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EFFECTS OF DIFFERENT SEASONS AND SOWING DENSITY ON YIELD PROPERTIES AND FATTY ACID COMPOSITIONS OF MILK THISTLE *(SILYBUM MARIANUM)*

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Abstract

The study was conducted to determine the effects of spring-fall seeding and different row and intra-row spacing on seed yield and yield properties of Milk Thistle plant in Tokat. The experiment was conducted during 2010-2011 and 2011-2012 growing seasons under Tokat-Kazova conditions. Seeds were planted in spring and fall seeding in the main plots two different row spacings (25 and 50 cm) and four different intra-row spacings (10, 20, 30 and 40 cm) in the sub plots. In this study, plant height, seed yield, 1000 seed weight, oil ratio, oil yield and oil composition were investigated. According to the findings, seed yield varied from 37,7 to 672,2 kg/da, and oil ratio % 20,9 to 25,5. The average seed yield of fall seeding (330,4 kg/da) was above that of spring seeding (107,4 kg/da). The seed yield and oil rate was higher (respectively, 25x20, 25x10 and 50x10 cm) than the other applications. Furthermore, oil ratio is approximately 4,9% higher in spring seeding than that in fall seeding. In terms of fatty acids, the ratios of linoleic acid, oleic acid, palmitic acid, and stearic acid in fall seeding and spring seeding are 43.77% and 46.87%, 31.31% and 28.78%, 8.93% and 8.47%, and 6.72% and 6.68%, respectively.

Keywords: Milk Thistle, Oil ratio and fatty acids, plant density, Seed yield, *Silybum marianum*, Spring and fall seeding

INTRODUCTION

As one of the medicinal plants which has recently come to the fore and been the subject of different studies in Turkey, Milk Thistle is an annual plant within the genus *Silybum* of the family *Asteraceae (Compositae*), both growing naturally and being commercially cultivated (Sanchez-Sampedro et al. 2008). Milk Thistle seeds contain 1% to 6% silymarin. Produced in America and Europe, condensed Milk Thistle extracts have a silymarin content of 70% to 80%. Silymarin is a pharmacologically active substance contained in Milk Thistle seeds and composed of flavonolignans such as silychristin, silydianin, diastereoisomers silybin and isosilybin (Gümüşçü et al. 1998; Demirezer et al. 2007; Sanchez-Sampedro et al. 2008). Many Milk Thistle seed preparations (either unaccompanied or as a mixture) are commercially available in the market and recommended for the treatment of toxicant-induced liver injury, cirrhosis and chronic liver injuries (Mills and Bone 2000; Sanchez-Sampedro et al. 2008).

Milk Thistle plant grows in South-West Europe, Australia, North Africa and Western Asia. It is widespread in Mediterranean countries and the southern parts of Russia and naturally exists in Kashmir, India and Pakistan. In Turkey, Milk Thistle grows along the roads and fields and in the wastelands of İzmir, Aydın, Denizli, Mersin, Adana and Antakya in Western and Southern Anatolia and of the coastal areas of Marmara Region (Gümüşçü et al. 1998). Consumed as a vegetable especially in the countries around the Mediterranean Sea, Milk Thistle has been used in treating liver diseases for long years. Every part of the plant can be used for nutritive or other purposes (Meriçli, 1984).

A study was conducted for the purpose of determining the effects of different sowing times (22 September, 1 March, 12 April, and 26 April) on yield and some plant characteristics in milk thistle (*Silybum marianum*) under the ecological conditions of Ankara. The highest seed yield (64.5 kg/da) was obtained in September, while 26 April produced the lowest one (31.5 kg/da) (Gümüşçü et al. 1998).

In 1996 and 1997, a study was performed to determine the effects of different sowing densities on yield in milk thistle (*Silybum marianum*) under the ecological conditions of Ankara. As a result of the study, the highest yield (93.9 kg/da) and the lowest yield (65.7 kg/da) were obtained from the combination of 60x10 cm-purple and of 60x50 cm-white, respectively (Gürbüz et al. 1998).

In order to determine the effects of different sowing times and densities on yield and flavonolignan content in milk thistle, a study was carried out in Poland between the years of 2004 and 2006. As a result, average seed yield was found to be 123 kg/da, and a silymarin content of 2.65 kg/da was obtained. However, with delayed sowing time, silymarin content increased by approximately 0.4%. In fruits, average silymarin content obtained was 2.18%. The ratio of silydianin to silychristin was 1:2.2, and the ratio of silybinin to isosilybinin in silydianin was 1:3.3 (Andrzejewska et al. 2010).

