# Effect of time of sowing on reproductive development of variegated thistle

R.J. Martin, B. Deo and J.A. Douglas[1]

New Zealand Institute for Crop & Food Research Limited, Private Bag 4704, Christchurch
[1]New Zealand Institute for Crop & Food Research Limited, Private Bag 3123, Hamilton, New Zealand

## Abstract

Variegated thistle (*Silybum marianum* (L.) Gaertn.) is grown overseas for its seed that contains silymarin, a liver detoxicant. To formulate sowing date recommendations for variegated thistle in New Zealand, seed was sown at monthly intervals over 12 months in two trials in Canterbury. Sowings made from April to September, when soil temperatures were below 13°C, had very poor emergence, whereas summer sowings had high emergence. Plants from late November through May sowings did not produce visible buds until the following spring, but then went through their reproductive stages rapidly. Winter sown plants were slow to emerge, but then went through their reproductive stages rapidly. Some spring sowings matured in the autumn, but others had poor emergence and/or flowered in the winter. Duration from first emergence to first visible bud was not simply related to thermal or photothermal time, but appeared to be related to thermal time, modified by daylength. Duration from first emergence to first bud required around 2000 degree-days over 5°C, up to around 13 hours daylength at first bud, but decreased as daylength at first bud increased beyond this. Recommendations are that the naturalized New Zealand variegated thistle be grown commercially as a biennial, sown in late summer. Sowing in spring as an annual crop is risky because of poor emergence with low spring soil temperatures, and also because of the risk of the crop maturing late in the autumn when it is likely to be difficult to harvest.

***Additional key words:*** *milk thistle,* Silybum marianum, *emergence, soil temperature, thermal time, photoperiod*

## Introduction

Variegated thistle (*Silybum marianum* (L.) Gaertn., also sometimes classified as *Carduus marianus* (L.), is an annual or biennial herb native to southern Europe, where it is known as milk thistle. It was introduced from Europe last century and is now a widespread weed throughout northern and eastern New Zealand (Dingwall, 1950). On dry hill country it is a vigorous colonizer of bare ground, such as cut tracks or stock camps, and can form dense thickets. It can cause poisoning of stock, due to high nitrate levels (Connor, 1977), and is classified as a noxious weed in Canterbury and other parts of New Zealand.

It is also an important weed of crops and pastures in many areas of South America, South Africa, Australia, the United States, and the Middle East (Holm *et al.*, 1997). However, it is easily controlled by herbicides and does not compete well with vigorously growing crops and pastures (Glue and Matthews, 1957).

Variegated thistle is a very prickly plant. It initially forms a rosette of green leaves up to 45 cm diameter. The leaves have distinctive white veins, which, according to a Christian legend, were formed when the milk of the Virgin Mary fell on them, hence the alternative common names of Milk Thistle, Holy Thistle, Blessed Thistle, Lady's Thistle, and St Mary's Thistle (Foster, 1991). When the plant goes reproductive, a branching flowering shoot up to 2 m in height is produced, with green flowering buds that open to purple flowers up to 7 cm in diameter. Flowering is spread over a number of weeks (Dodd, 1989). After flowering, the seed develops inside the spiny seed head, which is lined with hairs. The seed is the size of a rice grain (~25 mg) and appears to either fall to the ground or is spread after ingestion by birds. It is not spread by wind (Glue and Matthews, 1957). The earlier formed seed heads produce the most seed (Dodd, 1989).

The seeds contains 1.5-3% silymarin, a mixture of flavonolignans including silybinin, silychristin and silydiadin (Bissett, 1994). Silymarin is used in the treatment of most forms of liver disease, including cirrhosis, hepatitis, necroses and liver damage due to alcohol and drug abuse (Morazzoni and Bombardelli, 1995; Flora *et al.*, 1998). It has a wide range of activity in the liver (Bissett, 1994; Morazzoni and Bombardelli, 1995; Flora *et al.*, 1998), which can be grouped into three modes of action (Castleman, 1995). Firstly, it alters the structure of the outer cell membrane of liver cells, which blocks the absorption of toxins into the cells. Secondly, it acts as an antioxidant inside liver cells, reducing the damage induced by toxins. Thirdly, it stimulates the formation of new liver cells by increasing the rate of synthesis of cellular proteins.

