ABSTRACT

Lentil seeds of five cultivars were gathered from farms stores from various sites throughout Jordan in 2013 and 2014 to examine biological factors affecting the plant production. Germination, viability and purity percentages of the lentil seeds samples were evaluated by the international standards. Seed germination was ranged from 65 to 100% and seed viability was ranged from 80 to 100% depending on the lentil cultivar and the site of seed collection. The correlation between seeds germination and viability % was highly positive. The percentages of seeds purity were ranged between 80 and 99% in all lentil cultivars. Number of holes caused by the stored product insects in all seeds in the Red and Balady cultivars were significantly less than the other lentil cultivars. The most common insects species in the seeds samples of all cultivars from different parts of Jordan was Rhizophorpha dominica, followed by Bruchus rufimanus, Sitophilus granarium, Oryzaephilus surinamesis, Anthermus museorum, A. verbasci and Cryptolestes ferrugineus and from Amman governorate was B lentis, followed by B. rufimanus, Callosobruchus maculatus, B. incarnatus, B. pisorum and C. chinensis. Several agricultural practices were concluded and suggested to get high quality lentil seeds to be planted.

KEYWORDS: lentil; seeds cultivars; purity; viability; germination; legume stored insects.

INTRODUCTION

The lentil belongs to legumes. It is planted throughout the world to get seeds for human food and straw for animal feed [4, 3, 16]. The area planted with lentil is considered to be fourth of the plant areas in the Middle East. The lentil is a major food in the Middle East countries to get an important plant protein source need to human. Lentil is considered to be the second legume crop in importance planted in Jordan. The production from this plant provides only less than 3% of annually needed for Jordanian consumers [3]. Wheat from cereals and lentil from legumes are always considered to be strategic commodities worldwide including Jordan [4, 3, 16]. These crops must be stored for several months to meet the required amount for human to face major problem such as war, demand, drought and natural catastrophes [2, 15]. During storing for long time, the lentil grains might be infested by several pests particularly beetles and weevils. The infestation by the insect pest might cause severe direct and indirect damage. Stored legume grains must be protected, otherwise these grains will be changed physically in odor, appearance, lot of holes and size, and chemically lead to increase in temperature and humidity. The attacked insects might kill embryos and produce undesirable flour, and punctured and powdered
products [3,11]. A major concern with the presence of insects that the feed on seeds, insect molting skins, dead insect stages, potential to vector disease organisms, possess hair and exoskeletons [3, 11].

It is the aim of this study to find the biological parameters affecting lentil seeds germination and viability, and the various stored lentil beetles and weevils infesting several lentil cultivars collected from farms stores of different parts of Jordan to detect cultivars infestation by the weevils and beetles stored insects to help in finding effective control practices in lentil stores.

MATERIALS AND METHODS

Sampling method of lentil seeds:

From stores of farms:

Random samples of lentil seeds were gathered from various sources from stores of farmers for two years between 2013 and 2014. The sources of the gathered seeds were farms planted with lentil by the Jordanian farmers. These collected seeds were represented for all regions in Jordan. The samples were placed in paper bags, transferred to the laboratory, kept at room temperature and then taken for the required tests. Relative humidity in farm stores where they were gathered was ranged between 40 and 85%, while temperature was ranged between 5-40°C. There were five gathered lentil cultivars. The gathered samples for each of the cultivar were replicated from 4 to 7 replicates. The size for each sample was 2-3 kg taken from the amounts of seeds which were collected for different seed health tests. These seeds for each source were separately mixed and kept in well labeled bags including a code number, source of collection and then stored at room temperature until seed testing and seed insect examination. Temperature in stored room was in average of 22±5 and relative humidity was range between 40to 70%. Seeds of lentil samples were gathered from different parts of Jordan. The locations were Ajloon, Amman, Mwaqar, Madaba, Karak, Shoubak, Tafilah, Irbid, Maan, Mshaqar, Rumtha and Mafraq.

From Amman governorate:

Random lentil seeds samples were collected from Amman governorate from different stores of farmers for years extended between 2010 and 2015. The other needed important was as mentioned previously in section 2.1.a.

