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Some Germination and Emergence Characteristics of Cocklebur (*Xanthium strumarium* L.) Seeds

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HIGHLIGHTS

- The study determined that there was no dormancy in the large seeds of cocklebur, but there was a dormancy state in the small seeds.
- In germination studies conducted with seeds of cocklebur from different provinces; It was found that seeds had a wide germination temperature range between 10 and 40°C.
- The research demonstrated that the application of gibberellic acid (GA_3) had no effect on the germination rates of large seeds. However, it was observed that GA_3 increased the germination rates of small seeds.

Abstract

Cocklebur (*Xanthium strumarium* L.) is an annual broadleaf weed that causes significant yield losses in many crops and reproduces by seed. This study was conducted in 2020-2022 to contribute to alternative control by determining some biological characteristics of cocklebur seeds. For this purpose, in seed studies, the lower (large) and upper (small) seeds in the cocklebur fruit were placed in the incubator at 27±1°C and the germination rates were determined. Furthermore, the minimum, optimum and maximum germination temperatures of cocklebur seeds collected in 23 provinces of Türkiye were also determined. For this purpose, the studies were carried out at temperatures of 5, 10, 15, 20, 25, 30, 35, 40 and 45°C. In the studies to determine the optimum emergence depth of cocklebur; 25 cocklebur fruits were planted in pots at soil depths of 2, 5, 10, 15, 20 and 25 cm. The study found that the maximum germination rate of cocklebur was 93.1% for large seeds, while this rate was 21.2% for small seeds. The study revealed that the highest germination rate of cocklebur in the soil is at a depth of 2-10 cm. In the studies on seed germination temperatures, it was determined that the minimum temperature was 5°C, the optimum temperature was 15-35°C and the maximum temperature was 40°C, although it varied according to the province where seeds were collected. These findings indicate that this weed exhibits a substantial temperature requirement and will spread more in agricultural areas in different ecologies.

Keywords: Germination biology; germination temperature; depth; *Xanthium strumarium*

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1. Introduction

Weeds are defined as plants that are undesirable and grow spontaneously in their habitat. Weeds compete with cultivated plants for mineral nutrients, water, light and space, and due to this competition, they cause yield, quality and quantity losses in products (Güncan and Karaca 2014).

Knowledge of the biology of weeds is important for the development of economical and environmentally friendly weed control methods, which are the basic principles of integrated control. Weed biology refers to plant characteristics such as plant morphology, seed dormancy and germination, growth physiology, competitive ability and reproductive biology. The biological concepts of populations such as the dynamics of seed banks and root reserves in annual plants and the longevity of vegetative reproduction in perennial plants can be used to better the prediction of weed invasion and the evaluation of sustainable management strategies (Bhowmik 1997). Weeds differ in species and genetic diversity depending on geographic region, crop, season and planting date (Pala et. al. 2018). Therefore, the seeds of plants growing in different ecologies have different germination characteristics (Wang et al. 2023). Accurate information on seed germination biology is required for the successful control of any weed species

Cocklebur (*Xanthium strumarium* L.) is an annual, broad-leaved weed that belongs to the Asteraceae family and reproduces by seed (Venodha 2016; Eymirli and Torun 2015; Cesur et al. 2017; Özaslan 2021). The plant is capable of growing to between 20 and 150 centimeters in height (Weaver and Lechowicz 1983) and one plant can produce an average of 70 to 600 fruits (Eymirli and Torun 2015). Each fruit usually contains two seeds, one of which is usually larger than the other, and they differ in their germination requirements (Barton 1962; Hicks 1971).

Cocklebur is an extremely invasive and spreads rapidly. It spreads to agricultural land, pastures, national parks, rivers, dams, roadsides and urban green spaces, causing major problems for biodiversity and significant economic losses (Eymirli and Torun 2015; Tadesse et al. 2017; Chikuruwo et al. 2017; Machado et al. 2021; Özaslan 2021). It has been reported to cause significant yield losses in row crops such as soybean, cotton, corn and peanut in many parts of the world. (Amini and Izadkhah 2013; Tepe 2014; Özaslan 2021). In Turkey, cocklebur is a plant that can grow in almost all regions and under all types of climate and soil conditions (TBL 2024). It has been reported that cocklebur is currently distributed in the Mediterranean, Black Sea and Southeastern Anatolia regions and has the highest density in the Southeastern Anatolia region (Özaslan 2021). Additionally, cocklebur can increase invasion success with its high reproductive capacity, long-term seed survival and ability to adapt to different ecological conditions, as well as rapid spread and dispersal.

