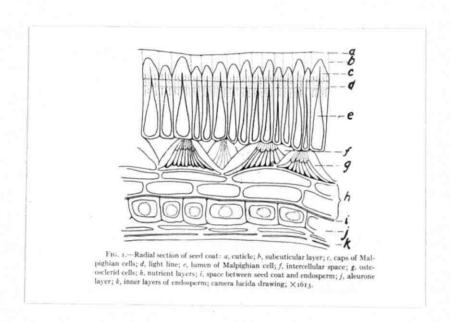
T. pratense, purple or purplish seed was heaviest and included most hard seeds. Yellow seeds germinated best in blotter tests. In T. hybridum black seeds were heaviest with yellow or yellowish classes being lowest in hard seed content. Intermediate colors were most numerous in most samples of the three species. Differences were greater in blotter than in soil germination tests. In pea and bean varieties, (19) no correlation was observed between smooth or wrinkled varieties and hard seed content. The sugar peas showed more hardshell than other classes. Legatt (40) tested Trifolium pratense and T. incarnatum at low and high altitudes. It was suggested that high altitude exerts its influence in this family by facilitating gas exchange through the seed coats.

The cuticle surrounding the seeds has received some consideration as a hindrance to water absorption. White (87) believed that the cuticular layer over the palisade cells or the coats determined the impermeability of small leguminous seeds, while in larger leguminous seeds the cuticle and a portion of the palisade cells combined to give the effect. According to Raleigh, (69) pectic substances change into water resistant substances as the seeds of the Kentucky coffee tree (Gymnocladis dioica) harden in ripening.

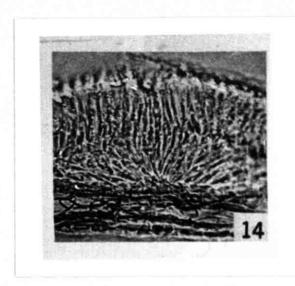
In the germination of sweet clover seeds initial water absorption was found to occur normally through the strophiole. The strophile which is usually recognizable near the hilum as a small elongated depression opposite the micro-

pyle, was found to be the region of the seed coat traversed by the vascular connections between the seed and funiculus.

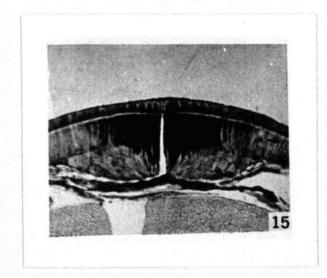
Hamly (22) used osmic acid as a stain to locate the areas in the seed coat where liquid could enter. Osmic acid (osmium tetroxide) is readily reduced through brown shades to black amorphous osmium when in the presence of active oxygen acceptors, such as unsaturated carbon compounds, the unsaturated fatty acids, and olefins. Von Altmann (3) has shown that the deep black staining occurs only in the presence of these compounds, and never with the saturated fats. (22) found out that the seed coat is bilaterally symmetrical about a plane extending through the center of the strophiolar region and hilum. On each side of this plane, the Malpighian cells are longer and narrower than normally is the case. On both sides of this, shorter Malpighian cells are found overlying loosely arranged cells, which show occasional linear arrangement. The lumina of the Malpighian cells when blackened by osmic acid clearly show that the cells have pulled apart forming a split along the plane of symmetry. split is characteristic of all sections that have stained at the strophiole, and it is without doubt the aperture by which liquids gain entrance to the soft seeds. Impermeable seeds showed no strophiolar discoloration and when sectioned there was no cleft between the Malpighian cells. On treating clover and alfalfa seeds with a fresh one percent aqueous solution of osmic acid, rapid penetration through any opening or permeable area takes place, as is evident from the



Reproduced from the Botanical Gazette, 92-93:346, 1932.



Unstained transverse section of strophiole of seed remaining hard after treatment with hydrochloric acid and ammonia; x 210.



Transverse section through strophiole of an osmic acidstained soft seed, showing elongated blackened Malpighian cells and cleft through which liquid entered; x 345.

Reproduced from the Botanical Gazette, 92-93:374, Plate V.

blackening in the lumina of the Malpighian cells where the osmic acid has been reduced by the unsaturated compounds present. The black stain develops in those cells around the openings in the seed coat before sufficient water has entered to produce visible swelling. After longer exposure to the solution swelling is noticeable, proceeding from the blackened areas until the whole seed is affected.

By a short osmic acid treatment therefore it is possible, not only to determine before hand whether a particular seed would or would not imbibe water, but also to identify areas of the seed coat which were permeable, and this without allowing the penetration of any liquid beyond the Malpighian layers of cells in the seed coat. The osmic acid reaction permits the classification of the soft clover and alfalfa seeds, according to the kind of permeable area, into the following groups.

- Seed scarified, showing irregular blackening around holes, cracks and abrasions in the seed coat.
- Seed softened by sulfuric acid treatment and subsequent maceration, presenting irregular permeable areas of variable sizes.
- Seed showing unlocalized permeable areas as the result of natural processes or the localized applications of heat.
- 4. Seed showing only a small permeable area at the strophiole, as a result of natural forces or heat.

Hamly (22) also found out that it was possible to open the cleft at the strophiole, in hard seeds, by moderate heating or by mechanical impacting or throwing against a hard surface. Porter (66) and Hutton and Porter (30) found impaction to be an effective method of making impermeable seeds permeable. Their findings substantiate the theories of Coe and Martin (8) and Lute (44) that impermeability is due to a condition of the cells, and refute the earlier views of Nobbe (61) and later beliefs of Nelson (59) that this was due to a waxy coating over the seeds.

Tests to determine the relation of moisture content to germinability and impermeability of seeds have been made by Hutton and Porter (30) by using seeds of Strophostyles helvola. Seeds with 57.87 percent moisture germinated 64 percent with no hard seeds, those with 43 percent moisture germinated 100 percent, and those with 20.18 percent moisture germinated 48.5 percent and contained 51.5 percent hard seeds. Seeds of Convolvulus arvensis were harvested at different stages of maturity by Brown and Porter (6). Seeds with 81 percent moisture were able to germinate at once, and when the moisture content was reduced to 13 percent, impermeability developed.

Methods of Reducing Seed Impermeability

Some of the methods of making hard seed coats permeable are:

1. Mechanical scarification

- 2. Concentrated and dilute sulfuric acid treatment
- 3. Impaction
- 4. Soaking in absolute alcohol
- 5. Liquid nitrogen treatment
- Burning holes in the seed coat with an electric needle.
- 7. Special temperature treatments
- 8. Hot water treatment
- 9. Pressure
- 1.- As stated before Hughes (29) in 1915, in Iowa invented a scratching machine called a scarifier. Today scarification to reduce the hard seed content is the only method of treatment which is used on a commercial scale. The most efficient machine is probably the Eddy Giant Huller and Scarifier which is equipped with carborundum discs that cause much less injury to seeds than sand paper, emery paper or a hard tile or cement surface. The other methods show promising results, but as yet have not been found adapted to use on a commercial scale. The disadvantage of severe scarification is that it breaks some of the seeds. As is generally recognized, this breakage takes place in both the seed coat and embryo. Injury to the embryo, which cannot be observed by an examination of the treated seed, results in broken sprouts when the seeds are germinated. It was also found that in germination tests of hard coated scratched legume seeds the experimental method employed influenced the percent-

age of abnormal sprouts produced (57). High moisture content of the seed bed increased and high temperature decreased the percentage of abnormal sprouts. Among the seeds tested abnormal sprouts were highest in <u>Trifolium Incarnatum</u> and considerable in <u>Medicago lupulina</u>.

- 2.- In the sulfuric acid treatment the seeds are put in concentrated or dilute sulfuric acid for a short time, then washed thoroughly with water and dried.
- 3.- In 1932, Hamly (22) described a new method for making hard seeds of sweet clover permeable. This consisted in shaking the seeds for ten minutes at three oscillations per second in a 500 ml corked Florence flask. Special mechanical impactors were also used. This treatment caused the formation of a strophiolar cleft through which water could enter. Hamly stated that the permeability of naturally soft seeds also occurred through the opening of a cleft at the strophiole. Scarification and sulfuric acid treatment. on the other hand, produced unlocalized permeable areas in the hard seed coat. The permeable areas of the coats were determined by osmic acid treatment. Hamly found out from his experiments that severity of impact is not necessary for seed softening in both hand shaking and mechanical impactions, it is important however, that enough impacts should occur so that at least one blow will strike at, or near, the strophiole. The mechanical impactors were faster and eliminated the uncertainties of hand shaking. In these machines, the seeds were dropped on a radially flanged revolving plate, and

were centrifugally thrown against a peripheral vertically ribbed wall. In each machine the revolutions per minute and the kind of flanging on the disk and ribbing on the wall differed in some respect, as did the efficiency per operation. Also he found in both hand and mechanical methods that softening is effectively carried out by light impacting. Heavy impacts are no more effective and are associated with scarification. While many blows are to ensure softening of the whole sample, the experiments indicate that softening is not a cumulative effect. On the other hand, the implication is that a large number of blows merely increases the chances of a single blow striking the strophiole. When this occurs with sufficient force to upset the stability of the tissue, which is in a state of tension, then a cleft appears between some of the long slender Malpighian cells, permitting the passage of water through it.

Porter (66) used the shaking method, described by Hamly, (22) to reduce the impermeability of Robinia pseudo-acacia. He shook approximately 100 grams of seed in a two liter glass bottle such as is used for shipment of nitric or hydrochloric acid. A second method of shaking was by using a thirty gallon steel drum mounted on a steel frame and equipped with a handle by which the drum could be slowly rotated. He found out that shaking seeds of black locust (Robinia pseudo-Acacia), either in a glass bottle or a steel barrel materially increased the percentage of permeable seeds. A sample of seed, which produced only 15 percent normal seed-

lings without treatment, produced 90 percent normal seedlings after being shaken for 20 minutes in the glass bottle. A larger quantity which was treated in the steel barrel for 20 minutes gave approximately 70 percent germination.