Germany is the leading country in the production of *Silybum marianum*. It is also produced in California on a large scale. In California and Germany, seeds are used in herbal medicine industry.

Milk Thistle seeds contain fixed oil (20%-30%), starch, tannin, silymarins (silybin, silydianin and slychristin) as well as tyramin, flavonid, histamine, resin, amine, albumin and agmatine (Tanker et al. 2003). According to a study, 75.1% of total fatty acids in seeds are unsaturated fatty acids (Tanker et al. 2007). *S. marianum* seeds are reported to be rich in linoleic acids (50%-60%), followed by oleic acids (20%-30%), palmitic acids (9%) and stearic acids (6.6%) (Wagner et al. 1986; El-Mallah et al. 2003).

In a study conducted on *S. marianum* seeds in Iran, the focal point was fatty acid amount and components, tocopherols, and antiradical activity. To determine antiradical activity, tocopherol amount (Vitamin E), and fatty acid components, researchers used DPPH radical, HPLC, and GC, respectively. The results of the study revealed that total oil content was 25%. Besides, they identified 9 fatty acids; namely, palmitic acid (8.25%), palmeotic acid (0.07%), stearic acid (6.67%), oleic acid (31.58%), isomer oleic acid (0.53%), linoleic acid (45.36%), linolenic acid (0.87%), arachidic acid (4.11%), eicozantoic acid (0.088%) and behenic acid (2,6%). α, γ, δ-tocopherol contents were calculated as 563.157 mg, 88.87 mg, and 163.791 mg, respectively. The studies performed suggest that *S. marianum* seed is a rich source of antioxidants (Hasanloo et al. 2008).

This study was conducted to determine the effects of spring-fall seeding and row and intra-row spacing on yield and yield properties in the seeds of Milk Thistle plant, which can be cultivated in Tokat region.

MATERIALS AND METHODS

Materials

This study was carried out 2010-2011 and 2011-2012 growing seasons in Tokat-Kazova conditions. Milk Thistle (*Silybum marianum*) seeds were taken in Ankara University, Agriculture Facultuy.

Methods

Experimental design was split plot in a randomized complete block with three replications. Seeds were sowed as two row spacings (25 and 50 cm) and four intra-row spacings (10, 20, 30 and

40 cm) on October and March months. Each plot consisted of four rows which were 4 m long. Equal amounts of nitrogen (10 kg/ha N), phosphorus (80 kg/ha P2O5) were applied homogenously to the plots. Half of nitrogen and all phosphorus were given to prepared plots and mixed with the soil before the transplanting of seedlings. The other half of nitrogen was given in bolting time. Study was conducted under rainfed conditions. The seeds were harvested during the physiological maturity period of milk thistle crops.

Oil Extraction

For each different sowing seasons and density, approximately 50 grams of milk thistle seed were ground by a laboratory blender. Of the said 50 grams of ground milk thistle seed, 10 grams were extracted with 30 mL hexane for 24 hours at room temperature. Following filtering by a filter paper and transferring to a tared flask, the solvent of the filtrate was removed under low pressure at 40°C. The total amount of oil was calculated based on 0% humidity, taking weight loss into consideration. For fatty acid analysis, the oil obtained was put into amber glass containers and kept at +4°C (Ahmadzadeh et al. 2013).

Formation of Fatty Acid Methyl Esters

For fatty acid analysis, fatty acid methyl esters were formed. To do so, 30 milligrams of extracted oil was solved in 3 mL hexane. After adding 3 M KOH solution prepared in 3 mL methanol, it was vortexed for 3 minutes. Then, it was kept waiting for 5-10 minutes at room temperature for phasing. 0,5 mL hexane containing methyl esters was put into vials and analyzed by GC-FID. The amount of each fatty acid was determined in percentages according to the rate of peak areas. In the determination of fatty acids, standard fatty acid mixture (Supelco 37 Component FAME mix 47885-U) was used (Demirtas et al. 2011).

Gas Chromatography Analysis Program

Fatty acids were analyzed by Perkin Elmer Clarus 500 Series gas chromatography (GC) using a FID (Flame Lonization Detector) detector. Apolar capillary column (TR-FAME 30 m X 0,25 mm X 0,25µm I.D) was employed. Split rate was set to 50:1. As the carrier gas, helium was used at a flow rate of 0,5 ml/min. Injection temperature and detector temperature were set to 250ºC and 260ºC, respectively. Being originally 100ºC, column furnace temperature was increased for 2ºC per minute and fixed to 220ºC.

