KSÜ Tarım ve Doğa Derg 25 (4): 511-520, 2022
KSU J. Agric Nat 25 (4): 511-520, 2022
DOI:10.18016/ksutarimdoga.vi.870090

Use of Extreme Low Temperatures Against *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae) in Storage Management

Onur AKER[1], Celal TUNÇER[2]
[1]Amasya University, Suluova Vocational School, Department of Plant Protection, Amasya, Türkiye, [2]Ondokuz Mayıs University, Faculty of Agriculture, Department of Plant Protection, Samsun, Türkiye
[1]https://orcid.org/0000-0002-9581-9697, [2]https://orcid.org/0000-0002-9014-8003
✉: onur.aker@amasya.edu.tr

ABSTRACT
This study was carried out to determine the effect of extreme low temperatures on different biological stages of *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae). Different extreme low temperatures were tested for egg, larva, pupa and adult stages in all experiments with different exposure times. For larval, pupal and adult stages, 100% mortality was obtained after 45 minutes at -16 °C, 30 minutes at -20 °C and 20 minutes at -26 °C. LT95 value was determined as 38.3, 39.0 and 36.8 minutes at -16 °C for larva, pupa and adult, respectively. LT95 value was determined as 27.4, 27.5 and 26.0 minutes at -20 °C for larva, pupa and adult, respectively. At the end of 120 minutes at -20 °C, 100% mortality was obtained in the egg stage and it was determined that the adults did not hatch from the eggs kept at -20 °C for 60 minutes. From these results, it is understood that extreme low temperatures are effective in all biological stages of *C. chinensis* that cause damage in storage.

Plant Protection

Research Article

Article History
Received : 01.02.2021
Accepted : 21.08.2021

Keywords
Callosobruchus chinensis
Low temperature
Exposure time
Storage
Chickpea

Depolama Yönetiminde *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae)'e Karşı Aşırı Düşük Sıcaklıkların Kullanılması

ÖZET
Bu çalışma, aşırı düşük sıcaklıkların *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae)'nin farklı biyolojik dönemleri üzerindeki etkisini belirlemek için yapılmıştır. Farklı maruz kalma sürelerine sahip tüm deneylerde yumurta, larva, pupa ve ergin dönemleri için farklı aşırı düşük sıcaklıklar test edilmiştir. Larva, pupa ve ergin dönemleri için, -16 °C'de 45 dakika, -20 °C'de 30 dakika ve -26 °C'de 20 dakika sonunda %100 ölüm oranı elde edilmiştir. LT95 değeri larva, pupa ve ergin için -16 °C'de sırasıyla 38.3, 39.0 ve 36.8 dakika olarak belirlenmiştir. LT95 değeri larva, pupa ve ergin için -20 °C'de sırasıyla 27.4, 27.5 ve 26.0 dakika olarak belirlenmiştir. -20 °C'de 120 dakika sonunda yumurta döneminde %100 ölüm sağlanmış ve -20 °C'de 60 dakika bekletilen yumurtalardan erginlerin çıkmadığı tespit edilmiştir. Bu sonuçlardan, aşırı düşük sıcaklıkların depolamada zarara neden olan *C. chinensis*'in tüm biyolojik dönemleri üzerinde etkili olduğu anlaşılmaktadır.

Bitki Koruma

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi : 01.02.2021
Kabul Tarihi : 21.08.2021

Anahtar Kelimeler
Callosobruchus chinensis
Düşük sıcaklık
Maruz Kalma süresi
Depolama
Nohut

To Cite: Aker O, Tuncer 2022. Use of Extreme Low Temperatures Against *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae) in Storage Management. KSU J. Agric Nat 25 (4): 511-520. DOI:10.18016/ksutarimdoga.vi.870090.

Atıf İçin: Aker O, Tuncer 2022. Depolama Yönetiminde *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae)'e Karşı Aşırı Düşük Sıcaklıkların Kullanılması. KSÜ Tarım ve Doğa Derg 25 (4): 511-520. DOI:10.18016/ ksutarimdoga.vi.870090.

INTRODUCTION

Legumes are an essential part of the human diet, as they are very rich in protein, carbohydrates and dietary fiber (Chibbar, 2009). Chickpea (*Cicer arietinum* L., Fabales: Fabaceae) is one of the most important legumes in many parts of the world and it originated in Turkey's southeast (Ali et al., 2009; Baloch and Zubair, 2010). According to FAO-2019 data, chickpeas are grown on an area of approximately 13.7 million hectares with 14.2 million tons of production in the world. India, Turkey, Russia, Myanmar and Pakistan are the biggest chickpea

producing countries in the world (Faostat, 2020). Chickpeas are highly nutritious and rich in energy in terms of high protein content and balanced fat, carbohydrate and fiber values (Ravi and Harte, 2009).

Chickpeas are exposed to the damage of many pest species both in the field and in storage conditions (Clement et al., 2000; Hill, 2003). Stored product pests are very dangerous because of the extent and prevalence of the irreversible losses they cause in legumes (Kumar et al., 2009; Jat et al., 2013). *Callosobruchus chinensis* L. is one of the most destructive pests of stored legumes, including chickpeas (Kutbay et al., 2011; Tessema et al., 2015). This pest not only causes loss of quantity and quality of chickpeas, but also reduces the economic value and nutritional content of the product (Righi-Assia et al., 2010; Homan and Yubak Dhoj, 2011).

Chemical management methods using fumigation with methyl bromide, phosphine or dusting with pirimiphos methyl are very successful against this pest in stored chickpeas (Athanassiou et al., 2004; Shaheen and Khaliq, 2005), however there are some disadvantages such as shipping hazards, pesticide residues, the possibility of developing resistance, health risks and environmental pollution, so there are a few limitations on chemicals (Boateng and Kusi, 2008; Boyer et al., 2012).

In recent years, due to the negative effects caused by chemical control methods, it has focused on alternative reliable control methods against stored product pests. One of these methods is to prevent the development of stored product pests by keeping the product at low temperature for a certain period of time (Fields and White, 2002; Zhong et al., 2013). Studies have shown that low temperatures can have negative effects on the growth of many stored product pests (Fields and Muir, 1996; Loganathan et al., 2011). During storage, pests are highly susceptible to dropping the storage temperature below 10 °C, and this sensitivity level can vary with biological stages (Zakladnoi and Ratanova, 1987; Maharjan et al., 2017). Also, susceptibility to deadly low temperatures; depends on pest species, exposure temperature, exposure time, sex of insects and ambient humidity (Mason and Strait, 1998; Zhong et al., 2013).