Porter (66) and Hutton and Porter (30) used the shaking test in the glass bottle for Lespedeza capitata, L.

virginia, Amorpha fruticosa. Their results show that seeds of L. capitata with 4.25 percent germination and hard seed content of 78 percent when shaken in a glass bottle for 10 minutes contained only 1 percent hard seeds and gave 83.5 percent germination. The same treatment applied to L.

virginia, which before treatment germinated 3.5 percent with 75 percent hard seeds, gave 33 percent normal seedlings and 34 percent hard seeds. A sample of seed of A. fruticosa before treatment produced 45 percent normal sprouts and had 25 percent hard seeds. The ten minute impaction treatment increased the percentage of normal to 67 percent and reduced the hard seed content to 0.5 percent.

In additional tests these investigators found the shaking method to be effective in making impermeable seeds of the following species permeable, namely Glycine max, Glycine usurriensis, Lathyrus odoratus, Pisum sativum, Vicia villosa.

If properly applied, impaction causes little or no injury to the seed, since it does not involve the use of acids, other liquids, sand or other abrasive materials. On the other hand it should not be anticipated that this princi-

ple can be applied to all species with impermeable seeds because Barton (5) tried this method and compared its effectiveness for members of three different subfamilies of the family Leguminosae. She found that seeds of the Papilionoideae were made permeable by shaking in a glass bottle for twenty minutes but seeds of the Caesalpinioideae with one exception, were not affected by shaking. The latter subfamily responded to soaking in absolute alcohol with transfer to water immediately after removal from the alcohol, which treatment was without effect on the Papilionoideae. Mimosoideae occupied an intermediate position, some being made permeable by shaking and some by alcohol soaking. The different responses of these two subfamilies to the two treatments indicate two types of causes of failure to absorb water. One, in the Papilionoideae, is the strophiolar cleft described by Hamly (22). The eight species of legumes for which Hutton and Porter (30) found shaking effective also belonged to this same subfamily.

4.- Verschaffelt (85) working chiefly with seeds of Gleditsia tricanthos concluded that the alcohol soaking permitted subsequent absorption of water by making a path through the coats which the water could follow. The alcohol was able to enter the integuments in the hilum region through interstices which water could not ordinarily pass. He showed, by using an aqueous solution of dyes, that water followed the path of the penetrated alcohol.

5.- Impermeable seeds of sweet clover have been made

to absorb water by plunging them in liquid nitrogen (-195.8°C) Barton (5).

6.- An electric needle (76) can be used in rendering the seed coats of large hard seeds permeable to water. The coats can be seared with the red hot point of this instrument with a minimum injury and there is consequently less danger of fungus attack than when the seeds are clipped or filed.

7.- Freezing has been found to reduce the number of impermeable alfalfa seeds (54). After the first freezing, however, subsequent freezing and thawing had little effect within the range of 0°C to -20°C. Busse (7) reported that freezing air dry impermeable seeds of sweet clover and alfalfa in liquid air (-190°C) made them permeable. Sweet clover seeds were kept in liquid air for 175 days without injury. Cooling to -80°C softened some of the impermeable alfalfa seed but had little effect on sweet clover seeds. Busse attributed the increased germination after freezing to formation of very tiny cracks in the impermeable seed coat. The very low freezing temperatures required for rendering hard sweet clover seeds capable of moisture absorption explains the ineffectiveness of freezing and thawing in soil plantings under natural weather conditions. Kamensky (36) found that the higher the water content of the hard seeds, the more effective was the freezing.

Rincker (71) applied dry heat treatments of varying temperatures and lengths of exposure, to alfalfa, sweet

clover, and red clover seeds. Within certain limits dry heat rendered impermeable seeds permeable and capable of immediate germination. By using the proper time-temperature relationship in treating alfalfa and red clover seed, the percentage of impermeable seeds was reduced as much as 81 for alfalfa and 69 for red clover, with a corresponding increase in germination. Impermeable seeds of sweet clover failed to respond very significantly to the same treatments. Heat treatments that cause impermeable seeds of alfalfa and sweet clover to become permeable do not produce any visible abnormalities in the resultant seedlings or plants. Heat treated alfalfa seed stored for seven months remained viable and exhibited the same effects of each treatment. These experiments indicate that a time-temperature relationship exists.

8.- Hutton and Porter (30) immersed seeds of Lespedeza capitata, Strophostyles helvola and Amorpha fruticosa in a water bath at temperatures of 65° and 85°C for one to eight hours. Exposures up to three hours at 86°C had no effect on permeable or impermeable seeds of Lespedeza capitata, but a five hour exposure reduced the percentage of hard seeds and increased the percentage of dead seeds of Strophostyles helvola. One or two hours at 85°C reduced the percentage of impermeable seeds of Amorpha fruticosa and two hours increased the percentage of dead seeds. Harding (23) recommended immersion of seeds of Acacia pycnantha and A. acuminate in boiling water for five seconds as the most satisfactory treatment and Wilson (94) found that a similar treatment for one

minute was effective for seeds of Robinia pseudo-acacia, R. hispida and R. viscosa.

9.- Davis (12) increased the germination of hard seeds of Medicago sativa and Melilotus alba 30 percent by exposing them to pressures ranging from 500 to 2000 atmospheres for five and ten minute periods. Rivers et al (72) found that pressures up to 30,000 pounds per square inch brought about germination of impermeable seeds of Cladrastis lutea. A pressure of 10,000 pounds at 25°C caused highest rate and final percent germination of the seeds of Ditremexa occidentalis. Seeds of Gymnocladus dioica germinated only after exposure to a pressure of 1,000 pounds at 50°C.

The weather factors effective in opening the coats of sweet clover, have been studied by Martin (47). He found that, in natural seeding in the field or in dry storage over winter in unheated open buildings in Iowa, 80-100 percent of the hard seeds softened by the middle of the following April. Although practically all of the openings of the seed coats to the absorption of water occurred in the interval from March 20 to April 20, a previous exposure of two months or more to fluctuations of temperature near the freezing point was required for effective softening. A constant temperature of 10°C as well as higher fluctuating temperatures were ineffective in making the seed coats permeable.

Martin (47) found also that hard seeds of sweet clover stored at constant temperatures -3°C, 2°C, 10°C and 15-25°C showed a maximum softening of 24 percent at the -3°C

and 2°C, and much less in the higher temperatures. Hard seeds buried in the soil at depths of 1, 3, 6 and 9 inches softened somewhat inversely proportional to their depth. Those buried at a depth of 30 inches where the temperatures seldom reached freezing showed no softening. The investigations showed that fluctuations of temperature in the realm of freezing are effective factors in softening hard seeds of sweet clover.

Work begun by Beal in 1879 and reported by Darlington (11) and Goss (20) has shown that seeds can survive long periods when buried in the earth. In Goss' experiments some seeds of red clover still remained hard after 20 years in the soil. In one lot 15.5 percent germinated when clipped.

samples of Melilotus alba and Vicia villosa. The samples were kept for 1-10 months in moist and dry storage under the following conditions: room temperature, 5°C, -10°C for one week followed by continuous storage at 5°C. At the close of the storage period for a sample, dead seeds and seeds which had produced radicles more than 1 cm long were discarded. The remainder of the sample was tested for germination on the basis of the original number of seeds in the sample as 100 percent. No low temperature storage treatment was found which would cause seed samples to give higher germination percentages than seed samples stored dry at room temperature. The various moist and dry-storage treatments caused softening of the hard seeds of Vicia villosa but did not soften hard

seeds of Melilotus alba.

The problem of evaluating hard seeds has received attention by many investigators during the past 25 years or more. As early as 1912, according to Pammer and Schindler (64), the union of Agricultural Experiment Stations, in Germany, required that hard seeds must be stated separately from the germination percentages in laboratory reports.

The interpretation of the plant producing value of hard seeds was first undertaken in America by Harrington (25) of the U.S. Dept. of Agriculture in 1916, at which time the following were recommended:

- For red clover, alsike clover, white clover and sweet clover when seeding late in fall, winter or early spring add all hard seeds to the germination.
- For alfalfa and crimson clover, add 2/3 of the hard seeds to the germination in all conditions.

Whitcomb (89), in 1921, separated hard seeds of alfalfa, sweet clover and red clover from permeable ones by means of subjecting lots of seeds to germination conditions in blotters for seven days. Hard seeds were tested in the laboratory and in pots set in soil in the field. From his results he concluded that during the first year alfalfa had the greatest value for the hard seeds (30-50 percent), red clover next (15-25 percent) and sweet clover least (10-15 percent).

Weihing (86) reported that samples of alfalfa seed

with less than 20 percent hard seeds germinated 61 to 64 percent in the field and samples with 20 to 62 percent hard seeds gave an average field germination of 57 to 60 percent. He concluded that samples with many hard seeds were practically equal to those with few or no hard seeds in field performance.

In 1928, Lute (44) reported observations and results of experiments in Colorado having to do with hard seeds in alfalfa during the period 1919-1927. These results are summarized as follows:

- Hardy varieties of alfalfa have a higher percentage of hard seeds than do less hardy ones.
- There was no consistant relationship between altitude and the production of hard seeds.
- 3. One half of the hard seeds of alfalfa became permeable during the first 3½ years of storage.
- 4. Dry heat at 80°C for two hours decreased the percentage of hard seeds in alfalfa and increased germination.

Legatt (39) decided, after many experiments, that impermeable alfalfa seeds are equal in value to permeable ones and that 50 percent of the hard seeds of sweet clover may be considered as germinable under Alberta conditions.

In 1938, Erickson and Porter (16) published the results of laboratory and field tests with a number of legumes in Iowa for the period of 1934-1937, and concluded with the following statement. "The work of other investigators,

coupled with the results herein reported, indicate that impermeable seeds of alfalfa should be considered as practically equal to viable, permeable seeds. Impermeable seeds of red clover and sweet clover may be taken to have 15-50 percent as much value as viable permeable seed.