Statistical Analysis

The results obtained were subjected to variance analyses according to Randomized Block Design. Averages were compared in accordance with Duncan Test (Açıkgöz 1993).

RESULTS AND DISCUSSION

In the research, the average plant height of milk thistles sowed in summer was 66.9 cm in 2010 and 53.8 cm in 2011. The highest value was found to be 74.4 cm (50x10 cm) in 2010 and 64.7 cm (50x40 cm) in 2011. On the other hand, in milk thistles sowed in winter, average plant height was 147.0 cm in 2010 and 127.6 cm in 2011. In winter sowing, the highest value was 154.3 cm (25x40 cm) and 134.2 cm (50x20cm) in 2010 and 2011, respectively. As can be seen, milk thistles sowed in winter were at least two times higher than those sowed in summer (Table 1).

It was determined that thousand-seed weights varied between 27.2 g and 29.3 g in summer sowing and between 25.8 g and 28.8 g in winter sowing and that milk thistles sowed in summer had a higher thousand-seed weight, which was not found to be statistically significant (Table 1). Statistical differences between sowing densities were not significant in summer sowing in the first year and in winter sowing in the second year, and a higher thousand-seed weight was obtained from the density of 50x10 cm.

Average seed yield was 165.0 kg/da in 2010 and 49.7 kg/da in 2011 in milk thistles sowed in summer but 469.5 kg/da in 2010 and 191.2 kg/da in 2011 in those sowed in winter. Two years' average was found as 107.3 kg/da for summer sowing but as 330.4 kg/da for winter sowing, demonstrating that the yield obtained from winter sowing was nearly three times higher than the yield obtained from summer sowing. It was seen that the effect of sowing densities on milk thistle plant seed yield was statistically significant and a higher sowing density generally provided higher yield. It was further determined that the densities of 25x10 cm and 25x20 cm were suitable for winter sowing and it could be 25x30 cm for summer sowing (Table 1).

The findings of the research indicate that, for obtaining a higher yield in milk thistle, the plant needs to undergo a very good vegetative development. In fact, the plants sowed in winter were at least two times higher and had nearly three times higher seed yield per decare. In the research, the plants sowed in winter went through a cooler rosette period and had a very good root development, resulting in a more positive effect on plant yield parameters for future periods. As a consequence, plants were less affected by ecological restrictions and had a higher yield.

While evaluating the research from the aspect of sowing density, row spacing (25 and 50 cm) and the number of plants per unit area (5, 10, 13, 20 and 40 plants/m2) along with intra-row spacing are required to be taken into account. A row spacing of 25 cm produced better results than 50 cm in both summer and winter sowings. For the plants sowed in winter, denser sowing led to increased number of plants per unit area. Having a more confined space, the plant had less sub-branches and fruits (small table) but larger fruits on the main stem. As a consequence of more plants per unit area and larger fruits as well as more seeds, seed yield per decare increased. For the plants sowed in summer, however, seed yield per decare decreased due to thinner sowing (row spacing of 50 cm). It is possible to say that, in summer sowing, soil moisture is used more efficiently if row spacing is less (25 cm). If row spacing is more, higher amount of water is lost because of higher evapotranspiration. In terms of the number of plants per unit area, the highest yield (672.7 kg/da) was obtained from 20 plants/m^2 (25x20 cm) in winter sowing, a value higher than 5, 10, 13 and 40 seeds/m^2 yields. For this reason, if 20 seeds are sowed per m^2 (25x20 cm), plants can better make use of environmental conditions and thus higher yields and better yield elements can be ensured.

In this study, a higher oil ratio was obtained from the milk thistle plants sowed in summer compared to those sowed in winter (Table 2). Oil ratio did not differ by sowing densities in milk thistle plants sowed in winter. But, in those sowed in summer, differences were found significant. However, sowing densities other than 50x30 and 50x40 cm were in the same group. In plants like milk thistle, not only oil ratio but also oil yield per decare is important. Accordingly, oil yield per decare ranged from 7.9 kg/da to 50.7 kg/da in summer sowing and from 23.7 kg/da to 162.8 kg/da in winter sowing, depending on sowing density, and was found to be statistically significant. Oil ratio is higher in summer sowing, while winter sowing has a higher seed yield. The highest oil yield was produced by the densities of 25x20 and 25x30 cm in summer sowing and by the densities of 25x10 and 25x20 cm in winter sowing.