Cocklebur not only causes significant yield losses in agricultural areas, but is also a highly toxic plant (Tepe 2014). The toxicity of cocklebur is due to the presence of xanthostrumarin and carboxyatractyloside glycosides in the seeds and seedlings of the plant. These glycosides are particularly harmful to cattle, horses, sheep and pigs. Ingestion of the cotyledons by grazing animals can result in a range of adverse effects, including vomiting, muscle cramps, liver damage and even death (Eymirli and Torun 2015).

Knowing some of the biological characteristics of cocklebur seeds such as germination temperature and emergence depth will help to use alternative control options. In the context of weed control, the emergence depth of weeds is a crucial factor in determining the appropriate tillage depth. This knowledge is also instrumental in determining the tillage depth of herbicides applied prior to sowing. (Güncan ve Karaca 2014). Although there are many studies on the ecological development characteristics of this weed, studies on germination temperatures and soil vigor under different ecological conditions are very limited. For this reason, this study was conducted to determine some germination and emergence characteristics of cocklebur seeds from different geographical regions of Türkiye.

2. Materials and Methods

2.1. Site and Plant Material

The main material of the research consisted of cocklebur fruits collected from 23 provinces in different regions of Turkey. (Table 1). The cocklebur fruits intended for the germination studies were stored in liquid nitrogen for a duration of 1-2 minutes, followed by a cracking process on the wood surface to remove the lower (large) and upper (small) seeds within the fruit.

The seeds were stored at +4°C until they were used in the germination experiments. The cocklebur seeds used to determine seed germination rates and emergence depth were obtained from the Diyarbakır province. The experiments were repeated twice, with four replications, according to the randomized plot design.

Table 1. Regions, provinces and altitudes where cocklebur seeds are obtained.

Regions	Provinces	Climatic properties	Altitudes (m)
Mediterranean	Antalya	Mediterranean	15m
	Adana	Mediterranean	2 m
Eastern Anatolia Region	Bitlis	Terrestrial	1793m
	Erzincan	Terrestrial	1290m
	Elazığ	Terrestrial	1089m
	Malatya	Terrestrial	815m
	Muş	Terrestrial	1959m
	Iğdır	Terrestrial	1455m
	Van	Terrestrial	1770 m
Aegean	Aydın	Mediterranean	22m
	İzmir	Mediterranean	5m
	Afyonkarahisar	Terrestrial	1066m
Southeastern Anatolia	Diyarbakır	Terrestrial	636m
	Batman	Terrestrial	569m
	Mardin	Terrestrial	602m
	Şanlıurfa	Terrestrial	445 m
	Siirt	Terrestrial	615m
Central Anatolia	Kayseri	Terrestrial	1180m
	Konya	Terrestrial	1184m
	Niğde	Terrestrial	1367m
Black Sea	Tokat	Transitionclimate of Black Seaand Central Anatolian	1272m
	Ordu	Temperate	521m
Marmara	Tekirdağ	Mediterranean	176m

2.2. Germination Experiments

The germination rates and germination speeds of the lower and upper seeds of cocklebur were determined according to their position within the fruit. In the experiments, the upper and lower seeds of the weed were placed in 9 cm Petri dishes containing Whatman filter paper No. 1 (moistened with 6 ml pure water). After 20 seeds had been placed in each Petri dish, they were placed in germination cabinets set to a temperature of 27±1°C and a 16-hour light/8-hour dark period. Pure water was added to the Petri dishes as required throughout the experiment. Observations were made every day after sowing. The plants whose radicle length reached 0.5 cm on day 14 were considered germinated and removed from the Petri dish.

Application of gibberellic acid (GA_3); to determine the effect of gibberellic acid application on seed germination, cocklebur seeds were subjected to two different applications of GA_3.

In the first application, the upper and lower seeds were immersed in pure water (control) and different concentrations of GA_3 (500, 1000, 1500, 2000 and 2500 ppm) for a duration of 24 hours (Cesur et al. 2017). After application, the cocklebur seeds were planted in 9 cm petri dishes. Following this, 6 ml of pure water was added to the Petri dishes and placed in germination cabinets.