The aim of this research was to determine the adverse effects of extreme low temperatures on the different biological stages of *C. chinensis*, thereby minimizing the damage this pest can cause to stored products. In this way, an alternative method will be developed for stored product pests.

MATERIAL and METHODS

Insect Rearing

C. chinensis adults were obtained from stock cultures grown on chickpea seeds at the Ondokuz Mayıs University, Faculty of Agriculture, Entomology Laboratory. *C. chinensis* were reared in glass jars (1 L) covered with muslin cloth in climate cabins at 28±1 °C, 70±5% RH and a 16:8 h (L:D) photoperiod.

The Extreme Low Temperature Experiments

The healthy chickpea seeds to be used in the experiments were sterilized in the oven at 50 °C for 24 hours, then the chickpea seeds were cooled. The moisture content of the sterile seeds was adjusted to 14% moisture and the seeds were made ready for experiments (Wright et al., 1987). The moisture content of chickpea seeds was monitored regularly throughout the experiments with a digital moisture meter (WILE Grain Moisture Meter). Low temperature experiments were performed in a temperature-controlled refrigerator (SIEMENS KG57NP01NE) and temperatures were monitored hourly using a digital data logger.

Five male-5 female *C. chinensis* adults were placed in sterile glass jars each containing 40 sterile chickpea seeds. *C. chinensis* adults were allowed to lay eggs on sterile seeds, and these egged seeds were transferred to sterile petri dishes. There were 20 chickpea seeds in each petri dish. In this way, more than 100 sterile petri dishes were prepared. The adults emerged from these eggs were collected daily. Some of the adults emerged from these eggs were used to determine the effect of low temperatures on adults. In addition, 2 male-2 female pairs from newly emerged adults were transferred to new sterile petri dishes each containing 10 sterile chickpea seeds. In this way, more than 300 new sterile petri dishes were prepared. Eggs laid on chickpea seeds in each petri dish by the pest pairs within 24 hours were used in all experiments.

Low-temperature experiments were performed on eggs, larvae, pupae and adults of 24-48 hours ages. Since larval and pupal stages occur in chickpea seeds, eggs between 0-24 hours at 28±1 °C - 70±5% RH were used to determine the biology of the *C. chinensis*. Accordingly, the larval stages started to form on the 6th day on average, and the pupal stages on the 22nd day on average. For 24-48 hour larval and pupal studies, 6-day-old chickpea seeds were used in the larval trials and 22-day-old chickpea seeds were used in the pupal trials. Different temperatures and different exposure times were used to determine the effects of extreme low temperatures on different biological stages of *C. chinensis*. The effects of low temperature on *C. chinensis'* eggs were studied at two constant temperatures (4 and -20 °C) and eight exposure times (5, 10, 15, 30, 45, 60, 120 and 180 minutes). The effects of low temperature on *C. chinensis'* larvae and pupae were studied at three constant temperatures (-16, -20 and -26 °C) and seven exposure times (10, 20, 30, 45, 60, 120 and 180 minutes). The effects of low temperature on *C.*

chinensis' adults were studied at six constant temperatures (8, 4, 2, -16, -20 and -26 ° C) and seven exposure times (10, 20, 30, 45, 60, 120 and 180 minutes). 480 eggs, 120 larvae, 120 pupae, and 60 adult pests were used in 4 replicates for each exposure time of each temperature. In addition, control groups with 4 replicates were formed under normal growth conditions for each biological stage in the experiments (28±1 °C, 70±5% RH, 16:8 h (L:D) photoperiod).

It was observed that adults started to emerge from chickpeas 28-36 days after the first egg was laid under normal growth conditions. It has been determined that the lifespan of *C. chinensis* is about 36-42 days. Egg, larva, pupa and adult stages exposed to different low temperatures in the experiments were taken to normal growth conditions after different exposure times. Eggs that have not completed their development or have not reached the larval stage are considered dead as a result of the controls made for 30 days after being placed in normal growing conditions. Larvae that did not complete their development or did not reach the pupal stage as a result of the controls made for 60 days after being placed in normal growth conditions were considered dead. Pupae that did not complete their development or did not mature as a result of the controls made for 60 days after being placed under normal growing conditions were considered dead. In order to determine whether the adults died as a result of the experiment, adults were touched with a fine-tipped brush and adults who did not respond were considered dead. Adults were checked for 30 days after being placed under normal growing conditions, and dead adults were counted again for control after 30 days.

Statistical Analysis

The data obtained from low temperature applications and exposure times were tried to be analyzed using a One-Way Anova program (SPSS 21). Mortality rates was considered significantly different at $P<0.05$. Statistical means are separated by Duncan's mean separation test. Statistical significance was established by comparing the p value in the "t" test table. The data obtained were corrected using Abbott's control formula (Abbott, 1925) and subjected to probit analysis (Finney, 1971) to estimate the LT50 and LT95 values (Polo Plus, LeOra Software, Robertson et al., 2003).

RESULTS and DISCUSSION

Effect of Low Temperatures on Eggs

Mortality and adult emergence rates of *C. chinensis'* eggs were studied at different temperatures and different exposure times (Fig. 1), and significant differences were found between exposure times at 4 °C and -20 °C ($P<0.001$). After the eggs were kept at 4 °C for 180 minutes, it was determined that 44% of the eggs died and 78% of the remaining eggs were emerged as adults. After the eggs were kept at -20 °C for 120