Silymarin is used widely in Germany (sales of US$18M in 1995), which, in 1988, imported 1800 t of thistle seed from cultivated crops in Argentina, China, Romania and Hungary (Anon., 1995). Cultivars are being bred in northern Europe for high levels of silymarin (Kazmierczak and Seidler-Lozykowska, 1997). The crop there is sown in March and harvested early September, yielding around 1 t seed/ha (Hecht *et al.*, 1992; Schunke, 1992).

In the wild in New Zealand, variegated thistle acts like an annual or biennial. On Banks Peninsula, seeds germinate in February-March, and the seedlings grow into rosettes up to 45 cm diameter, and remain in this state through the winter. Regrowth is rapid in the spring and flower stalks appear in October. Flowering begins in November and continues through December, with the seed ripening in January and February. The plants die when the seed has matured.

As part of a programme to develop variegated thistle seed production as a new crop in New Zealand, this trial was designed to determine the effect of time of sowing on reproductive development of naturalized variegated thistle, in order to formulate sowing date recommendations for growers.

## Materials and Methods

Two trials were carried out on Crop & Food Research's Lincoln farm on a Templeton silt loam soil. In the first trial, variegated thistle seeds gathered from wild populations on Banks Peninsula, and with a laboratory germination of 85%, were planted out at monthly intervals from October 1994 to September 1995 in 3 m by 2.5 m plots. Each sowing date was replicated three times in a randomized block experiment. In the second trial, seed saved from the first trial, with a laboratory germination of 80%, was planted out at monthly intervals from October 1997 to October 1998, excluding June. Each sowing was in 4 m by 4 m plots replicated four times in a randomized block experiment. Initial seedbed preparation was by ploughing and rota crumbling, with final cultivation by hand hoeing and raking.

Before sowing, the seeds were soaked overnight in 200 ppm gibberellic acid solution, which increased the laboratory germination by up to 15%. In the first trial, the treated seeds were sown by hand about 2 cm deep and spaced 5 cm apart in five rows 50 cm apart. In the second trial, the seeds were sown shallower, about 1 cm deep and spaced 25 cm apart in eight rows 50 cm apart.

Measurements were made weekly on the trials. Emergence counts were made from first emergence until there was no change in plant numbers. The counts were made on the middle three rows in each plot in the first trial and the middle six rows in the second trial. The date of first and last emergence was recorded along with the date when 50% of plants had emerged. The dates when the first visible bud, first visible flower and first matured head (when the dry seed head opened) were also recorded on six plants in each plot.

The plots were hand-weeded and irrigated to avoid stress to the plants, but no fertilizer was applied to the thistles.

Soil and air temperatures were measured at the Lincoln Meteorological Station, 300 m from the experimental sites. Daily thermal time was calculated as the higher of zero or the mean air temperature minus a base temperature of either 0 or 5°C.

Photothermal time was calculated by multiplying temperature above a base of 0°C by effective photoperiod, where effective photoperiod = (Actual photoperiod (including twilight) - base photoperiod)/(Saturated photoperiod - base photoperiod), where saturated photoperiod = 20 hours, and base photoperiod = 10 hours.

## Results

In both trials there was a wide range of final emergence, which was below 13% for sowings between April and September, and zero for May and June in the first trial. For the November to February sowings, emergence averaged over 90% in the first trial and over 55% in the second trial; the reason for the difference between the two trials is not known. Emergence was dependent on soil temperature with little emergence when soil temperature at 10 cm was below 11-13°C (Fig. 1). We did not check whether any seed remained dormant until the next season, as occurs with some thistle species (Roberts and Chancellor, 1979).

Time to first emergence varied considerably from 7 to 17 days for sowings from mid October to mid March, to 50-90 days for sowings from May to August (Fig. 2). During the summer months, seedlings in the shallower-sown second trial emerged an average of 6 days earlier than the first trial.