Seeds germination and purity tests:

Fifty grams from each lentil seed samples were taken with three replications. The gravel particles, other crops, weed seeds, and other impurities were removed and weighed adopting the international standards (the International Seed Testing [8,10,14]. The taken seeds were dipped in tap water for 24 hours to investigate the germination ability. Four hundred lentil seeds from the impregnated ones with three replicates were taken and then divided into four groups. There were 100 seeds in average per group. The seeds were transferred onto moistened filter papers in a plastic dish and then the dish cover was placed on to prevent seeds from drying. The seeds were placed under room temperature with the average of 25°C ± 2 and relative humidity of 60% ± 5. They examined after five days. The seedlings were categorized according to Klys[11] to healthy, not healthy and dead ones. The seeds were examined, counted and reevaluated after a period of two weeks to confirm the primary counting. The percentages of seeds germination were calculated according to the following formula:

\[ \% \text{ seed germination} = \frac{\text{Normal seedling}}{\text{Total number of seeds}} \times 100 \]

and then the average of for the four replication was obtained.

Seeds viability examination;

This test was conducted according to the ISTA [10]. The seeds were transferred on-to moistened filter paper for a period of two days to examine seed viability under the room temperature. The percentages of seeds germination were obtained by calculating the seeds with embryos that emerged. After a period of one week, the test was redone to obtain the normal seed germination.

Number of holes in the seeds:

Fifty grams from each replicate for each lentil cultivar were randomly and separately taken. These were transferred onto a glass Petri dish (10cm in diameter) to count the number of holes in all seeds by bare eyes and sometime under the binocular microscope when required. Total number of holes and means numbers of holes per one seed were obtained. The design was RCD and difference between means for number of holes for all examined seeds and per one seed for the five cultivars were obtained using one way analysis of variance (F-test) and LSD test.
Beetles and weevils in the cultivars of lentil seeds:

Fifty grams from each cultivar were randomly and separately were placed in the glass Petri dish from those samples collected between 2013 and 2014 from all regions of Jordan, and those collected between 2010 and 2015 from only Amman governorate. There were at least three replicates from each tested cultivar for insect examination. The occurred stored lentil insects were separated and counted for the same species. The key of Ptadt [16] was used to identify beetles and weevils that normally attacked stored cereal. Several keys were used to identify beetles and weevils that normally attacked stored legumes [3, 13, 17, 4]. The design was always the RCD. The difference between the number of each insect for the five cultivars or the Balady cultivar in Amman governorate were detected using one way analysis of variance (F-test) and the LSD test.

Results:

Seeds viability%:

Lentil seeds viability was generally ranged between 80 and 100% relying on the collection site, the store period in farm stores, and the cultivar. The southern locations of Jordan particularly from Madaba were with low viability %, but without significant differences. Lentil seeds gathered from farm stores in the northern sites such as Ajloon , Irbid and Rumtha and the middle parts of Jordan such as Amman, were with high viability %, but without significant differences (Table 1). Local cultivar was ranged between 80-100 % viability. Balady from Madaba was the lowest in viability%, followed by Turkey in Tafilah, Maan, Madaba and Irbid (Table 1). White, Red, Jordan 3 and sometimes Balady and Turkey was with 100% viability.

Table 1: Percentages of purity, germination and viability for lentil seeds of several cultivars in different parts of Jordan in 2013 and 2014.