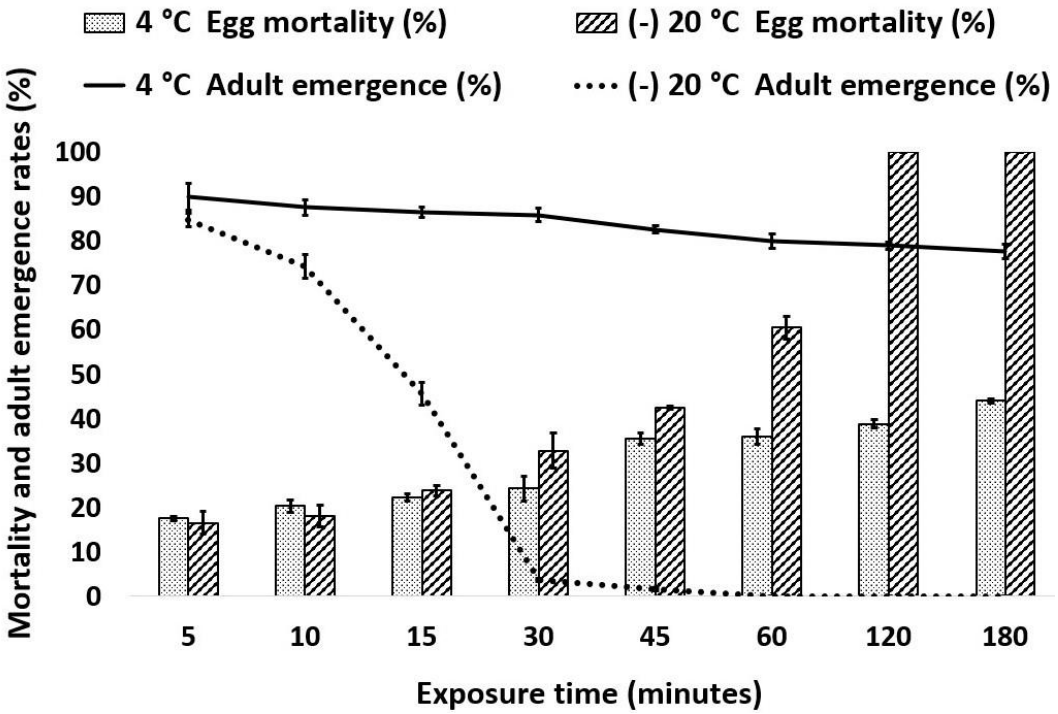

Figure 1. Mortality and adult emergence rates of *Callosobruchus chinensis'* eggs at different exposure times at 4 °C and -20 °C (P<0.001).

Şekil 1. 4 °C ve -20 °C'de farklı maruz kalma sürelerinde Callosobruchus chinensis yumurtalarının ölüm oranı ve ergin ortaya çıkma oranları (P<0.001).

minutes, it was determined that all the eggs died, and the eggs were kept at -20 °C for 60 minutes did not reach adulthood (Fig. 1). It was observed that egg mortality increased due to the increase in exposure time to -20 °C and at the same time the probability of reaching adulthood from egg was significantly reduced.

Effect of Low Temperatures on Larvae

Mortality rates of *C. chinensis'* larvae were studied, and significant differences were found between temperatures and exposure times (*P<0.001*). When the exposure time of the larvae to low temperatures was examined, it was observed that deaths occurred within the first 10 minutes. After the larvae were exposed to extreme low temperatures at -16 °C, -20 °C and -26 °C at the end of the first 10 minutes, mortality rates were determined as 7.36 %, 16.65 %, 54.24 %, respectively (*P<0.001*). As a result of the experiments, 100% larval mortality was achieved after 20 minutes, 30 minutes and 45 minutes at temperatures of -26 °C, -20 °C, -16 °C, respectively (Fig. 2).

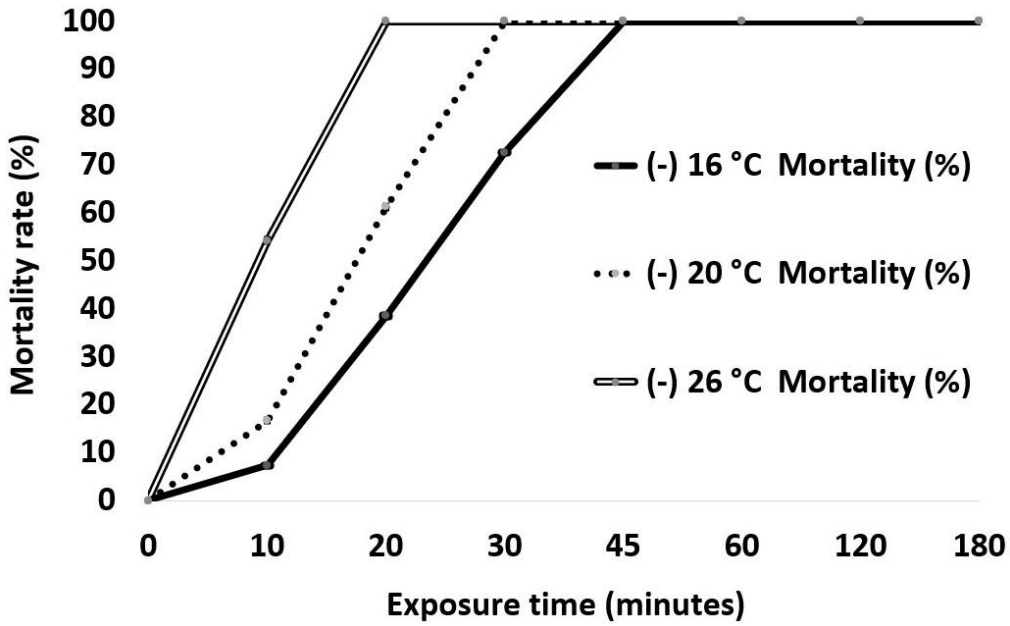

Figure 2. Mortality rates of *Callosobruchus chinensis'* larvae at different exposure times at -16 °C, -20 °C and -26 °C (P<0.001).

Şekil 2. Callosobruchus chinensis larvalarının -16 °C, -20 °C ve -26 °C'de farklı maruz kalma sürelerinde ölüm oranları (P<0.001).

Effect of Low Temperatures on Pupae

The differences between pupae death rates according to exposure times for each low temperature were found to be statistically significant (*P<0.001*). Mortality rates have been observed to increase as the exposure time to low temperatures increases. Deaths started in the first 10 minutes of the experiment, and this mortality rate progressed very rapidly. After the pupae were exposed to extreme low temperatures at -16 °C, -20 °C and -26 °C at the end of the first 10 minutes, mortality rates were determined as 8.42 %, 17.12 %, 57.64 %, respectively (*P<0.001*). As a result of the experiments, 100% pupal death was achieved at temperatures of -16 °C, -20 °C, -26 °C after 45 minutes, 30 minutes and 20 minutes, respectively (Fig. 3).

Effect of Low Temperatures on Adults

Comparing the 3 low temperatures applied to adults in 7 different exposure times, no deaths occurred in adults exposed to 8 °C after 180 minutes. 8.08 % and 3.31 % deaths were observed after 180 minutes at 2 °C and 4 °C, respectively (*P>0.05*) (Fig. 4). Comparing the 3 extreme low temperatures applied to adults in 7 different exposure times, the adults died after 10 minutes. 9.24 %, 18.64 % and 60.04 % deaths were observed after 10 minutes at -16 °C, -20 °C and -26 °C, respectively (*P<0.001*). Mortality increased with increases exposure to extreme low temperatures and 100% adult deaths were reached at -16 °C, -20 °C, -26 °C after 45 minutes, 30 minutes and 20 minutes, respectively (Fig. 4). It was determined that the control groups at each biological stage were between 95-100% successful.