There was wide variation in the time from first emergence to first visible bud, ranging from 63 to 67 days for July and August sowings up to 229-279 days for sowings from mid November to mid March (Fig. 2). For 18 of the 22 sowings that established, this condensed the 12-month range of sowing to a five-month range (October - February) over which the first bud became visible. The exceptions were the sowings in late October and early November where the first bud became visible in April or early May, and the sowings in late November and early December where the first bud became visible in late August - early September.

The time from first bud to first visible flower ranged from 5 to 27 days, but with no obvious trend with sowing date (Fig. 2). The time from first flower to first mature head generally ranged from 10 to 38 days. The exceptions were the 28 November 1997 and 9 December 1994 sowings (~58 days), where the plants flowered in late winter, and the 6 November 1994 sowing (168 days), where the plants flowered in May and the seed heads did not mature until October. Apart from these exceptions, there was no obvious trend with sowing date.

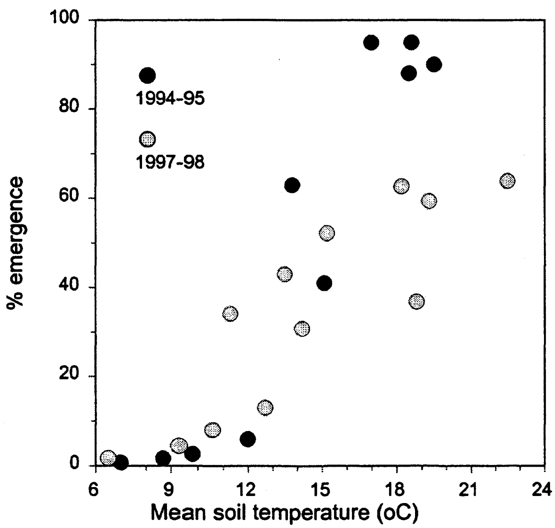


**Figure 1. Percentage variegated thistle seedling emergence versus mean soil temperature (10 cm depth) from sowing to the end of emergence. Closed symbols 1994-5, open symbols 1997-98.**

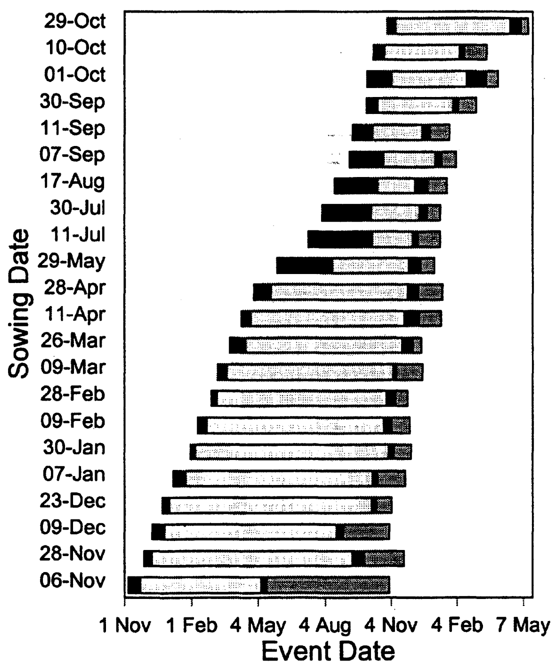


**Figure 2. Dates of sowing, first emergence, first bud, first flower and first mature head for all established sowings of variegated thistle. Shading on bars are, from left to right, sowing to first emergence, first emergence to first bid, first bud to first flower and first flower to first mature head.**

There was no consistent relationship between sowing date and thermal time (base temperature 0°C) from first emergence to first bud. However, with the exception of the November sowings, thermal time (base 0°C) from first visible bud to first mature head averaged 535 (s.e.m. 33.6), without any trend across sowing dates. Increasing the base temperature to 5 or 10°C, or using photothermal time, did not improve on these relationships (data not presented).