In the second application, the upper and lower seeds were planted in 9 cm Petri dishes. After planting the seeds, 6 ml of the solution prepared from pure water (control) and different concentrations of GA_3 (500, 1000, 1500, 2000 and 2500 ppm) were added to the Petri dishes (Ateş and Üremiş 2018). After 20 seeds were placed in each petri dish, they were placed in germination cabinets with a temperature of 27±1°C and a 16-hour light / 8-hour dark period. In the control application, pure water was added, while the solution prepared from different GA_3 concentrations was added as required in the other applications.

Since the difference between the two repetitions was insignificant, statistical analyses were performed by calculating the average of the two repetitions of the data obtained from the applications. As a result of the analyses, germination rate (G_{max}) and germination times (T_{50} and T_{90}) were calculated.

$$G_{max} = (G/T) \times 100 \quad (1)$$

G: Number of germinating seeds (number)
T: Total number of seeds used in the experiment (pieces)
T_{50} = Time taken for 50% of the germinated seeds to germinate (days)
T_{90} = Time taken for 90% of the germinated seeds to germinate (days)

2.2.1. Determination of germination temperatures of cocklebur seeds from different provinces

To determine the minimum, optimum and maximum germination temperatures of cocklebur seeds, seeds from 23 provinces were collected in order to represent each region. Large seeds of the cocklebur without dormancy were used for the germination temperature study. After surface sterilization with 1% (NaClO), large seeds were placed in 9 cm Petri dishes containing Whatman filter paper No. 1 (moistened with 6 ml pure water). Each Petri dish placed 10 seeds of similar size. The Petri dishes were then placed in germination cabinets, which were set to constant temperatures of 5, 10, 15, 20, 25, 30, 35, 40 and 45°C. Pure water was added to the Petri dishes as needed during the experiment. Observations were made every day from planting. Those whose radicle length reached of 0.5 cm by the 21st day were considered germinated and removed from the Petri dish (Üremiş and Uygur 1999).

2.2.2. Determination of the emergence depth of cocklebur seeds

In the studies conducted to determine the optimum emergence depth of cocklebur, 25 cocklebur fruits were planted in pots at soil depths of 2, 5, 10, 15, 20 and 25 cm. The soil used for the experiment was sterilized by placing it in an oven at 120°C for a duration of 30 minutes. The mixture of soil, peat and sand (1:1:1) was then added to the pots (Özkil 2021). After sowing was completed, observations were made at one-week intervals for 60 days and the germinated seeds were counted when they had cotyledons or two leaves and were removed from the pot (Erciş et al. 1993). In the evaluations, the emergence percentages were calculated.

2.2.3. Data analysis

The data obtained from the studies was analysed using a one-way Generalised Linear Model (GLM) with Analysis of Variance (ANOVA) in IBM SPSS 25 statistical software package. The difference between the applications was determined according to the significance level $P \leq 0.05$ using the Duncan multiple comparison test.

3. Results

3.1. Seed Germination Trials

In the seed germination experiments, the difference between the germination rates of large and small seeds was found to be statistically significant at the $P \leq 0.05$ level. However, no statistical difference was found in the germination times of the seeds.

In the control application, the maximum germination rate (G_{max}) of the large seeds of cocklebur was found to be 93.1% when pure water was applied, while the maximum germination rate (G_{max}) of the small seeds was 21.2%. When the seeds were evaluated for germination times, it was found that the time taken for 50% of both the large and small seeds to germinate T_{50} was 1.2 days and the time taken for 90% of the germinated seeds to germinate T_{90} was 2.5 days (Table 2).

Table 2. Effect of pure water application on seed germination rates (%) and duration

Pure water application	G_{max}	T_{50}	T_{90}
Large seeds	93.1±1.8a	1.2±0.2a	2.5±0.8a
Small seeds	21.2±1.2b	1.2±0.3a	2.5±0.6a

Each column was evaluated on its own.