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Sample source</th>
<th>Cultivar</th>
<th>% Purity</th>
<th>% Germination</th>
<th>% Viability</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Ajloon</td>
<td>Balady</td>
<td>99.36</td>
<td>100</td>
<td>95</td>
</tr>
<tr>
<td>3</td>
<td>Ajloon</td>
<td>Balady</td>
<td>98.80</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>4</td>
<td>Amman</td>
<td>Balady</td>
<td>96.40</td>
<td>80</td>
<td>80</td>
</tr>
<tr>
<td>18</td>
<td>Amman</td>
<td>Balady</td>
<td>98.88</td>
<td>80</td>
<td>95</td>
</tr>
<tr>
<td>19</td>
<td>Mwaqar</td>
<td>Balady</td>
<td>98.08</td>
<td>85</td>
<td>85</td>
</tr>
<tr>
<td>26</td>
<td>Madaba</td>
<td>Balady</td>
<td>98.96</td>
<td>65</td>
<td>80</td>
</tr>
<tr>
<td>39</td>
<td>Madaba</td>
<td>Balady</td>
<td>98.56</td>
<td>90</td>
<td>85</td>
</tr>
<tr>
<td>31</td>
<td>Amman</td>
<td>Balady</td>
<td>98.40</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>39</td>
<td>Karak</td>
<td>White</td>
<td>99.84</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>41</td>
<td>Karak</td>
<td>Red</td>
<td>99.76</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>45</td>
<td>Shoubak</td>
<td>Balady</td>
<td>99.52</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td>50</td>
<td>Tafilah</td>
<td>Turkey</td>
<td>99.20</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td>56</td>
<td>Shoubak</td>
<td>Balady</td>
<td>99.04</td>
<td>100</td>
<td>95</td>
</tr>
<tr>
<td>59</td>
<td>Shoubak</td>
<td>Balady</td>
<td>99.50</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>60</td>
<td>Irbid</td>
<td>Balady</td>
<td>96.70</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>64</td>
<td>Maan</td>
<td>Turkey</td>
<td>99.90</td>
<td>90</td>
<td>95</td>
</tr>
<tr>
<td>65</td>
<td>Maan</td>
<td>Balady</td>
<td>98.80</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>70</td>
<td>Madaba</td>
<td>Turkey</td>
<td>99.90</td>
<td>90</td>
<td>95</td>
</tr>
<tr>
<td>74</td>
<td>Madaba</td>
<td>Balady</td>
<td>96.40</td>
<td>100</td>
<td>85</td>
</tr>
<tr>
<td>81</td>
<td>Mshaqar</td>
<td>Jordan 3</td>
<td>99.10</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>95</td>
<td>Rumtha</td>
<td>Balady</td>
<td>98.08</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td>106</td>
<td>Mafraq</td>
<td>Turkey</td>
<td>98.56</td>
<td>90</td>
<td>100</td>
</tr>
<tr>
<td>112</td>
<td>Irbid</td>
<td>Balady</td>
<td>99.92</td>
<td>90</td>
<td>95</td>
</tr>
<tr>
<td>123</td>
<td>Irbid</td>
<td>Turkey</td>
<td>99.60</td>
<td>85</td>
<td>95</td>
</tr>
<tr>
<td>127</td>
<td>Ajloon</td>
<td>Balady</td>
<td>80.16</td>
<td>100</td>
<td>95</td>
</tr>
</tbody>
</table>

Seeds germination%:

% of lentil seed germination were ranged from 65 – 100% (Table1). The correlation between viability% and germination % was highly positive ($r^2 = 0.640$) (Table 1 and Table 2). Generally, germination % for those samples gathered from the middle and northern sites of Jordan was higher than those from the southern sites (Table 1), but without significant differences. The lowest % of germination was 65% for Balady cultivar from Madaba region (Table1).

Table 2: Values of correlation coefficient ($r^2$) between germination %, viability and purities for lentil seeds collected from different sources in Jordan during 2013, and 2014.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Correlation coefficient ($r^2$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seeds germination% vs seeds viability%</td>
<td>0.640</td>
</tr>
<tr>
<td>Seeds purity% vs seeds germination %</td>
<td>0.102</td>
</tr>
<tr>
<td>Seeds purity% vs seeds viability %</td>
<td>0.439</td>
</tr>
</tbody>
</table>

Seeds purity%:

The percentages of lentil seeds purity were ranged from 80.16 to 99.92 in all cultivars gathered from all sites in different regions in Jordan. Purity % of lentil seeds gathered from all sites of Jordan were generally high.
and did not differ significantly (Table 1). The correlation coefficient between viability % and purity % was positively medium (0.439) (Table 2). The correlation coefficient between seeds purity % and seed germination % was positively weak (0.102) (Table 2).

**Holes in seeds:**
Means numbers of holes in five lentil cultivars gathered from various sites distributed throughout Jordan in 2013 and 2014, caused by several weevils and beetles in all seeds and per seed are shown in Table (3). Number of holes in all examined seeds in the local cultivar called Jordan 3 was significantly higher than the other cultivars (Table 3). Balady cultivar contained holes in all seeds was significantly less than the other cultivars, followed by White and Turkey, but without significant differences. However, means numbers per seed in all cultivars had the same trend as commented in numbers of holes in all seeds (Table 3).

**Table 3:** Means numbers of holes caused by several stored product insects in five lentil cultivars collected from different parts of Jordan in 2013 and 2014.

<table>
<thead>
<tr>
<th>Lentil cultivar</th>
<th>No. of holes ± SE</th>
<th>All seeds</th>
<th>Per seed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Balady</td>
<td>47.22 ± 2.66c</td>
<td>0.062 ± 0.012 c</td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>54.67 ± 2.79 b</td>
<td>0.070 ± 0.014 b</td>
<td></td>
</tr>
<tr>
<td>Red</td>
<td>23.00 ± 1.91 d</td>
<td>0.029 ± 0.010 d</td>
<td></td>
</tr>
<tr>
<td>Jordan 3</td>
<td>92.00 ± 5.21 a</td>
<td>0.117 ± 0.018 a</td>
<td></td>
</tr>
<tr>
<td>Turkey</td>
<td>55.00 ± 2.81 b</td>
<td>0.071 ± 0.013 b</td>
<td></td>
</tr>
</tbody>
</table>

Means in columns sharing the same letter are not significantly different at 0.05 probability using LSD test.