However, when thermal time from first emergence to first visible bud was plotted against daylength at first visible bud, a two-stage relationship was obtained (Fig. 3). A base temperature of 5°C gave a better relationship than 0°C. Below 13.3 hours, thermal time (base 5°C) from first emergence to first visible bud averaged 1900 degree-days. Above 13.3 hours, there was a linear relationship between thermal time and daylength (in hours)

$$\text{Thermal time} = 7560\ (\pm 190) - 424\ (\pm 50.9) \times \text{daylength}$$
$$(r^2 = 0.812,\ \text{d.f.} = 16)$$

This relationship indicates that for every hour that daylength increased above 13.3 hours, the thermal time requirement decreased by 424 degree-days (base 5°C).

The few surviving plants from the winter sowings in the first trial were very short (less than 200 mm high). In the second trial, they were taller (around 500 mm). In the summer sowings, plants were up to 1.6 m tall. The number of flowers increased with plant size. There were some plant deaths in the trial, typically starting with dead patches on leaves spreading to other leaves and plants. Weak saprophytic fungi (*Stemphylium botryosum*, an *Alternaria* sp. and a *Cladosporium* sp.) and bacteria (*Pseudomonas* spp.), which are known to cause lesions and blight in other plants, were isolated from diseased areas of the plants, but it is not known if these caused the mortality.

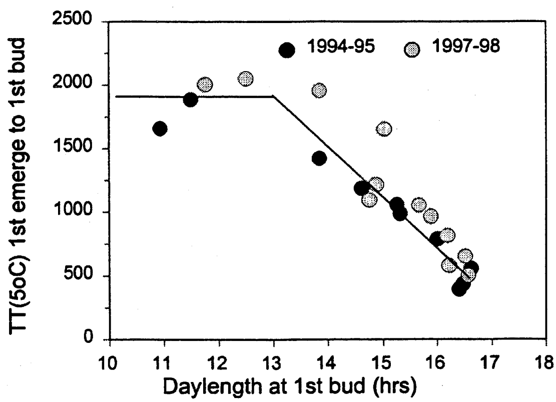


**Figure 3. Thermal time (base 5°C) from first emergence of variegated thistle to first bud versus daylength when first bud visible. Closed symbols 1994-5, open symbols 1997-98.**

## Discussion

This experiment has shown that New Zealand naturalized variegated thistle will grow as an annual or as a biennial at Lincoln. Plants from sowings from May through October matured in 4.5-6.5 months. However, they took a long time (one to three months) to emerge, and, except for the late September 1997 and the October sowings in both trials, had very poor emergence. This led to major weed infestation problems in our plots. The late September and October sowings had better, but still not good emergence, and did not produce the first mature heads until March to May, even in the 1998-99 summer, the warmest on record. With the indeterminant nature of the crop, a harvestable yield would not be available until very late in the season, and harvesting such a bulky crop in damper autumn conditions could be a problem. While spring sowing of variegated thistle is therefore a possibility, sowing too early could lead to reduced and slow emergence, and sowing too late runs the risk of the crop maturing very late in the autumn or else not until the following spring. In addition, the plants from these sowings were smaller than plants from summer and autumn sowings and were also observed to have fewer seed heads.

In the other sowings the plants acted as biennials, and did not produce mature heads until after they had experienced a winter. This took almost a year for November sown crops, decreasing to around nine months for autumn sowings. This is similar to the wild habit where seedlings establish in the early autumn and the plants produce seed around Christmas. Summer sowings (from November through February) had the highest and most rapid emergence, and produced the largest plants, with the most seed heads. Summer sowing would be the preferred option for seed from the New Zealand naturalised variegated thistle, with harvesting in November or December.

In an associated trial (Martin and Deo, unpublished data), a block of variegated thistle was machine drilled in March 1993, and quadrats harvested when mature. Total dry matter yields ranged from 3020 to 15 600 kg/ha, but seed yield was only 7-98 kg/ha. This gave a harvest index of 0.0005 to 0.01, much lower than the 0.5 of temperate cereals in New Zealand. Research will need to be directed at increasing this harvest index. Also, the ripe seeds are very attractive to birds, and early harvesting before the seed heads open may be necessary to prevent seed losses to birds.