When cocklebur seeds were kept in different GA_3 concentrations for a duration of 24 hours, it was found that large seeds showed 100% germination rate with the 24-hour application of 1500 ppm GA_3 and the 24-hour application of pure water (control). In the study, it was found that the application of GA_3 had no effect on the germination rates of large seeds of cocklebur. However, an increase in the concentration of gibberellic acid was observed to result in an increase in the germination rates of small seeds. Specifically, while the lowest germination rate of 45.0% was achieved in small seeds with a 24-hour application of pure water, the highest germination rate of 81.2% was achieved with a 24-hour application of 2000 ppm GA_3. When the effects of the different GA_3 applications on seed germination times were evaluated, the highest T_{50} (germination times of 50% of germinating seeds) and T_{90} (germination times of 90%) times were obtained for large seeds by the 24-hour application of 500 ppm GA_3. Conversely, for small seeds, the statistical difference in the T_{50} and T_{90} times of the applications was not significant (Table 3).

Table 3. Effect of different GA_3 applications on germination rates (%) and duration (days) of cocklebur seeds

	Large seeds			Small seeds		
Treatments	G_{max}	T_{50}	T_{90}	G_{max}	T_{50}	T_{90}
24 hours pure water	$100.0±0.0^{a}$	$1.2±0.2^{b}$	$2.0±0.4^{b}$	$45.0±3.5^{c}$	$1.1±0.9^{a}$	$1.4±0.2^{a}$
24 hours 500 ppm GA_3	$87.5±4.7^{b}$	$4.5±0.9^{a}$	$4.7±1.1^{a}$	$62.5±5.9^{b}$	$1.3±0.0^{a}$	$3.0±1.0^{a}$
24 hours 1000 ppm GA_3	$97.5±2.5^{a}$	$1.2±0.2^{b}$	$1.7±0.4^{b}$	$72.5±2.5^{ab}$	$1.2±0.8^{a}$	$3.5±0.8^{a}$
24 hours 1500 ppm GA_3	$100.0±0.0^{a}$	$2.0±1.0^{b}$	$3.5±0.8^{ab}$	$63.0±7.2^{b}$	$1.3±0.0^{a}$	$1.8±0.3^{a}$
24 hours 2000 ppm GA_3	$95.0±2.8^{ab}$	$1.2±0.2^{b}$	$2.0±0.0^{b}$	$81.2±1.2^{a}$	$1.2±0.8^{a}$	$3.2±0.8^{a}$
24 hours 2500 ppm GA_3	$97.0±2.5^{a}$	$2.0±0.40^{b}$	$2.7±0.7^{ab}$	$75.0±4.1^{ab}$	$1.4±0.8^{a}$	$3.4±0.7^{a}$

Each column was evaluated on its own. Values marked with the same letter are not statistically different. (P>0.05).

Table 4. Effect of different GA_3 applications on germination rates (%) and duration (days) of cocklebur seeds

	Large seeds			Small seeds		
Treatments	G_{max}	T_{50}	T_{90}	G_{max}	T_{50}	T_{90}
Pure water (control)	$98.7±1.2^{a}$	$2.0±0.0^{a}$	$2.0±0.0^{a}$	$18.7±2.3^{c}$	$1.7±0.1^{a}$	$2.5±0.7^{a}$
500 ppm GA_3	$97.5±1.4^{a}$	$1.7±0.2^{a}$	$2.2±0.2^{a}$	$40.0±4.0^{a}$	$1.8±0.4^{a}$	$3.0±0.8^{a}$
1000 ppm GA_3	$100.0±0.0^{a}$	$1.7±0.2^{a}$	$2.0±0.0^{a}$	$25.0±7.9^{bc}$	$1.7±0.1^{a}$	$3.7±0.9^{a}$
1500 ppm GA_3	$98.7±1.25^{a}$	$1.5±0.3^{a}$	$2.0±0.0^{a}$	$33.7±2.3^{ab}$	$2.2±0.8^{a}$	$3.6±0.7^{a}$
2000 ppm GA_3	$100.0±0.0^{a}$	$1.7±0.2^{a}$	$2.0±0.0^{a}$	$42.5±2.5^{a}$	$2.1±0.6^{a}$	$3.7±0.8^{a}$
2500 ppm GA_3	$100.0±0.0^{a}$	$1.7±0.2^{a}$	$2.0±0.0^{a}$	$36.7±3.7^{ab}$	$1.8±0.2^{a}$	$5.5±0.5^{a}$

Each column was evaluated on its own.