As a result of this study, however, milk thistle plants sowed in winter were found to be prominently superior to those sowed in summer in terms of numerous morphological and yield-related parameters. With regard to the relation between sowing density and seed yield, the highest seed yield per decare was obtained from the sowing density of 25x20 cm. In consequence, milk thistle plant can be commercially produced under the ecological conditions of Tokat, Kazova but, for higher seed and oil yields, is recommended to be sowed in winter with a sowing density of 25x20 cm.

It is a known fact that, in oilseeds, the effect of temperature on oil synthesis differs by plant species, ecology, and genetic structure. In a study, oil ratio in the seeds of Milk Thistle plants sowed in summer was found to be higher. However, high temperature and insufficient humidity resulted in lower yield, giving rise to lower oil yield. In summer sowing, oil synthesis in seeds is generally higher as a result of higher temperature. In winter sowing, however, unless any cold damage occurs, plants grow much better, below- and above-ground parts are stronger, and factors positively affecting yield improve. As a result, seed yield per decare increases. Katar et al. (2012) carried out studies on the subject and analyzed the effects of 8 different sowing times on oil ratio, oil yield and fatty acids in camelina plant. Eventually, they obtained the highest oil ratio from the plants sowed in November 15 and the lowest oil ratio from the ones sowed in May 1. On the other hand, the highest oil yield was produced by the first sowing date, October 1. Flagella et al. (2002) examined the effects of different sowing times on yield and yield components in hybrid sunflower plant. At the end of the study, they found that the oil ratio derived from the plants sowed earlier was higher. From the plants sowed earlier, they obtained higher seed yield and consequently higher oil yield. In another study, the effects of different sowing times on yield and yield components in safflower plant were analyzed. The researchers concluded that the effect of different sowing times on oil ratio was insignificant (Keles and Oztürk 2012).

As revealed by this study, fixed oil obtained from the seeds of Milk Thistle plant contains 16 different fatty acids (myristic, decanoic, decenoic, palmitic, palmiteoleic, heptadecanoic, heptadecenoic, stearic, oleic, linoleic, linolenic, arachidic, eicosanoic, arachidonic, cis-8,11,14-eicosatrienoic acid and eicosapentanoic). 8 of the said fatty acids (palmitic, stearic, oleic, linoleic, arachidic, eicosanoic, cis-8,11,14-eicosatrienoic and eicosapentanoic) had a higher ratio and were found to be statistically significant. Tables 3 and 4 show the distribution of those 8 fatty acids.

Findings related to the distribution of fatty acids in the seeds of Milk Thistle plants sowed in winter are given in Table 3. Accordingly, the effect of different sowing densities on the changes in stearic, oleic, linoleic, arachidic, cis-8,11,14-eicosatrienoic and eicosapentanoic fatty acids was not statistically significant. However, changes in palmitic ($p<0.05$) and eicosanoic ($p<0.001$) fatty acids were found to be statistically significant. The highest palmitic fatty acid ratio was obtained from the sowing density of 50x10 cm, while the sowing density of 25x10 cm resulted in the lowest palmitic fatty acid ratio. In eicosanoic fatty acids, the highest ratio and the lowest ratio were produced by the sowing densities of 25x30 cm and 25x10 cm, respectively (Table 3). As can be seen in the figures, increase in the number of plants per unit area gave rise to a decrease in palmitic and eicosanoic fatty acid ratios.

In the seeds of Milk Thistle plants sowed in summer, different sowing densities had a statistically significant ($p<0.05$) effect only on the change in eicosanoic fatty acid ratio (Table 4). The highest eicosanoic fatty acid ratio was obtained from the sowing density of 25x20 cm. However, the lowest was observed in the sowing density of 50x10 cm. Upon examining each fatty acid group in itself, it was concluded that, compared to thick sowing, thin sowing positively affected fatty acids other than linoleic acid ratio and that more linoleic acids were synthesized in thick sowing. Besides, linoleic acid synthesis was higher in the seeds of Milk Thistle plants sowed in summer (Table 5).

Milk Thistle seeds' fatty acid ratio varied by sowing time; namely, summer and winter. Accordingly, palmitic ($p<0.01$), oleic ($p<0.001$), linoleic ($p<0.001$), arachidic ($p<0.001$) and eicosapentanoic ($p<0.05$) fatty acids' changes were statistically significant (Table 5). In winter sowing, palmitic, oleic and arachidic fatty acids' ratios were higher. On the other hand, linoleic and eicosapentanoic fatty acids had higher ratios in summer sowing.