Promising approaches currently under study are the use of European cultivars, which have a more annual habit, the use of plant growth regulators to reduce plant size, and nitrogen fertilizer to increase seed yield. In preliminary trials (Martin and Deo, unpublished data), a German cultivar sown in mid October matured nearly two months earlier than the New Zealand line. Spraying with a plant growth regulator at bud formation reduced crop height by 17%, whereas adding 100 kg N/ha increased seed yield by 270%.

There were poor relationships between the length of the vegetative phase and either thermal or photothermal time. However, daylength and thermal time appear to interact as triggers for reproductive development. Other factors may also be involved, such as vernalization, but more detailed measurements would be required to separate out all the factors. However, the October sowing showed no sign of a vernalization requirement.

The period from first bud to first mature head was generally related to thermal time. The exceptions occurred when the seeds were maturing during winter when other factors such as humidity and frost may have slowed seed development.

These results are relatively coarse for two reasons. Firstly because measurements were only taken once a week, and secondly because finding a bud in a very dense prickly patch of thistles is not easy.

## Acknowledgements

This work is part of the Crop & Food Research New Crops Programme, which is funded by the Foundation for Research, Science and Technology.

## References

Anon. 1995. Market Report. *Herbalgram* ***39***, 68.

Bissett, N.G. 1994. Herbal Drugs and Phytopharmaceuticals. CRC Press, London. 566 pp.

Castleman, M. 1995. Milk thistle. Nature's liver protector. *Herb Quarterly* ***66***, 18-21.

Connor, H.E. 1977. The Poisonous Plants in New Zealand. New Zealand Department of Scientific and Industrial Research Bulletin 99. 247pp.

Dingwall, A.R. 1950. Variegated thistle and its control. *New Zealand Journal of Agriculture* ***81***, 141-142.

Dodd, J. 1989. Phenology and seed production of variegated thistle, *Silybum marianum* (L.) Gaertn., in Australia in relation to mechanical and biological control. *Weed Research* ***29***, 255-263.

Flora, K., Hahn, M., Rosen, H. and Benner, K. 1998. Milk thistle (*Silybum marianum*) for the therapy of liver disease. *American Journal of Gastroenterology* ***93(2)***,139-143.

Foster, S. 1991. Milk thistle *Silybum marianum*. *American Botanical Council Botanical Series* No. 305. 7 pp.

Glue, D.I. and Matthews, L.J. 1957. Weed identification variegated thistle. *New Zealand Journal of Agriculture* ***95***, 547.

Hecht, H., Mohr, T. and Lembrecht, S. 1992. Harvesting medicinal plants by combine. *Landtechnik* ***47 (10)***, 494-496.

Holm, L.G., Doll, J., Holm, E., Pancho, J. and Herberger, J. 1997. World weeds. Natural Histories and Distribution. Wiley, New York. 1129 pp.

Kazmierczak, K. and Seidler-Lozykowska, K. 1997. Silma - the Polish variety of milk thistle (*Silybum marianum* (L.) Gaertn.). *Herba-Polonica* ***43(3)***, 195-198.

Morazzoni, P. and Bombardelli E. 1995. *Silybum marianum* (*Carduus marianus*). *Fitoterapia* ***66(1)***, 3-42.

Polak, P. and Sustrova, H. 1991. A contribution to the cultural practice of field culture of holy thistle (*Silybum marianum* L. Geartn.). *Zahradnictvi* ***18(4)***, 305-314.

Roberts, H.A. and Chancellor, R.J. 1979. Periodicity of seedling emergence and achene survival in some species of *Carduus*, *Circium* and *Onopordum*. *Journal of Applied Ecology* ***16***, 641-647.

Schunke, U. 1992. Holy thistle. First experiences with cultivation and harvest. *Landtechnik* ***47(11)***, 548-550.