When cocklebur seeds of different GA_3 concentrations were added to 6 ml of Petri medium, it was found that the difference between the treatments was not statistically significant in terms of germination rates of the large seeds at the $P \leq 0.05$ level. In the study, it was determined that different concentrations of GA_3 had no effect on the germination rates of large seeds. When the germination rates of small seeds were evaluated, difference between the treatments was found to be statistically significant ($P \leq 0.05$). For small seeds, the lowest germination rate was obtained with the application of pure water (control) at 18.7%, while the highest germination rate was obtained with the application of 2000 ppm GA_3 at 42.5%. When the effect of applications on germination times was evaluated, it was found that the difference between the T_{50} and T_{90} times of large seeds was not statistically significant. When evaluating the effects of small seeds on germination times, it was found that the difference between the T_{50} germination times of the treatments was not statistically significant, while the effects on the T_{90} germination times were significant. The highest T90 germination time for small seeds was achieved with the application of 2500 ppm GA3 at 5.5 days (Table 4).

3.2. Germination Temperatures of Cocklebur Seeds from Different Provinces

In germination studies conducted with large seeds of cocklebur from different provinces; it was found that the lowest germination rates were at temperatures of 5 °C and 40°C. When the germination rates of cocklebur seeds at 5°C were examined, it was found that there was no statistical difference between the provinces. Notably, no germination was observed in 14 provinces (Bitlis, Diyarbakır, Elazığ, Iğdır, İzmir, Kayseri, Konya, Malatya, Mardin, Muş, Siirt, Şanlıurfa, Tokat, Van), while a very low germination rate of 2.5% was recorded in 9 provinces (Adana, Afyon, Antalya, Aydın, Batman, Erzincan, Niğde, Ordu, Tekirdağ). In the experiments, the most suitable temperature for seed germination was determined as 15-35°C, although it varies by province. (Table 5).

Table 5. Minimum, optimum and maximum germination temperatures of cocklebur from different provinces