In Milk Thistle sown in summer, oleic acid ratio decreased, while linoleic fatty acid ratio increased. As studies demonstrate, with temperature rise, there may be a decrease in the activity of the enzymes that catalyze the synthesis of linoleic and linolenic acids from oleic acid such as oleayl-PC desaturase and linoleayl-PC desaturase, respectively (Broun and Somerville 1997). As a result, high temperature can sometimes affect linoleic and linolenic acid synthesis negatively but oleic acid synthesis positively (Weiss 1983; Stryer 1986; Röbbelen et al. 1989). Pritchard et al. (2006), in a study they conducted in Victoria, presented that fatty acid composition in rapeseed (*Brassica napus*) varied by regions and years. While oleic acid content ranged from 0.4% to 60.3%, low temperature and precipitation reduced oleic acid content. In spite of higher temperature and lower precipitation, linoleic (0.3%-19.7%) and linolenic acid (0.3%-10.4%) had lower variability between regions. In a study performed by Gororo et al. (2003) in the leading rapeseed producing regions of Australia for 3 years, saturated fatty acid ratios were found to be lower in the rapeseeds grown in the regions with higher temperature.

Not different sowing densities but sowing time (summer and winter) had a statistically significant effect on the change in the oil ratios of Milk Thistle plant seeds. Compared to winter sowing, summer sowing led to an increased oil ratio of 3.9% on average. On the other hand, oil yield per decare was considerably lower in summer sowing than that in winter sowing. The reason of a higher oil yield per unit area in winter sowing compared to summer sowing is higher seed yield. The significant point here is seed and oil yield per unit area. Milk Thistle seeds' oil ratio is approximately 25% on average; oil yield per decare is nearly 110 kg in winter sowing and 42 kg in summer sowing. Therefore, it is recommended that, under the ecological conditions of Kazova, Tokat, Milk Thistle is sowed in winter and each decare is populated by minimum 10.000 plants.

As revealed by this study, thick sowing gave better results than thin sowing and the sowing density of 25 cm was more advantageous than that of 50 cm. Likewise, although there was no difference between intra-row spacing, thick sowing gave better results. In a study carried out by Yılmaz et al. (2013), thick sowing of Milk Thistle plants produced fewer branches. As a result, plants to be harvested were more homogenous than the ones sowed thinly. The same researchers reported that, in thin sowing, depending on branch creation, there were more differences between the fruits on each branch in terms of growth and maturation. Further density studies can be performed on the subject.

In Milk Thistle seeds sowed in winter, the effect of sowing density on fatty acid distribution was found to be statistically significant only in palmitic and eicosanoic acid content. The sowing density having the lowest palmitic acid ratio and the highest eicosanoic acid ratio was 25x30 cm. In summer sowing, the highest difference in eicosanoic acid content was generated by the sowing density of 25x20 cm. In other fatty acids, the difference was statistically insignificant in all sowing densities in both summer and winter sowings; they produced approximate values. The ratios of stearic, palmitic and oleic fatty acids decreased while linoleic fatty acid ratio increased in summer sowing. In a sowing time study conducted on 3 different safflower varieties, late sowing dates resulted in higher oleic acid, palmitic acid and stearic acid ratios but a lower linoleic acid ratio (Samancı and Ozkaynak 2003). In Mediterranean Region, early sowing of high oleic type sunflower hybrids led to a decrease in oleic and palmitic acid contents but an increase in linoleic and linolenic acid ratios (Flagella et al. 2002).

Milk Thistle seeds contain a low number of polyunsaturated fatty acids but a high number of unsaturated fatty acids, making its oil ideal for human nutrition. Besides, the plant contains slymarin, a substance used for the treatment of serious liver diseases such as cirrhosis and cancer. So, if included in the agricultural products of Kazova region in Tokat, where there are no ecological or physiological restrictions, Milk Thistle may contribute to both regional and national economy.

REFERENCES

Acikgoz N. 1993. Tarımda araştırma ve deneme metotları. E.Ü. Ziraat Fakültesi yayınları, no:478. İzmir

Ahmadzadeh, S., Kadivar M., Saeidi, G., 2013. Investıgatıon of oil properties and seed composition some safflower lines and cultivars. Journal of Food Biochemistry 38 (2014) 527–532

Andrzejewska, J., Sadowska, K., Mielcarek, S., 2010. Effect Of Sowing Date And Rate On Yield And Flavonolignan Content Of The Fruits Of Milk Thistle (Silybum Marianum L. Gaertn.) Grown On Light Soil İn A Moderate Climate. Industrial Crops And Products.