MATERIALS, METHODS AND PROCEDURES

The seeds for this study were collected from different parts of the world namely United States of America, Cyprus, Iran, Turkey and Lebanon. Some of the seeds when received from outside Lebanon were not labelled or were labelled incorrectly hence it was necessary to identify them. Seeds that were in pods or burs were threshed and cleaned. Threshing was done by hand. Whenever the sample was large enough and the seeds were not too small, two plantings were made, one in Beirut on the American University campus and the other on the American University Farm in the Bekaa. Some of the seeds which were too small to be planted directly in the field or when the sample was small were first germinated in petri dishes on filter paper in the laboratory and then transplanted to the field.

For the Beirut planting the field was first freed of weeds. Then the rows were marked and shallow furrows made with 50 centimeters between rows. The seeds were planted in November 1957 in the furrows by hand and then were covered with a rake. A few lots were planted in the spring of 1958.

On the American University Farm, in the Bekaa, the field was first cleaned and smoothed. Then furrows were opened by a tractor using a cultivator with shovels. The seeds were planted in furrows 80 centimeters apart. Some of the seeds were planted in November, some at the end of De-

cember, some in the middle of March and others as late as May.

The plots were irrigated and weeded by hand whenever necessary. In the spring in Beirut the plants in a few rows were infested by aphids so the plants were sprayed with 0.1 percent metasystex.

Whenever the seeds of any row were matured, they were harvested and as soon as possible were threshed by hand.

After threshing the seeds were separated from the chaff by using an air blast separator. Moisture determination and germination tests were made on portions of each sample.

Some of the samples were infested with weevils when harvested. In these cases the seeds were fumigated by a mixture of carbon tetrachloride and carbon disulfide at the ratio of 4:1. Infested seeds were discarded.

Moisture determination

Apparatus

- 1. Constant temperature oven
- 2. Electrical balance
- 3. Stainless steel moisture dishes with covers
- 4. Dessicator

Procedure

The empty dishes were dried in the oven for at least 5 hours at 70°C and cooled in the dessicator. Whenever there was enough seed, duplicate samples of approximately 2 grams each were prepared and weighed. When the sample was small or

when the seeds were small, less than 2 grams were taken for moisture determination. The seeds were dried in the oven at 70°C for 48 hours, cooled in the dessicator and weighed again. The loss of weight was determined and the percent moisture calculated as follows:

Wi - Wo x 100 = Percent moisture

where Wi = Initial (wet) weight of seed

Wo = Oven dry weight of seed

DETERMINATION OF IMPERMEABLE SEEDS

Three methods were employed for the determination of the percentage of impermeable (hard) seeds in the samples studied, namely (1) soaking in water, (2) testing the germinability of the samples with a count for hard seeds, and (3) soaking in osmic acid.

1. Soaking in water

For some tests it was desirable to have 100 percent hard seeds. To separate the soft seeds from the hard ones the seeds were soaked in water. After 2 days the swollen or germinated seeds were removed and the hard seeds were dried for future use.

2. Germination tests

Apparatus

A germinator with temperature and humidity control Aluminum pans 27 x 18 x 4 centimeters Special blotters for germination

Procedure

The temperature of the germinator was 18-20°C and humidity was controlled. Two methods were used for germination.

- 1. Seeds which were small or flat were germinated between folded blotters. For each kind 100 seeds were used for the germination test.
- 2. Seeds which were rounded or big were germinated in aluminum pans. A single blotter was put in the pan, then 100 seeds were placed on top and covered with 2 layers of blotters. Since the relative humidity in the germinator is less than 100 percent the amount of drying of the substrata is materially affected by the amount of moist substrata or the number of tests. The blotters were checked every day and water added to them whenever necessary. After the sixth day the first count of the seeds was made. If the sample contained less than 5 percent hard seeds then the sample was taken out of the germinator and the percentage germination recorded. If the sample contained more than 5 percent hard seeds, the germinated seeds were removed and the non germinated ones put back in the germinator for further observation. In many cases the percentage of hard seeds became less than 5 percent after some time. Those whose seeds did not germinate 95 percent, even after a month and in some cases after forty days, were left until shortage of pans made it necessary to remove those seeds from the germinator. Basically it was considered justifiable to determine the hard

seed content of samples by the number of non-swollen seeds at the end of the period of time recommended by the International Rules for Seed Testing for determining the percentage of germinability for each kind of seed shown in Tables 1 and 2. This length of time varies from 7 to 14 days.

3. Osmic acid test

The osmic acid test for permeability as previously described was used in this study by immersing different seed lots in a 0.1 percent aqueous solution of the acid for different periods of time. Osmic acid dissolves slowly in water but presents no problem in the preparation of a week solution. Freshly harvested samples, seeds stored in the laboratory for different periods of time and those shaken as described in the discussion on impaction were given the osmic acid test. In small petri dishes $4\frac{1}{2}$ centimeters in diameter, seeds of a given kind were placed and osmic acid was added. The permeable seeds absorbed the acid and gradually turned black. When left overnight the entire seed coat surface of the soft seeds became black. By this test only the hard and the soft ones could be differentiated since the hard ones did not absorb any of the osmic acid and did not change color. When the seeds were left in the osmic acid solution for more than two days the seeds actually germinated in the solution.

To test whether osmic acid affected the results of germination in permeable seeds, different kinds of seeds were put in osmic acid overnight. The next morning the blackened seeds were transferred to folded blotters and put in

the germinator. After 6 days the first count was made. The results showed that seeds blackened by osmic acid germinated as well as those that were not put in osmic acid.

TREATMENT OF IMPERMEABLE SEEDS

As described in the review of the literature many methods have been used to reduce seed impermeability. In this study only the impaction method originally described by Hamly (22) was used. The reason for using only this method was that larger samples than those available are required by some methods, others have been used extensively by many investigators but the impaction method is a new development and is readily adapted to small quantities of seed.

Impaction test

Hamly's method of making hard seeds of sweet clover and red clover permeable consisted of shaking the seeds for ten minutes at three oscillations per second in a 500 ml corked Florence flask. This treatment caused the formation of a strophiolar cleft through which water could enter. An application of this principle to the studies herein reported involved the use of pint, quart or, ½ gallon jars (normally used for canning food) and a two liter glass bottle (such as used by Porter (66). These different kinds of jars were used to find out which would give the best results in softening hard seeds. From the same seed sample which contained 100 percent impermeable seeds, six subsamples of 200 seeds

each were shaken in the pint, quart and ½ gallon jars for ten and twenty minutes at 90 shakes per minute, and 200 seeds were shaken in the two liter bottle for ten minutes at 90 shakes per minute. After the shaking 100 seeds at random were put in osmic acid and the remainder were placed in blotters and put in the germinator.

RESULTS AND DISCUSSION

I. Samples planted in the Autumn of 1957

During the summer of 1957 seed samples were collected from cultivated and wild species of Leguminoseae that were found growing in and around Beirut as well as at the American University Farm in the Bekaa Plain. Observations were made as to the percentage of impermeable seeds by placing them between moist blotters in a germinator maintained at 18-20°C. At the same time seed samples from lots (1) stored in the seed laboratory, (2) obtained from countries outside Lebanon, and (3) harvested from field plots planted by the Agronomy section in the Bekaa were collected and planted in the fall of 1957 at two locations as described in the section dealing with materials and methods. from some lots did not germinate hence tests could be made only from lots which produced plants and seed. the fall planting were harvested in May. June and July of 1958.

The original plan was to harvest seeds at different stages of maturity, and determine the moisture content each time as well as the percentage of germinability. Unfortunately the oven used for moisture determination was small, it required repairs frequently and the number of samples was so large that it was impossible to properly process the immature lots hence only the mature seeds which could be held a few

days without immediate examination were studied.

At the time a sample was threshed the moisture content of the seed was determined and a portion (100 seeds) was placed in the germinator. It was decided to determine the number of germinable and impermeable seeds at the end of 7 days, replace the sample in the germinator, make a second and a third count and thus determine if seeds which were impermeable at the beginning of a test would gradually become permeable.

The data obtained from the autumn planting at the two locations are shown in Table 1. For each sample are given the sample number, botanical name, the date of harvest, percentages of moisture, total germination (normal seedlings), and impermeable seeds if any at three different periods of time. The samples with (a) following the numbers were planted in the Bekaa and all others were planted at Beirut near the seacoast.

A number of comparisons can be made between lots listed in Table 1. One comparison is that of moisture content of two or more lots of the same species, a second is that between samples from the same lot grown in two locations, a third is between samples of the same species but from different sources, and a fourth comparison may be made between different species of the same genus or even between genera.

Chickpeas (<u>Cicer arietinum</u>) are widely grown as a winter legume in many subtropical countries and especially in the Middle East. Five seed lots of this legume were ob-

Percent moisture, germinability and impermeability of mature seed samples harvested in 1958 from plantings the previous autumn at Beirut and in the Bekaa plain* Table 1.

Sample	Kind of seed	Date of	Percent	1t	Perce	intage of h	ard seeds
Number		harvest	Moisture	Total germ.	At 7 days	At 7 At 14-20 At days days mor	At 25 or more days
н	Cicer arietinum* (Iran)	14/7/58	9,33	06	49	0	0
CN.	C. arietinum (Iran)	14/7/58	96*8	06	г	0	0
ю	C. arietinum* (Iran)	14/7/58	10,60	06	0	0	0
4	G. arietinum (Iran)	14/7/58	9,45	66	0	0	0
ß	C. arietinum (AUB Farm)	14/7/58	9.13	36	0	0	0
9	Lathyrus cicera (Libya)	89/9/02	10,37	66	83	14	0
4	L. sativus (AUB Farm)	20/6/58	9.45	94	CQ2	0	0
7a	L. sativus (AUB Farm)	14/7/58	9.12	66	03	0	0
8	L. sativus (AUB Farm)	80/9/08	9.42	88	03	0	0
88	L. sativus (AUB Farm)	14/7/58	10,20	26	3	0	0
6	L. sativus (AUB Farm)	18/5/58	10,31	86	03	0	0
10	L. sativus* (Iran)	1/1/58	10,80	68	0	0	0
11	L. sativus (Iran)	1/1/58	10.26	86	0	0	0
11a	L. sativus (Iran)	14/7/58	10,30	66	0	0	0

Table I. Contid.