Pests cause large amounts of damage in stored products, which can reach 5-10% in temperate climatic regions and 20-30% in tropical regions (Caswell, 1981; Nakakita, 1998). Temperature application is one of the most promising methods to control stored product pests (Fields, 1992). The lethal low temperature is highly effective in controlling the pests of stored seeds

(Loganathan et al., 2011). At low temperatures, the progeny of stored product pests decreases, and their development slows down to a certain point (Flinn and Hagstrum, 1990; Abdelghany et al., 2015). Stored product pests usually stop growing at temperatures below 20 °C, and stored product pests usually begin to die at temperatures below 5 °C (Fields and Muir, 1996). The results of this study have shown that extreme low temperatures have a lethal effect on all developmental stages of *C. chinensis*, and the egg stage is relatively more tolerant of cold than the other three stages (Fig. 5, 6, 7). Many extreme low temperature applications on *Callosobruchus* species also support these results (Mullen and Arbogast, 1979; Dohino et al., 1999; Johnson and Valero, 2000; Johnson and Valero, 2003).

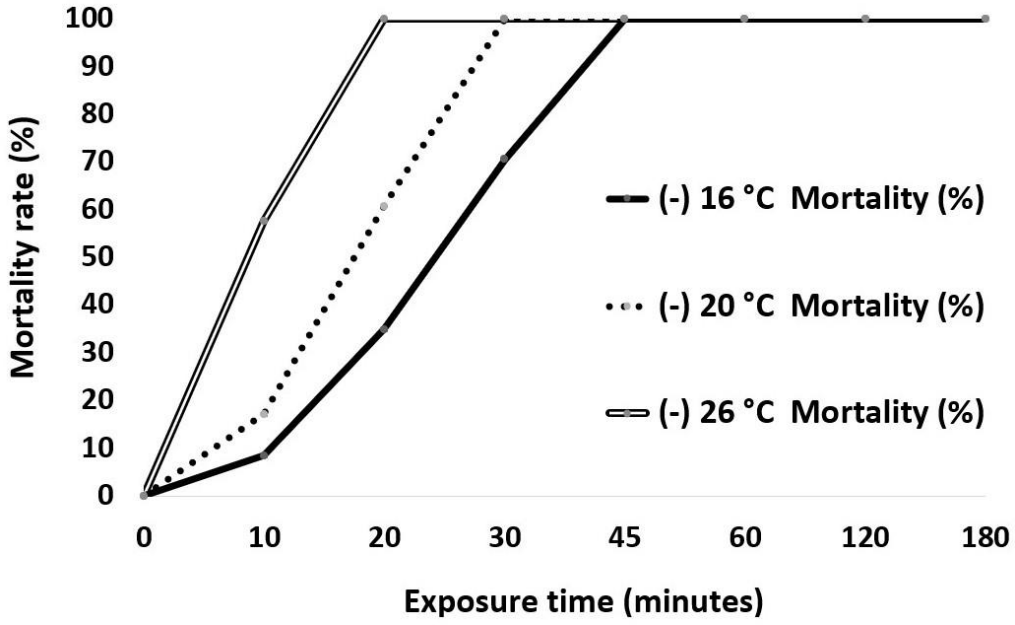

Figure 3. Mortality rates of *Callosobruchus chinensis'* pupae at different exposure times at -16 °C, -20 °C and -26 °C (P<0.001).

Şekil 3. Callosobruchus chinensis pupalarının -16 °C, -20 °C ve -26 °C'de farklı maruz kalma sürelerinde ölüm oranları (P<0.001).

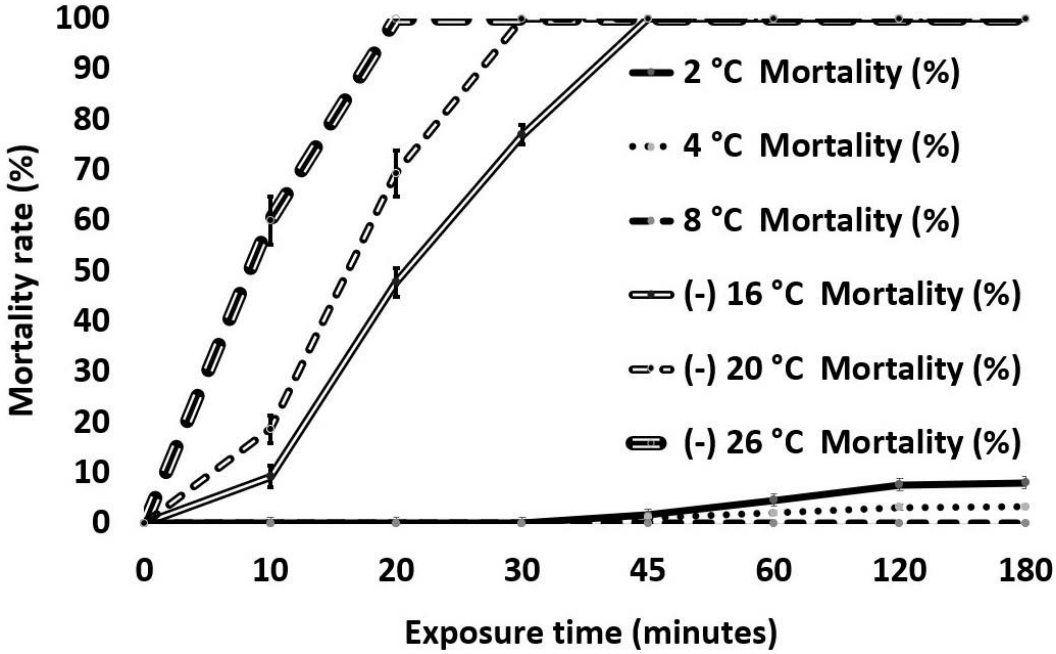

Figure 4. Mortality rates of *Callosobruchus chinensis'* adults at different exposure times at 2 °C, 4 °C, 8 °C and at -16 °C, -20 °C, -26 °C (P<0.001).

Şekil 4. Callosobruchus chinensis erginlerinin 2 °C, 4 °C, 8 °C ve -16 °C, -20 °C, -26 °C'de farklı maruz kalma sürelerinde ölüm oranları (P<0.001).