Broun, P., Somervılle, C. 1997. Accumulation of ricinoleic, lesquerolic and densipolic acid an seeds to transgenic arabidopsis plants that Express a fatty acyl hdroxlase cDNA from castor bean. Plant Phsiology, 113: 933-942

Demirezer Ö. Ersoz, T. Saracoglu I. Sener, B. 2007. Tedavide kullanilan bitkiler, FED Monografları, Medikal & Nobel Yayınevi. 265-270

Demirtas, I., Ayhan, B., Sahin, A., Aksit, H., Elmastas, M., Telci, I. 2011. Antioxidant activity and chemical composition of Sideritis libanotica Labill. ssp. linearis (Bentham) Borm. (Lamiaceae). Nat Prod Res: 25:1512–1523

El-Mallah, H., El-Shami, S., Hassanein, M. M., 2003. Detailed studies on some lipids of *Silybum Marianum* (L.) seed oil, national research centre, fats and oils department, dokki, cairo, 397-402 Egypt

Flagella, Z., Rotunno, T., Tarantıno, E., Caterına, A., De Caro, A. 2002. Changes in seed yield and oil fatty acid composition of high oleic sunflower (*Helianthus annuus* L.) hybrids in relation to the sowing date and the water regime. Europan Journal of Agronomy, 17: 221-230

Gororo, N., Salısbury, P., Rebetzke, G., Wayne, B., Bell, C. 2003. Genotypic variation for saturated fatty acid content of Victorian canola. 11th International rapeseed congress, the royal veterinary and agricultural university 6-10 July, Copenhagen, Denmark

Gumuscu, A., Arslan, N. Gurbuz, B. 1998. Farklı ekim zamanlarının meryemana dikeni (*S. marianum*)'nin verim ve bazı özelliklerine etkisi, Prooceedings of XII th international symposium on plant originated crude drugs, Ankara, 106

Gürbüz, B., Gümüşçü, A., Arslan, N. 1998. Farklı Bitki Sıklıklarının Meryemana Dikeni (Silybum Marianum)'nin Verimine Etkisi, Prooceedings Of XII th International Symposium On Plant Originated Crude Drugs, Ankara. 106.

Hasanloo, T., Sepehrifar, R., Bahmany, M., Kalantary. F. 2008. Tocopherols, antiradical activity and fatty acids in seeds of *Silybum Marianum* (L.) gaertn, biomed Proceedings 21, pp. 362–365

Katar, D., Arslan, Y., Subasi. I. 2012. Ankara ekolojik koşullarinda farkli ekim zamanlarinin ketencik (*Camelina Sativa* (L.) Crantz) bitkisinin yağ orani ve bileşimi üzerine olan etkisinin belirlenmesi. Tekirdağ Ziraat Fakültesi Dergisi. Cilt 9, Sayı 3, Sayfa:84-90. Tekirdağ

Keles, R., Ozturk, Ö.2012. Farklı ekim zamanlarinin bazi aspir çeşitlerinde verim ve kalite üzerine etkileri. Tarım bilimleri araştirma dergisi. 5 (1) s:112-117. Konya

Mericli, A. H. 1984. Türkiye'nin değişik bilgelerinde yetişen *Silybum marianum* türlerinin meyvelerinin flavonolignan bileşikleri yönünden incelenmesi. Doğa bilim der. 8(2). 203

Mılls, S. Y., Bone, K. 2000. Principles and practice of phytotherapy: Modern herbal medicine. Churchill livingstone, London

Robbelen, G., Downey, R. K., Ashrı, A. 1989. Oilcrops of the world. Mc Graw Hill, USA

Samancı, B. and E. Ozkaynak. 2003. Effect of planting date on seed yield, oil content and fatty acid composition of safflower (*Carthamus tinctorius L.*) cultivars grown in the mediterranen region of Turkey. J. Agronomy& Crop Science, (189): 359-360

Sanchez-Sampedro, M. A., Pelaez, R., Corchete, P. 2008. An arabinogalactan protein isolated from medium of cell suspensions cultures of *Silybum marianum* Gaernt. Carbohydrate polymers, 71:634-638

Stryer, L. 1986 Biochemistry. 30 th press. W. H. Freeman Comp. Inc., New York**.**

Tanker, M., Tanker, N. 2003. Farmakognozi. Cilt I. Ankara Üniv. Ecz. Fak. Yayınları No:58, Ankara. 213-214

Tanker, N., Koyuncu, M., Coskun, M. 2007. Farmasötik Botanik Ders Kitabı, Ankara Üniv. Ecz. Fak. Yayınları. 324