				-			
Sample	Kind of seed	Date of harvest	Percent Wolsture	Total	Perce At 7	Percentage of hard At 7 At 14-20 At days more	At 25 or more days
12	Lathyrus sativus (Iran)	1/1/58	17.6	66	1	. 0	
13	L. tingitanus (mottled)	19/2/28	10.70	78	42	24	16
13a	L. tingitanus (greenish)	10/6/58	8,38	46	81	49	90
14	L. sp*, **	13/6/58	10,70	83	16	ь	0
148	I. sp	10/6/58	9.65	74	39	30	24
158	Lens esculentum* (red)	86/6/58	11,93	88	44	18	0
168	L. esculentum (white)	26/6/58	10,50	66	88	42	0
17a	L. esculentum (Lebanon)	1/1/58	8.28	46	Q	0	0
20	Lupinus angustifolius var. Florida	18/5/58	10,10	100	74	03	0
218	L. termis (Lebanon)	14/7/58	8.71	100	65	0	0
27	Medicago sp. (Lebanon)	15/9/58	4.22	ю	66	46	26
28	M. tribuloides*, **	18/7/58	5,60	4.	88	78	92

more days At 25 or Percentage of hard seeds 94 94 93 0 0 0 0 At 14-20 days 93 96 96 0 0 0 days 93 6 18 96 0 0 Moistures Total germ. 95 16 86 94 96 88 90 26 100 97 91 Percent 7.65 8,50 7.20 9.75 9.05 7.00 0006 9,42 7.00 9.02 9,40 8.72 9.70 9,50 harvest Melilotus indica (Lebanon)29/7/58 89/1/68 29/1/58 14/8/58 14/8/58 16/7/58 10/1/58 16/7/58 10/1/58 Date of 10/7/58 16/1/58 86/6/28 16/1/58 86/6/28 M. indica (Lebanon) wild M. indica (Lebanon) wild arvense (winter pea) Pisum arvense (Turkey) arvense (Austrian winter pea) arvense (Austrian Winter pea) P. arvense (Turkey) Onobrychia sativa Kind of seed 学 arvense*, ** arvense*, arvense arvense 0. sp. ė. 히 Sample Number 37a 39a 388 40a 41a 30 36 35 38 40 32 37 31 41

Table 1. Contid.

more days Percentage of hard seeds 0 0 0 0 days 0 Q 0 0 0 0 0 days 26 0 0 Total germ. 86 100 98 86 95 06 100 87 90 66 Percent Moistures 9.44 8.88 8.88 8.88 9.70 8,60 90.6 10,25 8.50 10,32 10,25 9,05 Date of harvest 16/7/58 20/1/58 16/1/58 16/1/58 22/1/58 85/1/28 16/7/58 22/7/58 22/7/58 82/1/28 22/1/58 85/1/28 22/1/58 22/1/28 arvense (Canada field pea) U.S.A. pea) U.S.A. arvense (winter pea) arvense (winter pea) arvense* (Austrian arvense (Iran arvense (Iran (Iran) (Iran) (Iran) Kind of seed (Iran (Iran (Iran) winter pea) arvense arvense arvense arvense arvense arvense arvense ᆒ ونا e l ابم Sample Number 42a 448 43a 45a 47a 48a 49a 42 44 46 48 50 49

Table 1. Cont'd.

Table 1. Cont'd.

Sample Number	Kind of seed	Date of harvest	Percent Moistures	t Total germ.	Perce At 7 days	Percentage of hard At 7 At 14-20 At days mor	ard seeds At 25 or more days
518	P. arvense (Canada field pea) U.S.A.	14/7/58	9.52	93	0	0	0
52a	P. sativum var. Alaska (Cyplus)	26/5/58	10.48	86	0	0	0
64	Trigonella foenum-	1/1/58	00°6	86	7	0	0
65	T. foenum graecum	86/2/98	10,20	96	0	0	0
99	Vicia ervilia* (Lebanon)	18/6/58	10,32	82	4	0	0
66a	V. ervilia (Lebanon)	26/6/58	17,00	66	0	0	0
67a	V. ervilia	82/1/28	10.00	86	O3	0	0
89	V. ervilia (Iran)	18/6/58	9,25	80	38	21	18
69	V. narbonensis (Turkey)	14/7/58	9.75	86	Q	0	0
69a	V. narbonensis (Turkey)	14/7/58	10,35	86	Q	0	0
70	V. sativa (mottled) Lebanon	14/6/58	10,13	26	0	0	0
70a	V. sativa (mottled) Lebanon	26/6/58	9.03	86	03	0	0
71	V. sativa (greenish) Lebanon	14/6/58	10.50	ტ. ტ.	٦	0	0

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Sample		Kind of seed	f seed	Date of	Percent	t t	Perce	sntage of b	ard seeds
Number				harvest	Moistures	Total germ.	At 7 days	At 7 At 14-20 At 25 or days days more day	At 25 or more days
71a	·I	sativa Lebanon	sativa (greenish) Lebanon	26/6/58	9.03	86	01	0	0
72	١١	sativa	sativa (Turkey)	14/6/58	9,46	26	ю	0	0
73	اٰ⊲	sativa		14/7/58	10.15	66	7	0	0
73a	ا⊲	sativa		26/6/58	10,10	91	ч	0	0
74	اخ	sativa	(Lebanon)	10/6/58	10,64	26	0	0	0
748	اخ	sativa	(Lebanon)	26/6/58	10,40	96	C/3	0	0
75	ا⊲	sativa		10/6/58	8.98	94	03	0	0
75a	١٩	sativa		89/9/98	11,50	66	7	0	0
92	١٩	sativa	sativa**(Cyprus)	10/6/58	11,60	66	1	0	0
76a	١٩	sativa (Cyprus)	(Cyprus)	26/6/58	10.40	100	0	0	0
44	١ظ	sativa		18/6/58	10.00	86	03	0	0
77a	١٩	sativa		86/6/28	10,15	86	CQ.	0	0
78	*I	sativa (Iran)	(Iran)	24/7/58	9.37	100	0	0	0
62	١٨	sativa ((Iran)	22/5/28	11,13	66	п	0	0

Table 1. Cont'd.

Sample	K	Ind of	Kind of seed	Date of	Percent	t)	Perc	Percentage of hard seeds	ard seeds
Number				harvest	Moistures	Total germ.	At 7 days	At 14-20 days	At 25 or more days
808	V. sativa (Iran)	civa	(Iran)	26/6/58	10.29	74	40	32	24
818	V. sat	sativa		86/9/98	10,90	81	17	0	0
83	V. vi	llosa	villosa (Turkey)	14/7/58	07.6	84	58	17	15
82a	V. vi.	llosa	villosa (Turkey)	14/7/58	8,20	72	40	32	27
83	V. v13	villosa	(U.S.A.)	16/7/58	10,10	98	13	11	11
838	V. v1.	llosa	villosa (U.S.A.)	14/7/58	6.77	77	52	34	22
84	V. vill	llosa	villosa (A.U.B.)	16/7/58	10,70	88	18	11	11
84a	V. ville	Llosa	villosa (A.U.B.)	16/7/58	10.20	85	20	35	18
85	V. vil	llosa	V. villosa (Iran)	14/7/58	8,58	16	25	9	0
1									

* Numbers with (a) represent plantings in the Bekaa all others in Beirut

^{**} Diseased

^{***} Damaged by insects

tained and planted at Beirut. The moisture content at the time of harvest ranged from 8.96 to 9.45 percent which is not high nor is it exceptionally low. Possibly, if the seeds had been drier, impermeability would have been more pronounced. On the basis of the tests made, it appears that impermeability in this species is negligible and therefore of no practical importance.

Six different lots of Lathyrus sativus seed were obtained and planted in Beirut. Three of them were also planted in the Bekaa plain. The data in the table show that the moisture content varied from 9.12 to 10.80. In no case was the percentage of impermeability of any importance at the end of a 7 day test. It is possible that had the moisture content been lower the percentage of impermeability would have been higher. On the basis of the data obtained it appears that this species does not present any particular hard seed problem. It is also seen from the table that of the 3 samples planted in both locations there was no important difference in seed impermeability and in only one case, sample number 8 was there any important difference in the moisture content. It will be interesting to study this species further when the seeds have a much lower moisture content.

The sample of Tangier pea (<u>Lathyrus tingitanus</u>) was planted at the 2 locations. It is seen that the sample grown in the Bekaa had a moisture content significantly lower than the one planted in Beirut and a significantly higher percentage of impermeability.

Sample number 14 which was planted at both locations gave results similar to those of <u>L</u>. <u>tingitanus</u> in that the moisture content was significantly lower in the Bekaa than in Beirut and the impermeability was significantly greater. For both species the difference in impermeability on the basis of location may be due to the lower moisture content of the seed which was probably influenced by the drier atmosphere in the Bekaa.

Three species of Lens esculuntum were grown in the Bekaa. Two of them had high moisture content and the local one had a low moisture content, but the ones with the highest moisture content had also high impermeability. The low impermeability of the local sample even with low moisture content may be due to a varietal difference because in Lebanon people accept the lentils used for food which have a very low percentage of hard seeds; the growers plant varieties with seeds low in impermeability.

In the two samples of lupines that were tested the impermeability was quite high at 7 days but was lost at 14 days.