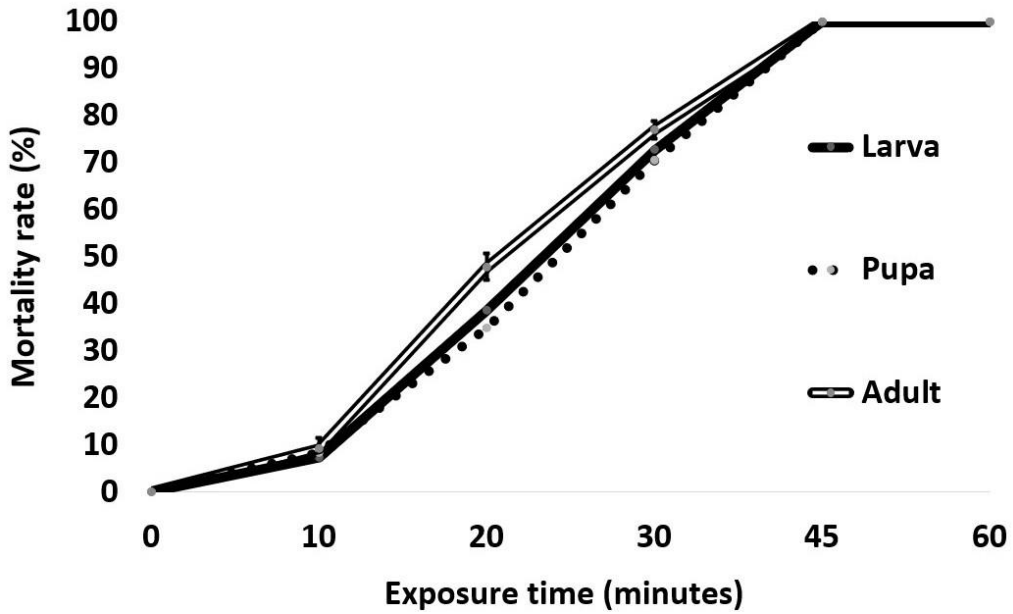

Figure 5. Mortality rates of *Callosobruchus chinensis'* larvae, pupae and adults exposed to -16 °C (P<0.001).
Şekil 5. -16 °C'ye maruz kalan Callosobruchus chinensis larva, pupa ve erginlerinin ölüm oranları (P<0.001).

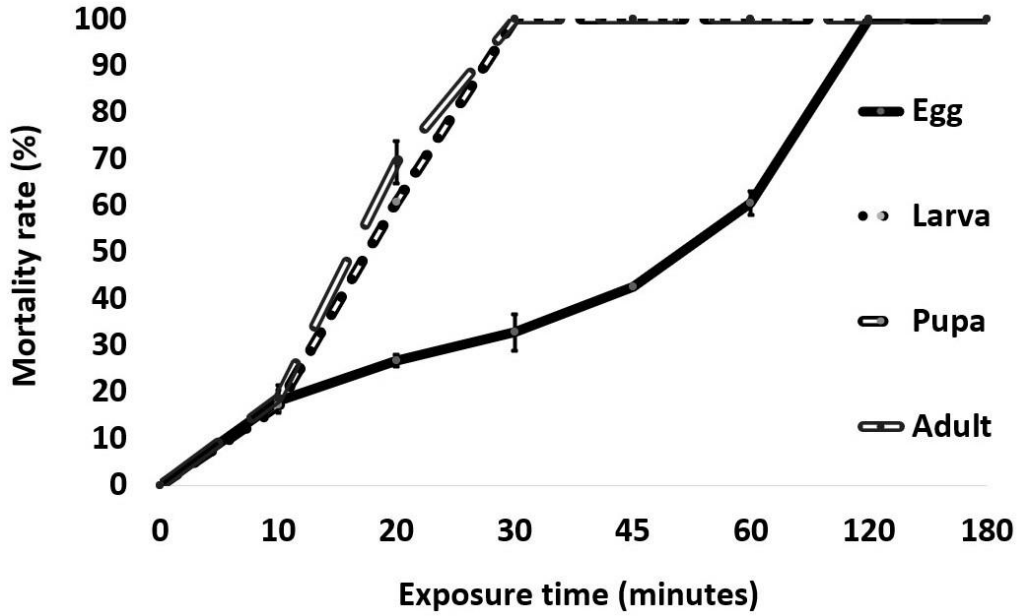

Figure 6. Mortality rates of *Callosobruchus chinensis'* eggs, larvae, pupae and adults exposed to -20 °C (P<0.001).
Şekil 6. -20 °C'ye maruz kalan Callosobruchus chinensis yumurta, larva, pupa ve erginlerinin ölüm oranları (P<0.001).

In this study, 100% death was achieved in the eggs after 120 minutes at -20 °C and it was observed that these eggs did not reach maturity after the eggs were kept at -20 °C for 60 minutes (Figure 1, 6). In other low temperature studies performed on *C. chinensis* eggs, LT50 and LT99 values at -5 °C were found to be 12.57 and 77.03 hours, respectively (Zhong et al., 2013). In another study, it was said that the time required for *C. chinensis* eggs to die completely at -4 °C should be about 20 days (Maharjan et al., 2017). In the study of Dohino et al (1999) on *Callosobruchus rhodesians* (Col: Chrysomelidae), they found a value very close to the results we found and recorded the LT99 value of the eggs as approximately 124 minutes at -18 °C. Johnson and Valero (2000) stated that after exposure of *Callosobruchus maculatus* (F) (Col: Chrysomelidae) eggs to an extreme low temperature such as -18 °C for 180 minutes, no egg reached maturity. As noted in another study by these authors, the complete mortality rate of *C. maculatus* eggs on *Vigna unguiculata* (L.) occurred after 14 days of storage in a commercial cold storage at -18 °C, and no adults emerged from these eggs kept at -18 °C for 24 hours (Johnson and Valero, 2003). Similarly, Loganathan et al. (2011) found that

the LT50 and LT95 values of *C. maculatus* eggs were 1.0 hour and 1.3 hours at -15 °C, respectively. A study on *Tribolium castaneum* (Col: Tenebrionidae), an important stored product pest, reported that more than 8 hours of exposure at -18 °C was required to kill all eggs (Arthur et al., 2015).