Provinces	5°C	10°C	15°C	20°C	25°C	30°C	35°C	40°C	45 °C
Adana	2.5 ±1.4$_{a}^{D}$	65.0±6.1$_{a}^{B}$	96.0±2.6$_{a}^{A}$	97.5±2.5$_{a}^{A}$	100.0±0$_{a}^{A}$	100.0±0a^{A}	98.7±1.2$_{a}^{A}$	35.0±3.5$_{ab}^{C}$	0.0±0.0$_{a}^{D}$
Afyon	2.5±1.4$_{a}^{D}$	30.0±7.9$_{d}^{B}$	67.5±3.2$_{bc}^{A}$	81.2±2.3$_{def}^{A}$	81.2±1.2$_{f}^{A}$	81.2±5.5$_{f}^{A}$	81.2±3.7$_{cde}^{A}$	16.2±7.4$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Antalya	2.5±2.5$_{a}^{E}$	61.2±3.7$_{ab}^{C}$	73.7±3.1$_{b}^{B}$	100.0±0$_{a}^{A}$	98.7±1.2$_{ab}^{A}$	100.0±0$_{a}^{A}$	95.0±3.5$_{ab}^{A}$	11.2±5.1$_{cde}^{D}$	0.0±0.0$_{a}^{E}$
Aydın	2.5±1.4$_{a}^{D}$	7.5±1.4$_{e}^{D}$	43.7±5.5$_{cd}^{C}$	78.7±5.1$_{def}^{B}$	98.7±1.2$_{ab}^{A}$	92.5±3.2$_{abcd}^{A}$	93.7±1.2$_{ab}^{A}$	6.2±3.1$_{e}^{D}$	0.0±0.0$_{a}^{D}$
Batman	2.5±1.4$_{a}^{C}$	41.2±3.1$_{bc}^{B}$	70.0±14.8$_{b}^{A}$	84.7±4.6$_{bcde}^{A}$	85.0±2.8$_{ef}^{A}$	86.2±3.1$_{def}^{A}$	87.5±4.7$_{abcde}^{A}$	17.5±5.2$_{bcde}^{C}$	0.0±0.0$_{a}^{C}$
Bitlis	0.0±0.0$_{a}^{F}$	8.7±1.2$_{e}^{E}$	47.5±4.7$_{bcd}^{C}$	88.7±3.1$_{abcd}^{AB}$	92.5±1.4$_{bcd}^{A}$	91.2±2.3$_{abcde}^{AB}$	83.7±3.7$_{bcde}^{B}$	17.5±1.4$_{bcde}^{D}$	0.0±0.0$_{a}^{F}$
Diyarbakır	0.0±0.0$_{a}^{D}$	5.0±3.5$_{e}^{D}$	72.7±8.1$_{b}^{B}$	95.0±2.0$_{abc}^{A}$	95.0±0$_{abcd}^{A}$	92.5±2.5$_{abcd}^{A}$	100.0±0.0$_{a}^{A}$	20.0±7.0$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Elazığ	0.0±0.0$_{a}^{D}$	2.5±1.4$_{e}^{D}$	26.7±4.1$_{d}^{C}$	71.7±3.8$_{fg}^{B}$	98.7±1.2$_{ab}^{A}$	93.7±1.2$_{abcd}^{A}$	76.2±6.5$_{e}^{B}$	21.2±5.1$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Erzincan	2.5±2.5$_{a}^{EF}$	13.7±4.2$_{e}^{DE}$	52.5±2.5$_{c}^{C}$	77.5±10.3$_{def}^{B}$	93.7±1.2$_{abcd}^{A}$	83.7±3.1$_{ef}^{AB}$	80.7±1.4$_{cde}^{B}$	17.5±5.9$_{bcde}^{D}$	0.0±0.0$_{a}^{F}$
Iğdır	0.0 ±0.0$_{a}^{D}$	6.2±4.7$_{e}^{D}$	77.5±7.5$_{b}^{B}$	97.5±2.5$_{a}^{A}$	100.0±0$_{a}^{A}$	100.0±0$_{a}^{A}$	98.7±1.2$_{a}^{A}$	50.2±1.6$_{a}^{C}$	0.0±0.0$_{a}^{D}$
İzmir	0.0 ±0.0$_{a}^{D}$	3.7±2.3$_{e}^{D}$	18.7±3.7$_{d}^{C}$	74.7±5.2$_{efg}^{B}$	73.7±3.7$_{g}^{B}$	95.0±2.8$_{abcd}^{A}$	100.0±0$_{a}^{A}$	12.5±6.2$_{cde}^{CD}$	0.0±0.0$_{a}^{D}$