A wild species of <u>Medicago</u>, which grew in Beirut, was tested. The moisture content of harvested seed was the lowest in all the samples being 4.22 percent and the percentage of hard seeds was high. Even after 25 days it had 97 percent hard seeds. Seed of <u>Medicago tribuloides</u> also had high impermeability. Many species of Medicago are well known for high seed impermeability.

Three local wild samples of <u>Melilotus indica</u> were planted in Beirut and tested. Their moisture content varied from 7.20 to 8.50 and all three had a very high percentage of hard seeds which was persistent even after 25 days. Seeds of the genus <u>Melilotus</u> are noted for a high and persistent degree of impermeability.

Fifteen different lots of <u>Pisum arvense</u> from Turkey, United States of America, Iran and the American University Farm in the Bekaa were planted in Beirut and in the Bekaa. The moisture content of the seeds when harvested varied from 7.00-10.48 percent. In only 2 cases samples 41a and 43a both of which were planted in the Bekaa, and had intermediate moisture content was there a significant amount of impermeability but even in these samples the percentage of impermeability decreased very rapidly after 14 days.

Three different lots of <u>Vicia ervilia</u> one from Lebanon grown in Beirut and in the Bekaa, another from the American University Farm in the Bekaa and a third from Iran were tested. The moisture content was high in all the samples ranging from 9.25 to 17.00 percent. Sample number 66a had exceptionally high moisture content and among the 4 samples it had the lowest percentage of hard seeds which is what one might expect. Sample number 68 had the lowest moisture content and the highest percentage of hard seeds. A comparison of samples 67a and 68 shows a difference in moisture content of only 0.75 percent which probably is not responsible for the marked difference in the degree of impermeability. A vari-

etal difference is more likely responsible for the difference.

The sample of <u>Vicia narbonensis</u> from Turkey planted both in Beirut and in the Bekaa had a moisture content of 9.75 and 10.35 at each respective location with impermeability practically negligible.

Twelve different lots of <u>Vicia sativa</u> from Turkey,

Cyprus Iran and Lebanon were planted in Beirut and in the

Bekaa. The average moisture content was relatively high

compared to other species ranging from 9.03 to 11.60 percent.

In only two cases, sample numbers 80a and 81a, was there a

considerable amount of hard seeds, both samples had a

moisture content above the average compared with the other

lots of <u>Vicia sativa</u>. Impermeability in these lots may be

a varietal characteristic. The relatively high moisture

content of most of the samples of <u>Vicia sativa</u> may account

for the low percentages of impermeability. Further study of

this condition should be made.

Three different lots of <u>Vicia villosa</u> from Turkey,
United States of America and the American University Farm in
the Bekaa were planted both at Beirut and in the Bekaa. One
number was planted only in Beirut. The moisture content
varied from 8.20 to 10.70 percent. Sample number 82 which
had 9.70 percent moisture had only 16 percent hard seeds
after 25 days while sample number 82a grown in the Bekaa had
8.20 percent moisture and 27 percent hard seeds after 25
days. The difference in impermeability is not significant.
In sample number 83 and 83a the one with the lower moisture

content had the higher percentage of impermeability. Although the difference in the moisture content was less than 0.5 percent the sample harvested from the Bekaa had 52 percent hard seeds at 7 days and 22 percent at 25 days compared with the one harvested in Beirut which had 13 percent at 7 days and only 11 percent at 25 days. The difference in impermeability in the 2 samples from the same lot is not large and is probably not significant. A further study of this species is desirable when seeds are much drier.

Samples 84 and 84a harvested from Beirut and from the Bekaa respectively had a significant difference in hard seeds after 7 days but after 25 days the difference in impermeability was not significant. The sample of <u>Vicia villosa</u> from Iran had the next lowest moisture content (8.50 percent) but the lowest impermeability at 14 days and no impermeable seeds at 25 days. The reason for this low impermeability may be a varietal characteristic.

In summary it may be said that among the species of leguminous seeds listed in Table 1 those intermediate in impermeability are <u>Lathyrus tingitanus</u>, <u>Lens esculentum</u>, one lot of <u>Vicia sativa</u>, <u>Vicia ervilia</u>, and <u>Vicia villosa</u>. Those with a high degree of impermeability are 2 species of <u>Medicago</u> and 3 lots of <u>Medicago</u> indica.

II. Samples planted in the spring of 1957.

In addition to the samples listed and described in Table 1, many seed lots were planted during May and June

1958 and the seeds were harvested between July and September. The data obtained from the spring planting in the Bekaa plain are shown in Table 2. For each sample are given the sample number, the date of harvest, percentages of moisture, total germination (normal seedlings), and impermeable seeds at three different periods of time. Samples numbered 55 and 56 were planted in the fall in Beirut but since they were harvested with the ones planted in the spring the results are included in Table 2.

Two varieties of Lotus corniculatus were tested. The difference in moisture content was quite high one being 9.25 percent and the other 12.60 percent yet both had very high percentages of impermeability namely 97 and 98 percent and even after 25 days the percentage of hard seeds was 97 and 94 percent respectively.

One sample of Medicago arabica was tested. The moisture content was quite low and at 7 days it had 100 percent impermeable seeds and after 25 days 95 percent hard seeds.

Two samples of <u>Medicago hispida</u> were tested. The moisture contents were 6.62 and 7.15 percent and the impermeability was very high, 99 percent at 7 days and 92 and 99 percent respectively after 25 days.

One sample of <u>Medicago</u> <u>orbicularis</u> was tested. It had a moisture content of 7.25 percent, 100 percent hard seeds at 7 days and 99 percent hard seeds after 25 days.

Medicago sativa which had an intermediate moisture

Percent moisture, germinability and impermeability of mature seed samples harvested during Summer of 1958 from plantings made in the Spring of the same year at Beirut and in the Bekaa plain* Table 2.

Sample	Kind of seed	Date of	Percent	ıt	Perc	Percentage of h	hand apply
		narvest	Moisture	Total germ.	At 7	At 14-20	At 25 or
18a	Lotus corniculatus (USA)	USA) 10/9/58	9.25	10	46	00	a day
19a	L. corniculatus var.	18/9/58	12,60	03	86	96	9.6
228	Medicago arabica (USA)	18/8/58	6.67	ro	100	ď	u
238	M. hispida (USA)	10/9/58	7,15	ю	000	3 3	9 0
24a	M. hispida (USA)	10/9/58	6,62	, н	0 00	# O	N 0
258	M. orbicularis (USA)	10/9/58	7,25	7	100	0 0	a 0
268	M. sativa (Hairy peruvian) (USA)	10/9/58	9.38	64	99	20	3 8
28a	M. tribuloides (Lebanon)	10/9/58	6.70	15	68	ď	90
298	M. tribuloides (Lebanon)	10/9/58	6,20	4	100	8 8	0 9
	M. indica (USA)	18/8/58	11,000	11	86	72	22.
	M. alba (Hubam) (USA)	18/8/58	8,75	Q	100	86	86
	Trifolium alexandrinum	15/8/58	7.92	46	54	53	45
54a	T. alexandrinum (Lebanon)15/8/58	15/8/58	10,35	75	38	27	80

Table 2. Contid.

Sample	Kind of seed	Date of	Percent	t t	Perce	Percentage of hard seeds	ard seeds
Number		harvest	Moisture	Total germ.	At 7 days	At 14-20 days	At 25 or more days
55	T. alexandrinum	2/8/28	8.70	87	10	ω	0
56	T. alexandrinum	2/8/28	6,17	73	74	20	14
57a	T. fragiferum (USA)	15/9/58	9,15	03	86	86	86
58a	T. hirtum (USA)	19/9/28	9.65	Q	86	86	86
59а	T. Incarnatum (USA)	15/9/58	9.45	18	92	93	88
60a	T. incarnatum (USA)	15/9/58	11,50	49	84	74	20
61a	T. pratense (USA)	15/9/58	8.43	89	96	99	06
62a	T. repens (Ladino) (USA)	19/9/28	6.75	03	86	86	86
63a	T. resupinatum (USA)	19/6/61	10,65	19	93	88	81

* Numbers with (a) represent plantings in the Bekaaa Plain, all others at Beirut.

content had 66 percent hard seeds at 7 days and only 36 percent hard seeds after 25 days. This was expected because many investigators have reported that alfalfa loses its impermeability rather rapidly.

Two samples of M. tribuloides were tested. Both had a low moisture content (6.70 and 6.20 percent) and a high percentage of impermeability (89 and 100 percent respectively) at 7 days and 86 and 96 percent hard seeds respectively after 25 days. Apparently impermeability is persistent in this species.

Melilotus indica with a high moisture content of 11.00 percent had 92 percent hard seeds at 7 days and 72 percent after 14 days. When compared with 2 other samples of M. indica in Table 1 it is seen that samples 31 and 32, which had 8.50 and 7.20 percent moisture, had 96 and 97 percent hard seeds at 7 days and 94 percent after 25 days. It is evident therefore that the lower moisture content the more persistent is the hard seed content.

Melilotus alba had a relatively low moisture content and 100 percent hard seeds at 7 days and 98 percent at 25 days.

Four different lots of <u>Trifolium alexandrinum</u> were planted, two in Beirut and two in the Bekaa. The moisture contents varied from 6.17 to 10.35 percent and the impermeability was variable. The one with the highest percentage of hard seeds had an intermediate moisture content. Sample number 56 had the highest percentage of hard seeds at 7 days

but only 14 percent at the end of 25 days. Sample number 53a had the next lowest moisture content and the percentage of hard seeds was 54 percent at 7 days and 45 percent at 25 days.

A sample of <u>Trifolium fragiferum</u> was tested. It had 9.15 percent moisture and 98 percent hard seeds even after 25 days. The same was true for <u>T. hirtum</u> which had 9.65 percent moisture and 98 percent at the end of the 3 testing periods.