Wagner, H., Horhammer, L. Munster, R. 1986. The Chemistry of silymarin (Silybin), The active principle of the fruits of *Silybum Marianum* (L.) Gaertn. *Arzneim-Forsch Drug Resp* 18:688–96

Yilmaz, G., Tasgin S., Kınay, A. 2013. Yazlık ve kışlık yetiştirilen meryemana dikeni (*Silybum marianum*) bitkisinde farkli ekim sıklıklarının verim ve verim özelliklerine etkileri, Türkiye X. Tarla Bitkileri Kongresi Bildirileri, Cilt: 3, Sayfa: 1106-1111, 12-15 Eylül, Konya

Weıss, E. A. 1983. Oilseed crops. Tropical agriculture series., Pub. By Longman Inc., Leonord Hill Boks, NewYork

Pritchard, F. M., Eagles, H. A., Norton, R. M., Salisbury, P. A., Nicolas, M. 2000. Environmental effects on seed composition of Victorian canola. Aust J Exp Agric 40:679–685

Table 1. Effects of different sowing density and time on plant height, 1000 seeds weight and seed yield

Sowing density	Plant height (cm)				1000 seeds weight (g)				Seed yield (kg/da)			
	2010-2011		2011-2012		2010-2011		2011-2012		2010-2011		2011-2012	
	Summer*	Winter*	Summer*	Winter	Summer	Winter*	Summer*	Winter*	Summer*	Winter*	Summer*	Winter*
25x10	71,1^{ab}	144,0^{a}	45,8^{c}	131,1	28,0	26,0^{b}	27,9^{ab}	28,8	189,7^{ab}	567,3^{b}	56,9^{ab}	257,8^{a}
25X20	71,5 ^{ab}	161,1^{a}	46,2^{c}	131,3	29,3	26,2^{ab}	27,4^{ab}	28,5	216,4^{a}	672,7^{a}	44,8^{b}	252,4^{a}
25x30	60,0^{c}	153,6^{a}	55,3^{abc}	130,9	27,4	27,5^{ab}	27,4^{ab}	28,1	160,8^{bc}	536,0^{bc}	69,2^{a}	186,8^{abc}
25x40	62,0^{bc}	154,3^{a}	53,8^{abc}	124,1	28,6	25,8^{b}	28,1^{ab}	28,7	191,2^{ab}	580,0^{b}	46,6^{b}	194,7^{abc}
50X10	74,4^{a}	152,6^{a}	52,0^{bc}	120,4	28,7	28,0^{a}	29,1^{a}	27,8	158,5^{bc}	454,1^{cd}	49,6^{b}	157,9^{bc}
50X20	69,6^{abc}	146,5^{a}	51,0^{bc}	134,2	29,1	26,0^{b}	27,4^{ab}	26,1	151,8^{bc}	420,9^{d}	46,3^{b}	230,0^{ab}
50X30	64,9^{abc}	142,3^{a}	61,8^{ab}	123,5	27,8	25,8^{b}	27,2^{b}	27,8	121,3^{c}	291,4^{e}	46,7^{b}	117,0^{c}
50X40	61,6^{bc}	121,7^{b}	64,7^{a}	125,6	28,5	27,4^{ab}	28,1^{ab}	27,6	130,1^{c}	233,6^{e}	37,7^{b}	133,2^{c}
Mean	66,9	147,0	53,8	127,6	28,4	26,6	27,8	27,9	165	469,5	49,7	191,2

*<0,05

Table 2. Effects of different sowing density and time on oil ratio and oil yield

Sowing density	Oil ratio (%)				Oil yield (kg/da)			
	2010-2011		2011-2012		2010-2011		2011-2012	
	Summer*	Winter	Summer*	Winter	Summer*	Winter*	Summer*	Winter*
25x10	25,5^{a}	23,0	23,0^{ab}	22,1	48,7^{ab}	130,2^{b}	13,1^{ab}	57,0^{a}
25X20	23,4^{ab}	24,2	23,3^{a}	21,0	50,7^{a}	162,8^{a}	10,5^{bc}	52,9^{ab}
25x30	23,4^{ab}	23,1	23,4^{a}	21,3	37,5^{bcd}	124,2^{b}	16,2^{a}	38,6^{b-e}
25x40	24,1^{ab}	23,4	22,5^{ab}	22,9	46,6^{ab}	135,5^{b}	10,5^{bc}	44,6^{a-d}
50X10	25,0^{a}	23,9	23,7^{a}	21,4	39,6^{abc}	108,6^{bc}	11,8^{abc}	33,8^{cde}
50X20	24,0^{ab}	22,0	23,8^{a}	21,2	36,8^{bcd}	92,7^{cd}	11,1^{bc}	48,5^{abc}
50X30	22,4^{b}	22,7	22,1^{ab}	20,4	27,1^{d}	66,7^{de}	10,2^{bc}	23,7^{e}
50X40	24,0^{ab}	23,3	20,9^{b}	21,2	31,2^{cd}	54,4^{e}	7,9^{c}	28,3^{de}
Mean	24,0	23,2	22,9	21,4	39,8	109,4	11,4	40,9