Kayseri	0.0 ±0.0$_{a}^{D}$	38.0±9.3$_{cd}^{C}$	75.0±2.0$_{b}^{B}$	95.0±0$_{abc}^{A}$	93.7±1.2$_{abcd}^{A}$	95.0±2.0$_{abcd}^{A}$	76.2±5.5$_{e}^{B}$	7.5±4.7$_{de}^{D}$	0.0±0.0$_{a}^{D}$
Konya	0.0 ±0.0$_{a}^{C}$	1.2±1.2$_{e}^{C}$	81.2±5.1$_{ab}^{B}$	93.7±3.7$_{abc}^{A}$	95.0±2.0$_{abcd}^{A}$	92.5±4.3$_{abcd}^{A}$	93.7±2.3$_{ab}^{A}$	7.5±3.2$_{de}^{C}$	0.0±0.0$_{a}^{C}$
Malatya	0.0±0.0$_{a}^{D}$	7.5±3.2$_{e}^{CD}$	62.5±6.6$_{bc}^{B}$	90.0±2.0$_{abcd}^{A}$	100.0±0$_{a}^{A}$	97.5±1.4$_{abc}^{A}$	98.7±1.2$_{a}^{A}$	16.2±6.2$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Mardin	0.0 ±0.0$_{a}^{F}$	2.5±2.5$_{e}^{F}$	22.5±2.5$_{d}^{E}$	78.7±2.3$_{def}^{C}$	88.7±1.2$_{de}^{B}$	92.5±1.4$_{abcd}^{B}$	98.7±1.2$_{a}^{A}$	28.7±3.7$_{bc}^{D}$	0.0±0.0$_{a}^{F}$
Muş	0.0±0.0$_{a}^{F}$	12.5±4.7$_{e}^{E}$	26.2±3.7$_{ed}^{D}$	66.7±4.8$_{g}^{C}$	91.2±1.2$_{cde}^{A}$	96.2±1.2$_{abc}^{A}$	80.0±3.5$_{cde}^{B}$	10.0±2.0$_{cde}^{E}$	0.0±0.0$_{a}^{F}$
Niğde	2.5±1.4$_{a}^{D}$	10±4.5$_{e}^{D}$	30.0±3.5$_{d}^{C}$	82.5±4.7$_{cdef}^{B}$	92.5±3.2$_{bcd}^{AB}$	96.2±1.2$_{abc}^{A}$	91.2±1.2$_{abc}^{AB}$	27.5±7.5$_{bcd}^{D}$	0.0±0.0$_{a}^{D}$
Ordu	2.5 ±1.4$_{a}^{D}$	50.0±3.5$_{bc}^{C}$	78.7±7.7$_{ab}^{B}$	96.2±1.2$_{ab}^{AB}$	97.5±1.4$_{abc}^{AB}$	100.0±0$_{a}^{A}$	95.0±0.0$_{ab}^{AB}$	36.2±15.9$_{ab}^{C}$	0.0±0.0$_{a}^{D}$
Siirt	0.0±0.0$_{a}^{D}$	5.0±2.0$_{e}^{D}$	49.0±2.4 $_{bc}^{C}$	90.0±2.8$_{abcd}^{A}$	95.0±2.0$_{abcd}^{A}$	90.0±2.0$_{bcde}^{A}$	78.7±4.2$_{de}^{B}$	6.2±3.7$_{e}^{D}$	0.0±0.0$_{a}^{D}$
Şanlıurfa	0.0±0.0$_{a}^{F}$	8.7±2.3$_{e}^{EF}$	16.2±3.1$_{d}^{E}$	63.7±6.5$_{g}^{C}$	81.2±2.3$_{f}^{B}$	93.7±2.3$_{abcd}^{A}$	97.5±1.4$_{a}^{A}$	34.0±5.5$_{ab}^{D}$	0.0±0.0$_{a}^{F}$
Tekirdağ	2.5±2.5$_{a}^{E}$	48.7±3.1$_{c}^{C}$	65.0±7.3$_{bc}^{B}$	90.0±0$_{abcd}^{A}$	95.0±3.5$_{abcd}^{A}$	98.7±1.2$_{a}^{A}$	88.7±3.7$_{abcd}^{A}$	36.2±6.2$_{ab}^{D}$	0.0±0.0$_{a}^{E}$
Tokat	0.0±0.0$_{a}^{D}$	7.5±1.4$_{e}^{D}$	51.2±4.2$_{c}^{B}$	83.7±1.2$_{bcdef}^{A}$	93.7±1.2$_{abcd}^{A}$	90.0±4.0$_{bcde}^{A}$	85.0±6.7$_{bcde}^{A}$	18.7±6.2$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Van	0.0±0.0$_{a}^{D}$	10.0±2.8$_{e}^{D}$	41.2±3.1$_{bcd}^{C}$	96.2±1.2$_{ab}^{A}$	85.0±4.5$_{ef}^{A}$	88.7±3.7$_{cdef}^{A}$	62.5±8.5$_{f}^{B}$	6.2±4.7$_{e}^{D}$	0.0±0.0$_{a}^{D}$