Two samples of <u>T. incarnatum</u> were tested. They had a moisture content of 9.45 and 11.50 percent respectively. The one with the lower moisture content had 95 percent hard seeds while the one with the higher moisture content had 84 percent hard seeds at 7 days and at 25 days the impermeability decreased to 50 percent. This decrease in impermeability in this sample may be due to its high moisture content because the one with the lower moisture content retained more of its impermeability being reduced from 95 to 82 percent.

A sample of <u>Trifolium pratense</u> had a moisture content of 8.43 percent, and the impermeability was 96 percent at 7 days and 90 percent after 25 days.

Trifolium repens which had a low moisture content of 6.75 percent, showed a hard seed content of 98 percent even after 25 days.

Trifolium resupinatum which had a relatively high percentage of moisture had 93 percent hard seeds at 7 days and 81 percent after 25 days.

In summary it can be said that seeds of small-seeded legumes presented in Table 2, with few exceptions, had very high percentages of impermeability and in most cases they retained impermeability after 25 days. Medicago sativa had an intermediate amount of hard seeds, and the samples of Trifolium alexandrinum were intermediate to low in the amount of hard seeds.

III. Effect of laboratory storage on imported seed.

One of the characteristics of leguminous seeds is the persistence of impermeability. This characteristic is valuable in several ways as it aids in the maintenance of species of plants in a given area by the gradual loss of seed impermeability accompanied by annual appearance of new plants. Also impermeable seeds remain viable longer in storage than permeable ones. One disadvantage is that the seeds must be treated so that they will germinate when planted in the field.

Persistence in impermeability was studied by using many kinds of introduced and native leguminous seeds which had previously been collected or imported and stored in the seed laboratory in Beirut. The samples were harvested in 1957 with the exception of two lots one of which is a wild Medicago sativa, probably harvested in 1954 and stored in pods in the laboratory for about 4 years. The other, Medicago tribuloides, was harvested about 1956 and stored in the laboratory for about 2 years.

Table 3 shows the origin, year harvested, date tested,

Table 3. Germinability and Impermeability of Imported Leguminous Seeds Stored in Beirut.

Kind of Seed	Origin	Тави	Data		0		
	0	harvested	tested	Normal	Abnormal	Dead	Hard
Desmodium tortuosum	USDA~USA	1957	24/10/58	1	4	17	0
Lathyrus hirsutus	USDA~USA		24/10/58	81	0	ю	16
Lotus corniculatus	Asgrow-USA	=	6/10/58	26	ы	12	88
L. comiculatus	USDA-USA	=		63	4	27	9
L. uliginosis	USDA-USA	=		53	ß	14	52
Medicago arabica*	USDA~USA	=	z	89	0	0	86
M. hispida	USDA-USA	=	=	80	CQ.	11	7
M. hispida	Peppard-USA	=	=	67	9	6	18
M. orbicularis	Peppard-USA	=	*	57	4	21	18
M. sativa*(wild)	Lebanon	1954	=	62	Q	ß	31
M. tribuloides	Lebanon	1956		38	CQ2	ч	29
Melilotus indica	Asgrow-USA	1957		64	ю	ß	12
M. indica (wild)	Lebanon			4	Q	ю	16
M. indica (wild)	Lebanon		=	1	0	ч	86
M. indica (wild)	Lebanon	=	=	ю	1	ю	83

Table 3. Cont'd.

Telana USA	Kind of Seed	Origin	Year	Date		Percent	ıt	
raria thunbergiana USA 1957 3/11/58 30 folium ambiguum USDA-USA " 6/10/58 58 fragiferum Asgrow-USA " 6/10/58 58 fragiferum USDA-USA " " 71 hirtum Peppard-USA " " 45 hirtum Peppard-USA " 45 Hybridum Peppard-USA " 93 incarnatum Asgrow-USA " " 95 subterraneum Asgrow-USA " " 95			harvested	tested	Normal	Abnormal	Dead	Hard
folium ambiguum USDA-USA " 6/10/58 58 fragiferum Asgrow-USA " 6/10/58 58 fragiferum USDA-USA " " 71 hirtum Peppard-USA " " 45 hirtum Peppard-USA " 45 Hybridum Peppard-USA " 93 incarnatum Asgrow-USA " " 91 subterraneum Asgrow-USA " " 91	Pueraria thunbergiana	US A	1957	3/11/58	30	0	CQ.	89
fragiferum Asgrow-USA " " 74 fragiferum USDA-USA " " 71 hirtum Peppard-USA " " 65 hirtum Peppard-USA " " 45 Hybridum Peppard-USA " " 93 incarnatum Asgrow-USA " " 91 subterraneum Asgrow-USA " " 91	Trifolium ambiguum	USDA-USA	=	6/10/58	99	ιΩ	10	27
fragiferum USDA-USA " " 71 hirtum Peppard-USA " " 1 hirtum USDA-USA " " 45 Hybridum Peppard-USA " " 95 Incarnatum Asgrow-USA " " 91 subterraneum Asgrow-USA " " 95	L. fragiferum	Asgrow-USA	=		74	4	ω	14
hirtum Pepperd-USA " " 1 hirtum USDA-USA " " 45 hirtum Peppard-USA " 45 Hybridum Peppard-USA " 93 incarnatum USDA-USA " " 91 subterraneum Asgrow-USA " " 95	l. fragiferum	USDA-USA		:	17	4	9	16
hirtum USDA-USA " 65 hirtum Peppard-USA " 45 Hybridum Peppard-USA " 93 incarnatum USDA-USA " 91 subterraneum Asgrow-USA " " 91		Peppard+USA		=	1	0	42	20
hirtum Peppard-USA " 45 Hybridum Peppard-USA " " 93 incarnatum USDA-USA " " 91 incarnatum Asgrow-USA " " 95 subterraneum Asgrow-USA " " 91	I. hirtum	USDA-USA			65	CN	п	32
Hybridum Peppard-USA " " 93 incarnatum USDA-USA " 91 incarnatum Asgrow-USA " 95 subterraneum Asgrow-USA " 91	[hirtum	Peppard-USA	=	=	45	2	O	41
Incarnatum USDA-USA " " 91 incarnatum Asgrow-USA " " 95 subterraneum Asgrow-USA " " 91	F. Hybridum	Peppard-USA			93	ю	Q	Q
incarnatum Asgrow-USA " # 95 subterraneum Asgrow-USA " # 91		USDA-USA	=	=	91	9	ю	0
Asgrow~USA " 91	[. incarnatum	Asgrow-USA	=	=	98	0	Q	ы
	T. subterraneum	Asgrow-USA	=	=	16	2	0	4

* The seeds were not threshed until the date tested.

germinability and impermeability of 25 samples of leguminous seeds. Twenty of these were imported from United States of America and 5 of them were collected locally. It is probable that, with the exception of Medicago arabica, all the imported seed samples had been scarified. The sample of M. arabica and a wild species of M. sativa, which was collected from Lebanon, had the seeds in pods until the date tested.

Desmodium tortuosum had no hard seeds when tested in October 1958. Lathyrus hirsutus had 16 percent hard seeds. Two samples of Lotus corniculatus were tested. had 6 and 29 percent hard seeds respectively. Lotus uliginosis had 52 percent hard seeds. A sample of Medicago arabica which was stored in pods until tested contained 92 percent hard seeds. Two samples of Medicago hispida were tested. They had 7 and 18 percent hard seeds respectively. A sample of Medicago orbicularis had 18 percent hard seeds. A wild species of Medicago sativa had 31 percent hard seeds in October 1958. The Medicago tribuloides which was probably threshed at the time of harvest in 1956 had 59 percent hard seeds in October 1958. A sample of Melilotus indica which was imported from the United States of America had only 12 percent hard seeds while 3 samples of M. indica harvested the same year in Beirut had 91, 93 and 98 percent hard seeds respectively. The low percentage of impermeability of the imported sample is probably due to scarification. A sample of Pueraria thunbergiana had 75 percent hard seeds. A sample of Trifolium ambiguum which was scarified contained

27 percent hard seeds. Two samples of <u>Trifolium fragiferum</u> were tested. They had 14 and 16 percent hard seeds respectively. Three samples of <u>Trifolium hirtum</u> were tested. They had 20, 32 and 41 percent hard seeds respectively. A sample of <u>Trifolium hybridum</u> had only 2 percent hard seeds. Two samples of <u>Trifolium incarnatum</u> were tested; one had no hard seeds and the other 3 percent. A sample of <u>Trifolium subterraneum</u> had 4 percent hard seeds.

In summary it can be said that leguminous seeds vary in their persistence of impermeability. Seeds of Medicago arabica in pods, of Medicago tribuloides, and Trifolium hirtum retain impermeability well in storage.

IV. Effect of storage on seed lots harvested in Lebanon.

Seventeen different samples of leguminous seeds which had a relatively high percentage of hard seeds at harvest time were tested again after 2-5 months to check the persistence of impermeability. The results are shown in Table 4.

Lathyrus cicera which had 83 percent hard seeds at harvest time had only 20 percent after 25 days. Lathyrus sp. which had 72 percent hard seeds at harvest time had only 17 percent 7 weeks later. L. tingitanus which had 49 percent at harvest time had only 18 percent 7 weeks later.