*<0,05

Table 3. Effects of different row and row spacing on fatty acids ratio of winter sowing in milk thistle

Row spacing (cm)	Intra-row spacing (cm)	Palmitic asid	Stearic asid	Oleic asid	Linoleic asid	Arachidic asid	Eicosanoic asid	Cis-8,11,14-eicosatrienoic asid	Eicosapentanoic asid
25	10	8.86 a	6.71	31.26	43.97	4.24	0.78 a	2.98	0.76
	20	8.89 a	6.58	31.56	43.45	4.25	0.94 bc	2.97	0.76
	30	8.73 a	6.62	31.38	43.97	4.23	0.98 c	2.96	0.76
	40	9.02 ab	6.75	31.37	43.42	4.30	0.96 bc	3.02	0.77
	Mean	8.88	6.67	31.39	43.70	4.26	0.92	2.98	0.76
50	10	9.36 b	6.99	31.25	43.14	4.37	0.88 b	2.91	0.72
	20	8.74 a	6.65	31.52	44.12	4.19	0.91 bc	2.83	0.72
	30	8.83 a	6.69	31.31	43.83	4.27	0.94 bc	3.00	0.78
	40	8.99 ab	6.73	30.85	44.30	4.13	0.95 bc	2.95	0.75
	Mean	8.98	6.77	31.23	43.85	4.24	0.92	2.92	0.74
Duncan ^{a,b,c}		0.43 *	ns	ns	ns	ns	0.61 ***	ns	ns

ns= no significant, *=p<0.05, ***=p<0.001, In each column, the same letter was not significantly different

Table 4. Effects of different row and row spacing on fatty acids ratio of summer sowing in milk thistle

Row spacing (cm)	Intra-row spacing (cm)	Palmitic asid	Stearic asid	Oleic asid	Linoleic asid	Arachidic asid	Eicosanoic asid	Cis-8,11,14-eicocsatrienoic asid	Eicosapentanoic asid
25	10	7.49	6.44	28.11	48.34	4.01	0.95 bc	2.83	0.75
	20	8.63	6.47	29.03	46.70	4.10	0.98 c	2.93	0.76
	30	8.29	6.69	29.12	47.00	4.16	0.96 bc	2.99	0.79
	40	8.63	6.71	28.84	46.67	4.16	0.91 ab	2.98	0.80
	Mean	8.26	6.58	28.78	47.18	4.11	0.95	2.93	0.78
50	10	8.96	6.75	28.82	46.39	4.13	0.89 a	2.94	0.76
	20	8.67	6.78	29.08	46.20	4.18	0.94 abc	2.98	0.77
	30	8.63	6.70	28.14	47.40	4.11	0.95 bc	2.89	0.81
	40	8.51	6.87	29.10	46.25	4.25	0.95 abc	2.96	0.77
	Mean	8.69	6.78	28.77	46.56	4.17	0.93	2.94	0.78
Duncan [a,b,c]		ns	ns	ns	ns	ns	0.42*	ns	ns

ns= no significant, *=p<0.05, In each column, the same letter was not significantly different

Table 5. Effects of different sowing season on fatty acids

Sowing season	Palmitic asid	Stearic asid	Oleic asid	Linoleic asid	Arachidic asid	Eicasonoic asid	Cis-8,11,14-eicocsatrienoic asid	Eicosapentanoic asid
Winter	8.93 a	6.72	31.31 a	43.77 b	4.25 a	0.92	2.95	0.75 b
Summer	8.47 b	6.68	28.78 b	46.87 a	4.14 b	0.94	2.94	0.78 a
Mean	8.70	6.70	30.05	45.32	6.32	0.93	2.95	0.77
Duncan [a,b,c]	0.14**	ns	0.74 ***	0.70 ***	0.15 ***	ns	ns	0.06 *

ns= no significant, *=p<0.05, **=p<0.01, ***=p<0.001
In each column, the same letter was not significantly different