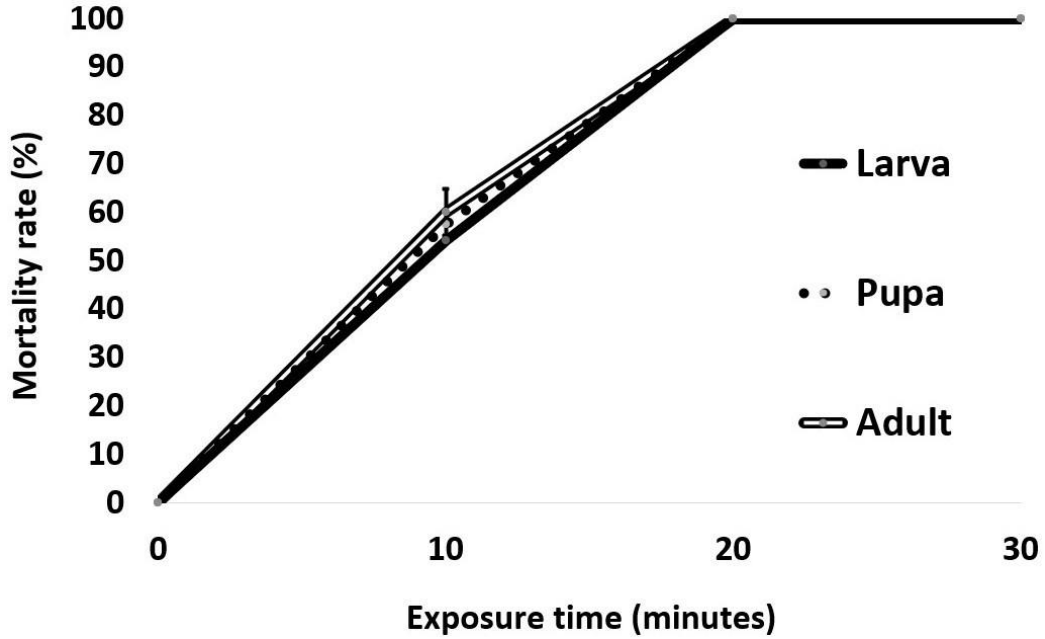

Figure 7. Mortality rates of *Callosobruchus chinensis'* larvae, pupae and adults exposed to -26 °C (P<0.001).
Şekil 7. -26 °C'ye maruz kalan Callosobruchus chinensis larva, pupa ve erginlerinin ölüm oranları (P<0.001).

This study showed that 100% mortality was obtained in larvae, pupae and adults after 20 minutes, 30 minutes and 45 minutes at -26 °C, -20 °C, -16 °C, respectively (Fig. 5, 6, 7). As a result of the findings, lethal time of the various life stages of *C. chinensis* exposed to -16 °C and -20 °C, it is understood that the lethal times of larvae and pupae are very similar. In addition, the adult mortality rate is very close to the lethal time of larvae and pupae. In this study, that LT95 value at -16 °C was 38.3, 39.0 and 36.8 minutes for larvae, pupae and adults, respectively. LT95 value at -20 °C for larvae, pupae and adults were 27.4, 27.5 and 26.0 minutes, respectively (Table 1, 2).

Table 1. LT50 and LT95 values (minutes) of *Callosobruchus chinensis'* larvae, pupae and adults exposed to -16 °C.
Çizelge 1. -16 °C'ye maruz kalan Callosobruchus chinensis larva, pupa ve erginlerinin LT50 ve LT95 değerleri (dakika)

Stage	LT50 (95% CL)	LT95 (95% CL)	Slope ± SE	Intercept ± SE	X^2 (df)
Larva	23.4 (22.1 - 24.8)	38.3 (35.9 - 41.3)	0.111 ± 0.008	-2.594 ± 0.196	3.36 (4)
Pupa	23.8 (22.5 - 25.2)	39.0 (36.6 - 42.1)	0.108 ± 0.008	-2.584 ± 0.193	3.51 (4)
Adult	21.8 (19.9 - 23.7)	36.8 (33.6 - 41.4)	0.109 ± 0.011	-2.375 ± 0.257	2.71 (4)

Table 2. LT50 and LT95 values (minutes) of *Callosobruchus chinensis'* larvae, pupae and adults exposed to -20 °C.
Çizelge 2. -20 °C'ye maruz kalan Callosobruchus chinensis larva, pupa ve erginlerinin LT50 ve LT95 değerleri (dakika)

Stage	LT50 (95% CL)	LT95 (95% CL)	Slope ± SE	Intercept ± SE	X^2 (df)
Larva	17.1 (15.9 - 18.1)	27.4 (25.6 - 29.6)	0.160 ± 0.013	-2.720 ± 0.232	6.461 (4)
Pupa	17.0 (14.9 - 19.2)	27.5 (24.5 - 32.8)	0.157 ± 0.013	-2.671 ± 0.226	7.411 (4)
Adult	16.1 (14.6 - 17.6)	26.0 (23.8 - 29.4)	0.166 ± 0.019	-2.672 ± 0.329	1.741 (4)

In another extreme low temperature study on *C. chinensis*, LT50 values at -5 °C were 24.93, 30.54 and 15.76 hours for larvae, pupae and adults, respectively. In the same study, LT99 values for *C. chinensis* at -5 °C were 171.36, 189.70 and 126.11 hours for larvae, pupae and adults, respectively. In addition, *C. chinensis* pupal LT99 values were found as 189.70, 33.81 and 2.90 hours at -5, -10 and -20 °C, respectively (Zhong et al., 2013). When two studies were compared in terms of extreme low temperature application applied to the pupal stage at -20 °C, the LT95 value in this study was found to be 27.5 minutes, and the LT99 value was found to be 175 minutes in the study conducted in 2013. In another similar study, all the

larvae and pupae of *C. chinensis* used in the studies died at -4 °C after 25 and 10 days, respectively (Maharjan et al., 2017).

Looking at similar studies on *C. maculatus*, Johnson and Valero (2000) noted that the newly emerged adults died completely after 50 minutes of exposure to -18 °C. In another study by the same authors, it was stated that adults living on seeds died completely within 40 minutes at -18 °C (Johnson and Valero, 2003). In a study on mites, all nymphs and adults of *Tyrophagus putrescentiae* (Acaridae) died within 30 minutes after cooling to -18 °C (Eaton and Kells, 2011). It took about 30-60 minutes for all life forms of *Plodia interpunctella* (Hübner) (Lep: Pyralidae) to die at -18 °C (Gvozdenac et al., 2019). The results of these four studies are very similar to the values found at -16 °C for this study. However, Loganathan et al. (2011) showed that the LT50 and LT95 values of *C. maculatus* pupae were 1.8 hours and 2.5 hours at -15 °C, respectively.