Two lots of Lens esculentum which had 77 and 92 percent hard seeds respectively at harvest time had no hard

Table 4. Comparison in impermeability of seeds at the time of harvest and after some time of storage in the seed Laboratory

Le tingitanus Le tingitanus Le tingitanus Le seculentum Le esculentum Medicago sativa Medicago sativa Medicago sativa Medicago sativa Medicago sativa Trifolium alexan Trifolium alexan Te alexandrinum Te alexandrinum Te incarnatum Te incarnatum Te incarnatum Te incarnatum Te incarnatum Te incarnatum Te incarnatum			/	0000	207 27000
Lathyrus cicera L. sp. L. tingitanus Lens esculentum Medicago sativa M. tribuloides M. indica M. indica Trifolium alexan Trifolium alexan Trifolium alexan T. alexandrinum T. alexandrinum T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum	Date	e tested	Percentage of hard seeds*	Date tested	Percentage of hard seeds*
L. tingitanus L. tingitanus L. esculentum M. sativa (hairy M. tribuloides M. indica M. indica Trifolium alexan T. alexandrinum T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum T. incarnatum T. incarnatum T. incarnatum	8	26/6/58	83	85/1/28	20
Lens esculentum Le esculentum Medicago sativa Medicago sativa Medicago sativa Medicago sativa Medicago sativa Trifolium alexan Trifolium alexan Te alexandrinum Te alexandrinum Te incarnatum Te incarnatum Te incarnatum Te incarnatum Te incarnatum	Š	26/6/58	72	7/8/58	17
L. esculentum Medicago sativa M. sativa (hairy M. tribuloides M. indica M. indica Trifolium alexan T. alexandrinum T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum T. incarnatum T. incarnatum	Č.	86/6/28	49	7/8/58	18
L. esculentum Medicago sativa M. sativa (hairy M. tribuloides Melilotus indica Trifolium alexan T. alexandrinum T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum T. incarnatum	8	86/6/58	44	7/8/58	0
M. sativa (hairy M. tribuloides M. tribuloides M. indica Trifolium alexan T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum T. incarnatum T. incarnatum	Š	80/6/58	86	7/8/58	0
M. tribuloides M. tribuloides Melilotus indica Trifolium alexan T. alexandrinum T. alexandrinum T. incarnatum T. incarnatum T. incarnatum	_	17/12/57	09	7/8/58	43
	peruvian)	2/9/28	99	10/10/58	25
		11/8/58	98	10/10/58	56
		16/5/57	46	6/10/58	16
		16/5/57	86	6/10/58	93
타타타다		11/8/58	74	10/10/58	4
H H H		85/8/28	54	10/10/58	34
H H.		22/8/58	38	10/10/58	4
I.		22/8/58	95	20/10/58	06
		22/8/58	84	10/10/58	94
		14/7/58	72	20/10/58	56
83a V. villosa		16/7/58	777	20/10/58	22

* At the end of 7 days in blotters.

seeds at 7 and 8 weeks later.

A wild species of <u>Medicago sativa</u> which was probably harvested in 1954 and stored in pods until the time tested had 60 percent hard seeds in December 1957, 43 percent in August 1958 and 31 percent in October 1958 as shown in Tables 3 and 4. A sample of hairy Peruvian alfalfa which had 66 percent hard seeds at harvest time showed only 25 percent hard seeds 5 weeks later.

Medicago tribuloides which had 86 percent hard seeds at harvest time had 56 percent hard seeds 2 months later.

Two samples of <u>Melilotus indica</u> which had 97 and 98 percent hard seeds respectively at harvest time still had 91 and 93 percent hard seeds respectively 5 months later. The difference in the hard seed contents is not significant so it can be said that <u>M. indica</u> had the same hard seed content at the 2 dates tested.

Three samples of <u>Trifolium alexandrinum</u> were tested. They had 74, 54 and 38 percent hard seeds respectively at harvest time and 4, 34 and 7 percent hard seeds respectively $6\frac{1}{2}$ and 8 weeks later. The reason the second sample retained its impermeability longer may be a varietal characteristic.

Two samples of T. incarnatum had 95 and 84 percent hard seeds at harvest time respectively and 90 and 76 percent hard seeds after 7 weeks.

Two samples of <u>Vicia villosa</u> which had 72 and 77 percent hard seeds respectively at harvest time had only 26 and 22 percent hard seeds about 3 months after the ini-

tial test.

The data in Table 4 indicate that in certain species the decline in impermeability is rapid as shown by samples numbered 6, 14, 15a, 16a, 54a and 56. In other species represented by numbers 31, 32, 59a and 60a the decline is much slower. In a third group may be found certain species which are intermediate in the rate of decline in impermeability as shown by numbers 13a, 26a, 28a, 82a and 83a.

V. Measurement of Impermeability by Osmic acid and germination tests.

Determination of impermeability in seed lots by the osmic acid test has received only limited attention by botanists. The method requires a minimum of a few hours and a maximum of 24 to 36 hours. The reliability of the method in comparison with other methods has not been determined, hence a series of comparative tests were made using the osmic acid test and the germination test in blotters.

Twenty one lots of leguminous seeds with a high percentage of impermeability were selected. From each lot 200 seeds were taken and subdivided at random into 2 subsamples of 100 seeds each. One subsample was placed in a 0.1 percent solution of osmic acid for 24 hours and the other was placed between moist blotters in a germinator at 20°C and kept for 7 to 14 days, the number of days being the requisite recommended in the International Rules for Seed Testing for those kinds listed.

The results of the comparison are shown in Table 5 from which it may be noted that remarkably close agreement was obtained. In no case was the difference in hard seed content significant.

To check whether osmic acid had any effect on the germination of seeds, different lots of seeds were put in osmic acid overnight. The next day the blackened seeds were removed from the osmic acid and put in a germinator at 20°C and kept there for 7 days. The results showed that the blackened seeds germinated as well as those that were not put in osmic acid.

The advantage of using the osmic acid method is that it is much quicker as it requires a maximum of 36 hours while the germination in blotters requires a minimum of 7 days.

The disadvantages of using osmic acid are (1) both the liquid and its vapors are poisonous when handled or inhaled; (2) osmic acid cannot be used with dark colored or black seeds since it would be difficult to detect the difference between natural color of the seed and the black color due to action of the acid; (3) the osmic acid test is expensive except for small seeds.

VI. Effect of Impaction on seed impermeability Fourteen different lots of seed which were tested previously and found to contain a high percentage of hard

seeds, were used in the shaking test. As described in the

Impermeability of leguminous seeds as determined by the osmic acid and germination tests, using 100 seeds per sample. Table 5.

Sample	Kind of Seed	Date Tested	Percent Impermeable by Osmic acid Blotters test	rmeable by Blotters	Percent normal* seedlings in blotters	Days Final count
18a	Lotus corniculatus (USA)	18/9/58	95	76	٤	12
228	Medicago arabica (USA)	11/9/58	100	100	0	4
23a	M. hispida (USA)	11/9/58	100	66	1	**
25a	M. orbicularis (USA)	18/9/58	66	96	4	i
268	M. sativa (halry peruvian) (USA)	11/9/58	63	99	34	7
288	M. tribuloides (Lebanon)	18/9/58	86	06	10	1
298	M. tribuloides (Lebanon)	11/9/58	92	86	63	1
27	M. sp.(Wild)(Lebanon)	11/9/58	100	46	ю	1
30	Melilotus indica (wild) (Lebanon)	11/9/58	46	93	7	14
31	M. indica (wild) (Lebanon)	11/9/58	66	26	ю	14
32	M. indica (wild) (Lebanon)	11/9/58	86	26	ю	14

Table 5. Cont'd.

Sample	Kind of Seed	Date	Percent Imp	rmeable by	Percent normal*	Days
Number		Tested	Osmic acid Blotter test	Blotters	seedlings in blotters	count ISTA**
33a	M. indica (USA)	11/9/58	94	86	8	14
348	M. alba (Hubam) (USA)	18/9/58	100	98	Q	4
53a	Trifolium alexandrinum	11/9/58	09	63	37	7
548	T. alexandrinum (Lebanon)	11/9/58	45	41	29	4
58a	I. hirtum (USA)	18/9/58	86	86	œ	0
59 a	T. Incarnatum (USA)	11/9/58	94	. 98	2	4
60a	T. incarnatum (USA)	11/9/58	98	85	15	4
618	T. pratense (USA)	11/9/58	95	86	62	7
62a	T. repens (Ladino) (USA)	18/9/58	100	86	Q2	7
63а	T. resupinatum (USA)	18/9/58	95	94	9	4
			The state of the s			-

* At the end of normal period of germination as indicated in the international rules for seed testing.

** ISTA * International Seed Testing Association.

*** Samples kept in the germinator for 7 days.

section on materials and methods the same type of bottle described by Porter (66) was used for this test and the seeds were shaken for 10 minutes. Only impermeable seeds were used and after the shaking the percentage of hard seeds was determined by 2 methods namely the osmic acid method and germination test in blotters. In the osmic acid test the seeds were observed at 2 periods, one after 24 hours and the other after 36 hours. The results are shown in Table 6.

It was found a few hours after the seeds were put in osmic acid that some of the permeable seeds began darkening and turning black while others took a longer time. It is evident that the shaking in the bottle made cracks in the seed coat of some seeds so that after the shaking the acid entered through the abrasions while in others only the strophiole was opened. In the latter case the seeds required a longer time to absorb the acid and the strophiole blackened first.

Medicago arabica seeds gave a good result by showing 78 percent permeable seeds in osmic acid with a germination of 81 percent in blotters. Seeds of Medicago hispida showed 12 percent permeable seeds in osmic acid and germinated 11 percent in blotters. Seeds of Medicago orbicularis showed 2 percent softening in osmic acid and germinated 3 percent in blotters. Wild Medicago sativa seeds showed 54 percent permeable seeds in osmic acid and ge minated 50 percent in blotters. Seeds of Medicago sp. showed 62 percent permeable seeds in osmic acid with a germination of 58 percent in blotters.