**Stored product insects in seeds samples:**

**Various regions in Jordan:**
Means numbers of seven species of stored lentil insects in five lentil cultivars seeds gathered from various sites in Jordan in 2013 and 2014 are shown in Table (4). The most common species in the lentil seeds samples of the five cultivars was the lesser grain borer, *Rhyzopertha dominica*, followed by the large faba bean beetle *Bruchus rufimanus* (Table 4). The Balady lentil cultivar was significantly attacked by the lesser grain borer (8.539) with a higher mean compared with the other cultivars infestation (Table 4). Jordan 3 cultivar was significantly the least infested with the borer, followed by Turkey, White and Red. The White lentil cultivar was significantly infested with the large faba bean beetle (16.333) with a highest mean compared with the other cultivar infestation (Table 4), however, followed by Balady and Jordan 3. Turkey cultivar was significantly infested with the wheat weevil, *Sitophilus granarium*. The other insects namely *Oryzaephilus surinamensis*, *Cryptolestes ferrugineus*, *Anthrenus museorum*, *A. verbasci* had infested the different lentil cultivars in a few numbers (Table 4)

**Table 4:** Means numbers of seven stored product insects for five lentil cultivars seeds collected from different parts of Jordan in 2013 and 2014.

<table>
<thead>
<tr>
<th>Lentil cultivar</th>
<th>No. of counted insects/cultivar ± SE</th>
<th><em>Rhyzopertha dominica</em></th>
<th><em>Sitophilus granarium</em></th>
<th><em>Oryzaephilus surinamensis</em></th>
<th><em>Bruchus rufimanus</em></th>
<th><em>Cryptolestes ferrugineus</em></th>
<th><em>Anthrenus museorum</em></th>
<th><em>Anthrenus verbasci</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Balady</td>
<td>8.539 ± 0.161 a</td>
<td>0.00 ± 0.00 c</td>
<td>0.00 ± 0.00 b</td>
<td>4.428 ± 0.09</td>
<td>0.072 ± 0.01 a c</td>
<td>7.0 ± 0.02 a</td>
<td>0.00 ± 0.00 b</td>
<td>1.00 ± 0.02 a</td>
</tr>
<tr>
<td>White</td>
<td>2.00 ± 0.04 c</td>
<td>0.50 ± 0.01 b</td>
<td>1.00 ± 0.02 a b</td>
<td>16.333 ± 0.291a</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>1.00 ± 0.02 a</td>
<td>0.00 ± 0.00 b</td>
</tr>
<tr>
<td>Red</td>
<td>4.00 ± 0.08 b</td>
<td>0.00 ± 0.00 c</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00c</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
</tr>
<tr>
<td>Jordan 3</td>
<td>0.00 ± 0.00 e</td>
<td>0.00 ± 0.00 c</td>
<td>0.00 ± 0.00 b</td>
<td>5.00 ± 0.05</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
</tr>
<tr>
<td>Turkey</td>
<td>1.00 ± 0.02 d</td>
<td>5.00 ± 0.06 a</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 c</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
<td>0.00 ± 0.00 b</td>
</tr>
</tbody>
</table>

Means in columns with the same letters are not significantly different at 0.05 probability using LSD test.

**Amman governorate:**
Table 5 shows means numbers of six stored product insects separated from Balady cultivar from Amman governorate between 2010 and 2015. The most common species in the lentil seeds samples of the cultivar was significantly *Bruchus lentis* (ervi), followed by *Bruchus rufimanus*, *Callosobruchus maculatus* and *B. incanatus* with significant differences, *B. pisorum* and *C. chinensis* which was significantly the lowest (Table 5).
Table 5: Means numbers of six stored product insects for Balady cultivar from Amman governorate in Jordan between 2010 and 2015.