Generally, -18 °C was used against stored product pests and generally the same results were obtained. Donahaye et al. (1995) determined that approximately 180 minutes of exposure at -18 °C was required to kill all life stages of *T. castaneum, Oryzaephilus surinamensis* (Col: Silvanidae) and *Ephestia cautella* (Lep: Pyralidae). In a similar study, Ferizli et al. (2004) found that eggs, larvae, pupae, and adults of *C. maculatus* can be fully controlled by keeping them at -18 °C for 180 minutes. However, Gvozdenac et al. (2019) stated differently that the LT50 and LT99 values of *P. interpunctella* larvae died at -18 °C in a very short time such as 1.9 minutes and 12.8 minutes, respectively.

When extreme low temperature studies were examined, it was seen that there were differences in the same extreme low temperature results applied to the same stored product pest. Eliopoulos et al. (2011) found that the LT50 value of larvae of *T. granarium* at -16 °C was less than four hours. However, Abdelghany et al. (2015) found that the LT50 value of larvae of *T. granarium* at -16 °C was more than one day. In another extreme cold application study on *P.interpunctella, P.interpunctella* adults died after 120 minutes at -15 °C (Athanassiou et al., 2018), and *P.interpunctella* larvae died after 180 minutes at -15 °C. (Gvozdenac et al., 2019).

Low temperature studies have been conducted on *C. chinensis*, but not extreme low temperature studies as in this study. However, very low temperature studies have been done on similar stored product pests. While some researchers came to different conclusions that eggs are the most sensitive stage in low temperature applications (Maharjan et al., 2017), some researchers have reached different conclusions that eggs are the most resistant stage in low temperature applications (Johnson and Valero, 2003). In this low temperature study, the egg stage was found to be the most durable stage and the adult stage to be the most susceptible stage. The larval and pupal stages in this study have almost similar results. Adult stage results are close to larval and pupal stage results. There may be many reasons why different biological stages of *C. chinensis* and other stored product pests show different sensitivity and different responses to extreme low temperatures. These; There may be different factors such as the species of pests, the geographies where the pests are located, climates and temperature requirements, the species of stored product that the pests live on, the storage conditions of the product where the pests live. All these different factors can cause similar or different results in low temperature studies on stored product pests. Therefore, it is important to carry out low temperature studies carefully for each region. It would be in the interest of valuable scientists working in this field to come together in comprehensive regional and global studies on stored product pests.

CONCLUSION

Today, it is known that healthy protection of agricultural products is as important as the safe cultivation of agricultural products. It is known that the use of chemical pesticides in the storage of agricultural products has some side effects. As can be understood from previous studies, low temperature applications as a reliable way to control of stored product pests have gained importance in recent years and have been defined as an alternative management strategy against pests. As a result of this study, it was found that *C. chinensis* was susceptible to extreme low temperatures and deaths increased rapidly due to the increase in exposure time. This study will set an example for future low temperature studies, thus shedding light on the development of new strategies for a healthier and more reliable storage of stored agricultural products for a long time.

ACKNOWLEDGEMENT

This study was supported by the funding of the Scientific Research Projects Department of Ondokuz Mayıs University (Project Number: PYO. ZRT. 1904. 10.027).

Author's Contributions

The contribution of the authors is equal.

Conflicts of interest

The authors declare no conflict of interest.

REFERENCES

Abbott WS 1925. A Method of Computing the Effectiveness of an Insecticide. Journal of Economic Entomology, 18(2): 265-267.

Abdelghany AY, Suthisut D, Fields PG 2015. The effect of diapause and cold acclimation on the cold-hardiness of the warehouse beetle, *Trogoderma variabile* (Coleoptera: Dermestidae). The Canadian Entomologist, 147(2): 158-168.

Ali MA, Nawab NB, Abbas A, Zulkiffal M, Sajjad M 2009. Evaluation of selection criteria in *Cicer arietinum* L. using correlation coefficients and path analysis. Australian Journal of Crop Science, 3(2): 65-70.

Arthur FH, Hartzer KL, Throne JE, Flinn PW 2015. Susceptibility of *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Trogoderma inclusum* (Coleoptera: Dermestidae) to cold temperatures. Journal of Stored Products Research, 64:45–53.

Athanassiou CG, Arthur FH, Hartzer KL 2018. Efficacy of low temperatures for the control of all life stages of *Plodia interpunctella* and *Liposcelis bostrychophila*. Journal of Pest Science, 91:1363–1369.

Athanassiou CG, Papagregorioub AS, Buchelos C 2004. Insecticidal and residual effect of three pyrethroids against *Sitophilus oryzae* (L.) (Coleoptera:Curculionidae) on stored wheat. Journal of Stored Products Research, 40: 289-297.

Baloch MS, Zubair M 2010. Effect of nipping on growth and yield of chickpea. The Journal of Animal and Plant Sciences, 20(3): 208-210.

Boateng BA, Kusi F 2008. Toxicity of Jatropha Seed Oil to *Callosobruchus maculatus* (Coleoptera: Bruchidae) and its Parasitoid, *Dinarmus basali* (Hymenoptera: Pteromalidae). Journal of Applied Sciences Research, 4(8):945-951.

Boyer S, Zhang H, Lempérière G 2012. A review of control methods and resistance mechanisms in stored- product insects. Bulletin of Entomological Research, 102(2):213-229.

Caswell GH 1981. Damage to stored cowpea in the northern part of Nigeria. Samaru Journal of Agricultural Researchrecd, 1(1): 11-19.

Chibbar R 2009. Grain legumes for ensuring nutritional security. Conference on Grain Legumes: Quality Improvement, Value. Addition and Trade, 14-16 February 2009, Indian Institute of Pulses. Research, Kanpur, India.

Clement SL, Wightman JA, Hardie DC, Bailey P, Baker G, McDonald G 2000. Opportunities for integrated management of insect pests of grain legumes. In: Linking research and marketing opportunities for pulses in the 21st century. R. Knight, (ed.), 467-480. Kluwer Academic, ISBN 0-7923-5565-2, Proceedings of the Third International Food Legumes Research Conference. Dordrecht, The Netherlands.

Dohino T, Masaski S, Matsuoka I, Tanno M, Takano T 1999. Low Temperature as an Alternative to Fumigation for Disinfesting Stored Products. Research Bulletin of the Plant Protection Service Japan, 35:5-14.

Donahaye E, Navarro S, Rindner M 1995. Low temperature as an alternative to fumigation for disinfecting dried fruit from three insect species. Journal of Stored Product Research, 1(1): 63-70.

Eaton M, Kells SA 2011. Freeze mortality characteristics of the mold mite *Tyrophagus putrescentiae*, a significant pest of stored products. Journal of Economic Entomology, 104:1423-1429.