Table 6. Effect of shaking impermeable seeds in a 2 liter bottle for 10 minutes using 100 seeds

Sample	Kind of seed	+ +	permeable	Percent ge	germinable by	blotter	er test
Number		After 24 hours	After 36 hours	Normal seedlings	Abnormal seedlings	Dead	Imper-
	Medicago arabica	53	78	81	0	0	19
24a	Medicago hispida	10	12	11	0	0	68
25a	M. orbicularia	03	cα	ю	0	7	96
	M. sativa (wild)	45	54	20	QZ	ю	45
27	w sb.	37	629	58	0	0	42
28	M. tribuloides	50	37	37	0	0	63
34a	Melilotus alba (Hubsm)	73	73	70	0	Q	28
30	M. Indies	1	1	0	0	0	100
31	M. Indica	0	0	Q	0	0	86
57a	Trifolium fragiferum	12	22	80	-1	0	42
58a	T. hirtum	45	80	96	0	0	14
59а	T. incarnatum	95	95	93	ю	O.	Q
61a	T. pratense	88	92	96	0	н	ю
63a	T. resupinatum	12	15	50	0	0	8

Seeds of Medicago tribuloides showed 37 percent softening in osmic acid and germinated 37 percent in blotters. Hubam sweet clover seeds showed 73 percent permeable seeds in osmic acid and germinated 70 percent in blotters. Two samples of Melilotus indica seed were tested. The seeds showed 1 and 0 percent permeable seeds respectively in blotters. Failure of Melilotus indica seeds to soften by shaking may be caused by the hulls which adher to the seeds very firmly and when threshed some parts of the hulls remain attached to the seed. If those remaining parts are near or on the strophiolar region then the shaking may not open up the cleft in the strophiole and the seeds would remain hard. Further studies on this species are needed. Trifolium fragiferum, softened 22 percent in osmic acid and germinated 20 percent in blotters. Trifolium hirtum seeds showed 80 percent permeable seeds in osmic acid and germinated 86 percent in blotters. Trifolium incarnatum seeds showed 95 percent permeable seeds in osmic acid and germinated 93 percent in blotters. Seeds of Trifolium pratense showed 95 percent permeable seeds in osmic acid and germinated 96 percent in blotters. Trifolium resupinatum seeds showed 15 percent permeable seeds in osmic acid and 20 percent in blotters.

It can be concluded from the data in Table 6 that the shaking is effective in softening the seeds of some species like <u>Trifolium pratense</u>, <u>T. incarnatum</u>, <u>Melilotus alba</u> and <u>Medicago arabica</u>. It has a slight or insignificant effect on species like <u>Melilotus indica</u>, <u>Medicago orbicularis</u>

and <u>Medicago</u> <u>hispida</u>. And its effect is intermediate with some species like <u>Medicago</u> <u>sativa</u>, <u>M. tribuloides</u>, <u>Trifolium</u> <u>fragiferum</u> and <u>T. resupinatum</u>.

Variability in the response of impermeable seeds to the shaking test as used in the experiments herein described is greater than was anticipated, inasmuch as all genera tested belong to the subfamily Papilionoideae. In the preceding discussion a suggestion was made concerning a possible reason why seeds of Melilotus indica were not made permeable. Seeds of Medicago tribuloides, and Medicago arabica are all characteristically reniform and the strophiole is so located on the incurved surface that it cannot receive a direct blow during the shaking process. In some cases the bumping may cause fissures at the base of the hilum, which permits entrance of water. Seeds of Medicago sativa are so variable in shape that many of them can be struck at the strophiole, whereas others are shaped as in Medicago arabica. Failure of seeds of Trifolium fragiferum and T. resupinatum to be made permeable by bumping is not readily explainable because seeds of other species of Trifolium do respond well. This phase of the study should be continued.

SUMMARY

In this study the degree of impermeability in 117
seed lots representing 14 genera and 38 species of
Leguminosae was determined. All species studied belong
to the subfamily Papilionoideae. Most if not all the
species can be grown along the seacoast of Lebanon during
winter when the rainfall is abundant. If irrigation water
is available some of the species can be grown in summer.
At higher elevations, such as are found in the Bekaa plain
(around 1000 meters) the major portion of the species that
were investigated can be planted in the fall or winter
months. Growth is slow until March but becomes rapid thereafter if rain comes or irrigation water is available. Some
species grow well in the summer with occasional irrigations.

Seed impermeability is a characteristic condition among members of the <u>Leguminosae</u> family and its persistence is more pronounced in some species than in others, often preventing germination at the time of planting and for long periods thereafter. There are no published reports of seed impermeability among leguminous plants grown in Lebanon, hence this study was undertaken to find out to what extent the condition exists, how long it persists in storage, and the moisture content of impermeable seeds at harvest time. In addition methods of determining and reducing impermeability in seed lots were included in the study.

The most common winter legumes that can be grown in the Bekaa plain are species of <u>Cicer</u>, <u>Lens</u>, <u>Lathyrus</u>, <u>Pisum</u>, <u>Onobrychis</u>, <u>Vicia</u> and <u>Medicago</u>. Members of all these genera can be grown in winter along the seacoast as well as species of <u>Lupinus</u>, <u>Lotus</u>, <u>Trifolium</u>, <u>Trigonella</u>, and <u>Melilotus</u>. During the summer months of 1958 the species that grew in the Bekaa and produced seed were members of the genera <u>Lotus</u>, <u>Medicago</u>, <u>Trifolium</u>, <u>Trigonella</u> and <u>Melilotus</u>. Plants of <u>Pueraria thunbergiana</u> grew luxuriantly but produced no seed.

The results of the investigations are summarized briefly as follows:

- 1.- The winter legumes in general were low in impermeability with the exceptions of Lathyrus sp., Lathyrus tingitanus, a wild species of Medicago, M. tribuloides, 3 lots of Melilotus indica and a sample of Vicia ervilia. The moisture contents varied from 4.22 to 17.00 percent. The sample having the lowest moisture content had 97 percent impermeable seeds after 25 days. This was a wild species of Medicago, native to Lebanon and impermeability is well-known among many members of this genus.
- 2.- Samples planted in the spring of 1957 were mostly from small-seeded species in which seed impermeability was high in most of them. The moisture content ranged from 6.17 to 12.60 percent, the average being 8.65 percent. Species with the percentage of impermeability 72 up to 99 included Lotus corniculatus, Medicago arabica, M. hispida, M. orbicularis, M. tribuloides, Melilotus alba, M. indica,

Trifolium fragiferum, T. hirtum, T. incarnatum, T. pratense, T. repens and T. resupinatum.

3.- Persistence of impermeability after harvest was variable. In some species the decline was rapid as shown by seeds of Lathyrus cicera, L. sp., Lens esculentum, Trifolium alexandrinum and Vicia villosa. The decline was relatively small in seed lots of Medicago tribuloides, Melilotus indica, and Trifolium incarnatum. Of special interest is a sample of wild alfalfa (Medicago sativa) which was harvested in 1954 and kept in the pods in the laboratory until December 1957 at which time the impermeability was 60 percent, which is unusual for this species.

4.- Soaking seeds in a 0.1 percent solution of osmic acid for 12 to 36 hours, caused blackening of permeable seeds, thus indicating permeability which correlated closely with tests in blotters to determine the percentage of germinable and permeable seeds.

5.- Shaking is effective in softening the seeds of some species like <u>Trifolium pratense</u>, <u>T. incarnatum</u>,

<u>Melilotus alba</u> and <u>Medicago arabica</u>. It has slight or insignificant effect on species like <u>Melilotus indica</u>,

<u>Medicago orbicularis</u> and <u>Medicago hispida</u> and its effect is intermediate with some species like <u>Medicago sativa</u>,

<u>M. tribuloides</u>, <u>Trifolium fragiferum</u> and <u>T. resupinatum</u>.

6.- The data obtained indicate that most of the winter species of leguminous plants which were studied present no particular problem of hard-seededness but many

small seeded species in the genera Melilotus, Medicago,

Lotus, and Trifolium require treatment before planting to

provide a high percentage of readily germinable seeds.

Temperature and Relative Humidity for 5 Summer Months in Lebanon 1952 to 1958***

				Temperature	bure			Re	a tive	Relative Humidity	
		May	June	July	Aug.	Sept	May	June	July	Augo	Sept.
1952	Seacoast*	20.6	23.3			86.9	69	75	72	73	7.1
	Bekaa**	16.4	20.0	22.7	24.1	24.4	20	44	41	35	35
1953	Seponest	19.8	1			24.5	72	74	44	94	17
	Bekaa	16,1	20,3	23.9	23,1	20.7	54	20	20	21	49
1954	Seponsest	21,8		28.6	29.2	27,3	02	20	29	99	63
	Bekaa	17.2	20.6	24.4	25.0	20.8	20	44	40	41	49
1055	+ a a c c a a c c	1.99	4	28.2		27.0	72	68	64	64	65
2	Bekaa	17.2	17.2	23,1	22.7	21,5	26	38	40	46	52
1956	Seacoast	21.0				87.8	20	99	67	89	19
	Bekaa	15,7	21.2	24,3	25,9		51	41	32	35	1
1957	Seacoast	21.6	26.		e	27.7	7.1	70	69	68	88
	Bekaa	13,8	19	22,6	23,1	21.7	64	46	40	35	42
1958	Seacoast	228	26			27.2	68	65	99	49	63
	Bekaa	15,8	19.4	22.4	23,6		44	42	36	38	ı
40				1							

taken from the weather station at the Observatory at the American University * The data for the seacoast for 1952 and 1953 were obtained from the weather station of the Beirut International Airport and from 1954 on the Data were of Beirut,

The data for the Bekaa for 1952 to 1956 were from the weather station at Rayak and for 1957 and 1958 from the American University Farm in the Bekaa. 本本

*** Inasmuch as seed maturation occured between May and September, weather data are given only for that period.

APPENDIX B

Insects found in seed lots harvested in 1958.

Bruchus pisorum in Pisum sp.

Bruchus sp. in Lens esculentum

Bruchidius sp. in Trifolium alexandrinum

Bruchus sp. in Medicago tribuloides

These insects, which hibernate during the winter as adults in seeds remaining in the field or planted in the fall, emerge in the spring, feed on young leaves and blossoms, copulate, and the females deposit their egges on the immature pod. The eggs hatch into tiny grubs, which enter the seeds, eat part of the contents, pupate inside and remain there for varying periods of time. The beetles mature more rapidly in stored seeds than in the field but in any case there is only one generation per year.

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