<table>
<thead>
<tr>
<th>Insect species</th>
<th>No. of counted insects ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Callosobruchus maculates</td>
<td>10.10 ± 0.362 c</td>
</tr>
<tr>
<td>Bruchus rufimanus</td>
<td>15.13 ± 0.381 b</td>
</tr>
<tr>
<td>Bruchus incarnatus</td>
<td>9.12 ± 0.301 c</td>
</tr>
<tr>
<td>Bruchus pisorum</td>
<td>5.13 ± 0.281 d</td>
</tr>
<tr>
<td>Bruchus lentis (ervi)</td>
<td>25.11 ± 0.521 a</td>
</tr>
<tr>
<td>Callosobrachus chinensis</td>
<td>3.03 ± 0.191 e</td>
</tr>
</tbody>
</table>

Mean in columns sharing the same letter are not significantly different at 0.05 probability using LSD test.

Discussion:

Lentil is planted throughout the world to get seeds for human food and straw for animal feed. Wheat from cereals and lentil from legumes are always considered to be strategic commodities worldwide [4, 3, 16] including Jordan [16]. Stored grains and legumes are subject to insect infestation and deterioration from fungi and bacteria [2,13]. Harein and Meronuch, [7] estimated the postharvest losses in the United states particularly from insects were to be $500 million per year. Many stored insects possess hairs, moulting skins and dead insects which are available to contaminate the stored commodity. However, lentil seed should be carefully protected from the direct insects to minimize the damage and the loss during storing. In addition, some stored product insects might move or transfer to the stored seed through the field infestation [3]. Insect populations can increase to outbreaks when left unchecked in bins and store houses, causing high environmental temperatures and moisture that accelerate fungi, insect development within the stored legumes and grains, increasing losses and bad quality of the stored commodity [5, 3, 13]. Seeds after harvesting should store in the proper way to have healthy ones in the clean, well prepared stores, free of pests, with less than 12% water content, free of impurities, weed seeds, well ventilated, proper temperature and relative humidity and well equipped for insect fumigation when required. Small, broken and shrunken seeds should be excluded because their germination and viability % might be low [18, 1]. However, lentil seeds attacked by the stored product insect pests in this study caused sometimes low in seeds vigour. These results agreed with those found to similar work in Jordan, but on wheat [18] and barley [1]. Generally, in case of lentil seeds, beetles and weevils and their stages might be transferred by them from the field before and after harvesting to the bins and store houses [18]. Selection of lentil seeds is an important factor in the agricultural practices. Seed technology is usual taken in consideration seed germination, viability, purity and pests transferred by the seeds through production, packing, sold and cultivated in many countries. The previous practices will give chance for the beetles and weevils to spread all over the world, causing high loss in lentil and cereal yields [12,18].

In this investigation, stored product insects found in the seeds of the five cultivars are divided to two groups. The first is those normally attacked cereal seeds such as R. dominica, S. granarium and O. surinamensis [1,9]. The first two pests are considered to be primary insects which are to attack healthy, non healthy, broken and crushed seeds [6,18]. The second group is those normally attacked legumes including stored lentil seeds such as B. rufimanus, C. ferrugineus, A. museorum, A. verbasci, in seeds collected from the five cultivars from different parts of Jordan, and B. lentis, B. rufimanus, C. maculates, B. incarnatus, B. pisorum and C. chinensis in Balady seed collected from Amman governorate. The seed beetles attacking legumes are called bruchids. These bruchids develop in legume seeds such as lentil, peas and beans causing holes and crushed and powdered products [9,13,4]. Seeds of the local lentil cultivar namely Balady and the imported Red cultivar from the present investigation have shown less in number of holes in all seed and per seed the in the experimental seeds. The results might encourage Jordanian farmers to plant and store them. Plant breeders with entomologists could cooperate to find new cultivars with high yield and less infestation with insect pests.

The primary mentioned stored product insects and the bruchids can dig holes in the healthy stored lentil seeds encouraging other pests to attack [2, 18]. Larvae of these pests might develop and live within stored seeds causing high mortality to embryos and declining viability and seeds germination [15, 18].

Conclusions:

Lentil seeds with high quality are required to have high germination, viability, and high production. The seeds should be harvested as soon as possible after ripening to avoid insect infestation and then screened before storing to get rid of impurities, weed seeds, small, broken, crushed and shrunken seeds. Lentil stored seeds should be free of insect and disease infection, and dry enough. Before storing, stores should be clean, free from insects and well equipped and established to make the fumigation easy to carry out. The use of tolerant or resistant cultivars are needed. Storage of lentil seeds in air-tight stores and containers are useful, with regular monitoring for temperature and relative humidity.
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Declaration Of Interest:
The authors declare that there are no conflict of interest.

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