While uppercase letters represent groups between temperatures, lowercase letters represent groups between provinces ($P \leq 0.05$).

3.3. Determination of the Emergence Depth of Cocklebur Seeds

The studies show that the emergence rates of cocklebur at soil depths of 2, 5 and 10 cm are statistically in the same group and have the highest emergence rate. The highest emergence rates of cocklebur seeds at soil depths were observed to be 90.6% at a depth of 2 cm, 99.3% at a depth of 5 cm and 91.8% at a depth of 10 cm. The lowest emergence rate of cocklebur fruits was observed at a soil depth of 25 cm with 0.6% (Table 6).

Table 6. Emergence rates of cocklebur seeds according to different soil depths (%)

Soil depths (cm)	Emergence rate (%)
2	90.6a±4.8
5	99.3a±0.3
10	91.8a±4.0
15	50.0b±5.1
20	36.2c±4.8
25	0.6d±0.3

Values marked with the same letter are statistically the same ($P>0.05$).

4. Discussion

Studies have shown that dormancy is not to be found in the large seeds of the cocklebur, but in the small seeds. Similar to the results of our study, various researchers reported in their studies on dormancy that there are two seeds in the cocklebur, that the germination of these seeds is different, that there is no dormancy in the large seeds, that the small seeds do not germinate under normal growing conditions, and that they germinate about one year after the germination of the large seeds (Barton 1962; Esashi et al. 1983).

In the study to determine the highest germination rate of seeds as a result of different applications of GA_3, the highest germination rate of large seeds, 100%, was obtained by applying 1500 ppm GA_3 for 24 hours. Cesur et al. (2017) reported that the highest germination rate of 100% in different GA_3 applications in the germination of cocklebur seeds was obtained by applying 1500 ppm GA_3 for 24 hours, and they provided the same results as our study. Karimmojeni et al. (2010) reported in dormancy-breaking studies that the combination of cold (5±2 °C for 2 months) and GA_3 led to a slight increase in germination in cocklebur seeds. In the results of the study, it was found that the different applications of GA_3 had no effect on the germination of large seeds, but there was a slight increase in small seeds. In the study, it was found that different applications of GA_3 had no effect on the T_{50} and T_{90} germination times of cocklebur seeds. Similar to our study, Cesur et al. (2017) reported that different applications of GA_3 for 24 hours was had no effect on the germination time of seeds.

The study revealed that the optimal temperature range for cocklebur seeds in the Mediterranean region is 15-35°C, with a minimum of 5°C and a maximum of 40°C. Similar to our study results, in the study conducted by Kadıoğlu (1997) in the Mediterranean region, it was reported that the optimum germination temperature of cocklebur seeds was 25-30°C, with a maximum of 40°C. In the temperature studies conducted in the Aegean region, it was determined that the cocklebur seeds germinated under a minimum of 10°C, while the maximum germinated temperature was recorded as 35°C. (Kaya and Nemli 2004). In the present studies, the minimum temperature in the Aegean region was found to be 5°C, the optimum temperature was determined to be 25-35°C, and the maximum temperature was found to be 40°C. These results are consistent with the studies conducted in the Aegean region. When the germination temperature of cocklebur seeds in the Southeastern Anatolia region was examined, it was found that the highest seed germination rate was obtained at 35°C. This finding aligns with the results of other researchers, who have also reported that the optimal germination temperature for cocklebur seeds ranges from 25 to 40 °C (Bükün 1997; Norsworthy and Oliveira 2007; Saric-Krsmanovic et al. 2012; Goharian et al. 2019). In studies conducted in different countries, Saed et al. (2020) found that germination of cocklebur seeds in Pakistan occurred at temperatures between 10 and 45 °C, and Ahmedi et al. (2024) found that seeds germinated at temperatures between 10 and 40 °C in Iran. The study shows that there are differences in the germination temperatures of seeds from different provinces. In the study conducted by Karaman and Tursun (2021), it was reported that the germination temperatures of seeds grown in different ecological regions differ.

In the study, it was found that the highest emergence rate of cocklebur seeds in soil was at a depth of 2 to 10 cm, and the lowest emergence rate was at a depth of 25 cm. Similar our study, Kadıoğlu (1997) found that the highest emergence rate of cocklebur was 2 to 10 cm at different soil depths and the lowest emergence was 25 cm. In a separate study, Tao et al. (2021) reported that the majority of cocklebur fruits emerged between 0 and 3 cm, while Toledo et al. (1993) stated that the highest emergence rate of hogweed seeds occurred at a soil depth of 0 to 8 cm James et al. (2016) have stated that there is no emergence of cocklebur seeds at a depth of 10 cm. However, the our study found that emergence occurred even at a depth of 25 cm.

5. Conclusions

In the study, it should be taken into consideration that there are differences in the germination temperatures of seeds from different provinces and therefore the cocklebur control period may vary from province to province. A further finding of the study is that the seeds have a wide germination temperature range between 10 and 40 °C, which increases the invasive character of this weed species and causes considerable damage to the ecosystem and agricultural land. One of the most important practices in weed control is tillage. Tillage can influence on the distribution and germination of weed seeds in the soil. This study will help determine the depth of tillage by determining the depth of cocklebur emergence from the soil. Thus, it will help reduce the population of this weed species in agricultural areas by indicating to growers the depth at which tillage should be done. The results of this research will form the basis for future scientific studies to control the damage that cocklebur may cause in agricultural areas.

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