Eliopoulos PA, Prasodimou GZ, Pouliou AV 2011. Time-mortality relationships of larvae and adults of grain beetles exposed to extreme cold. Crop Protection, 30(8): 1097-1102.

Faostat 2020. Food and Agriculture Organization of the United Nations. Viale delle Terme di Caracalla, 00153 Rome, Italy. Date of access: 20.12.2020.

Ferizli AG, Emekci M, Tutuncu S, Navarro S 2004. Utilization of freezing temperatures to control *Callosobruchus maculatus* Fab. (Coleoptera, Bruchidae). Integrated Protection of Stored Products-IOBC Bulletin/wprs, 27(9): 213-217.

Fields PG 1992. The control of stored-product insects and mites with extreme temperatures. Journal of Stored Products Research, 28: 89-118.

Fields PG, Muir WE 1996. Physical Control. Chapter 5:195-221. Ed.B. Subramanyamand D.W. Hagstrum, Integrated Management of Insects in Stored Products, 426p.

Fields PG, White NDG 2002. Alternatives to Methyl Bromide treatments for stored-product and quarantine insects. Annual Review of Entomology, 47:331-359.

Finney DJ 1971. Probit analysis. Cambridge University. Press, U.K. Cambridge, 333p.

Flinn PW, Hagstrum DW 1990. Simultaneous comparing the effectiveness of various stored grain management practices and to control. *Rhizopertha dominica*. Environmental Entomology, 19: 725-729.

Gvozdenac S, Tanasković S, Ovuka J, Vukajlović F, Čanak P, Prvulović D, Sedlar A 2019. Low temperature tolerance of *Plodia interpunctella, Sitophilus oryzae* and *Sitophilus zeamais* - the prevalent pests of stored maize in Serbia. Acta Agriculturae Serbica, 24(48): 143-155.

Hill DS 2003. Pests of Stored Food stuffs and Their Control. Kluwer Academic Publisher, 476p.

Homan R, Yubak Dhoj GC 2011. Eco-friendly management of pulse beetle. The Journal of Agriculture and Environment, 12:81-90.

Jat NR, Rana BS, Jat SK 2013. Estimation of losses due to pulse beetle in chickpea. The Bioscan, 8(3): 861-863.

Johnson JA, Valero KA 2000. Control of cowpea weevil, *Callosobruchus maculatus*, using freezing temperatures. Proceedings of the Annual International Research Conference on Methyl Bromide Alternatives and Emission Reductions, Abstract 90-1, November 5-9, 2000, San Diego, CA.

Johnson JA, Valero KA 2003. Use of Commercial Freezers to Control CowpeaWeevil, *Callosobruchus maculatus* (Coleoptera: Bruchidae), in Organic Garbanzo Beans. Journal of Economic Entomology, 96(6): 1952-1957.

Kumar A, Shukla R, Singh P, Singh A, Dubeya N 2009. Use of essential oil from *Mentha arvensis* L. to control storage moulds and insects in stored chickpea. Journal of the Science of Food and Agriculture, 89: 2643-2649.

Kutbay F, Varol I, Bayram M, Ozdemir A 2011. The effect of carbon dioxide at high pressure under different developmental stages of *Callosobruchus maculates* (F) hosting on chickpea. African Journal of Biotechnology, 10(11): 2053-2057.

Loganathan M, Jayas DS, Fields PG, White NDG 2011. Low and high temperatures for the control of cowpea beetle, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) in chickpeas. Journal of Stored Products Research, 47(3): 244-248.

Maharjan R, Yi H, Young Y, Jang Y, Kim Y, Bae S 2017. Effects of low temperatures on the survival and development of *Callosobruchus chinensis* (L.) (Coleoptera: Bruchidae) under different storage durations. Journal of Asia-Pacific Entomology, 20(3): 893-900.

Mason LJ, Strait CA 1998. Stored Product Integrated Pest Management with Extreme Temperatures. Chapter 6. In Hallman G.J. Denlinger, D.I. (Eds.): 141-177p, Temperature sensivity in Insects and Application in Integrated Pest Management. Westview Press, Boulder, USA.

Mullen MA, Arbogast RT 1979. Time-temperature mortality relationships for various stored-product insect eggs and chilling times for selected commodities. Journal of Economic Entomology, 72: 476-478.

Nakakita H 1998. Stored rice and stored product insects. In: Rice Inspection Technology Manual. A.C.E. Corporation, Tokyo, Japan, 49-65.

Ravi R, Harte JB 2009. Milling and physiochemical properties of chickpea (*Cicer arietinum* L.) varieties. Journal of the Science of Food and Agriculture, 89:258-266.

Righi-Assia A, Khelil M, Medjdoub-Bensaad F, Righi K 2010. Efficacy of oils and powders of some medicinal plants in biological control of the pea weevil (*Callosobruchus chinensis* L.) African Journal of Agricultural Research, 5:1474-1481.

Robertson JL, Preisler HK, Russell RM 2003. PoloPlus Probit and Logit Analysis User's Guide; LeOra Software: Petaluma, CA, USA, 36p.

Shaheen FA, Khaliq A 2005. Management of Pulse Beetle, *Callosobruchus chinensis* L. (Coleoptera: Bruchidae) in stored Chickpea using ashes, red soil powder and turpentine oil. Pakistan Entomologist, 27:19-24.

SPSS 2013. IBM SPSS Statistics 21.0 for Windows. Armonk, NY.

Tessema K, Kurabachew H, Tadesse FT 2015. Evaluation of the Efficacy of Plant Powders, Cow Dung Ash and Malathion Dust against *Callosobruchus chinensis* L. (Coleoptera: Bruchidae) On Chickpea in Jole Andegna: Southern Ethiopia. Journal of Agricultural Studies, 3(2):129-144.

Wright VF, Mills RB, Willcutts BJ 1987. Methods for Culturing Stored-Grain Insects. 74-83p. In Toward Insect Resistant Maize for the Third World. Proceedings of the International Syposium on Methodologies for Developing Host Plant Resistance to Maize Insects. CIMMYT, UNDO, GTZ and USAID. 327p.

Zakladnoi GA, Ratanova VF 1987. Stored-Grain Pests and Their Control. Oxonian Press Private Limited. New Delhi, 268p.

Zhong JF, Wan ZH, Li L, Chen HW, Wu GH 2013. Effect of Low and High Temperatures on Controlling Azuki Bean Beetle (*Callosobruchus chinensis* L., Coleoptera: Bruchidae) in Storage. Scientia Agricultura Sinica, 46(1):54-59.