



# Influence of seeding date and seeding rate on cow cockle, a new medicinal and industrial crop



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## ABSTRACT

There has been keen interest in the development of cow cockle (*Vaccaria hispanica* (Mill.) Rauschert) as a medicinal and industrial crop in North America. However, very limited agronomic information exists regarding the cultivation of cow cockle as a crop. The objective of this study was to determine how seeding date and rate affected stand establishment, seed yield, and seed weight of cow cockle at various sites across western Canada. Field experiments were conducted at three locations across western Canada from 2006 to 2010 to evaluate the effects of seeding date (early May, mid May, early June, mid June) and seeding rate (50, 100, 200, 400, 800, and 1600 seeds m<sup>-2</sup>) on cow cockle stand establishment, seed yield, and thousand seed weight (TSW). Delayed seeding had an inconsistent effect on plant stand establishment, but resulted in significant declines in seed yield and TSW in nearly all site-years. Plant stand establishment increased linearly with increasing seeding rate. A significant curvilinear increase in seed yield occurred with increasing seeding rate, while TSW declined significantly at increased seeding rates. The results of this study clearly show that seeding at an optimum rate of 400 seeds m<sup>-2</sup> as early as practical in the growing season will maximize seed yield and TSW in cow cockle crops.

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## 1. Introduction

Recent interest in growing crops for industrial and medicinal purposes has been growing on a global scale. These crops have a multitude of uses, including their utility as flavorings, cosmetics, and for medicinal purposes (Ferrie, 2007). Research conducted over the past few years has demonstrated that the seed of cow cockle (*Vaccaria hispanica* (Mill.) Rauschert) is rich in flavonoids, cyclopeptides, and contains significant amounts of mono- and bisdesmosidic triterpene saponins (Balsevich et al., 2006, 2012). Saponins are important nutraceuticals with numerous bioactivities (Sharma et al., 2012). They are also important constituents of several traditional medicines and possess valuable pharmacological properties that may have utility in the pharmaceutical industry (Augustin et al., 2011). In addition, seed extracts from *V. hispanica* were found to possess potential anti-cancer activity (Campbell et al., 2002; Shoemaker et al., 2005; Ma et al., 2008). Due to these unique properties, there has been growing interest in the potential cultivation of cow cockle as a medicinal and industrial crop in North America. Saponin Inc., based out of Saskatoon, SK, Canada, had been developing cow cockle into an alternative crop for western Canada under the trade name Prairie Carnation® (Willenborg and Dosedall, 2011), but the company fell into receivership in

2011. However, Incubate Canada has recently formed and is pursuing the development of cow cockle as an industrial crop for Canada (M. Oelck, Incubate Canada, personal communication, 2012).

*V. hispanica* is a member of the pink family (Caryophyllaceae) and is an herbaceous annual weed that can be found in cropped fields, roadsides, and waste areas in the Northern Great Plains region of North America (Royer and Dickinson, 1999). Although it is considered a weed, its abundance and distribution from the 1970s to the 2000s has declined according to weed surveys conducted on the Canadian Prairies (Thomas and Leeson, 2007). Stems are erect, smooth, and branched with swollen nodes. Plant height ranges from 30 to 90 cm (Biliaderis et al., 1993) with sessile leaves 2–8 cm in length. Cow cockle has round, black seeds approximately 2.0 to 2.5 mm in diameter, similar to canola (*Brassica napus* L.) in size, shape, and color. Small plot (2 m × 6 m) yields range between 1300 kg ha<sup>-1</sup> and 6500 kg ha<sup>-1</sup> and vary significantly among genotypes (Willenborg and Dosedall, 2011). Seed weight also varies significantly among genotypes, ranging from 3.7 mg in small-seeded genotypes to 8.8 mg in large-seeded genotypes. Whole mature seeds of cow cockle contain approximately 60% starch, 15% protein, and 2–3% lipids, and are rich in saponins (Goering et al., 1966; Mazza et al., 1992). As a nutraceutical, many saponins contain anti-cholesteremic, anti-fungal, and immunostimulant properties (Kernan and Ferrie, 2006). Industrially, saponins can be used as emulsifiers, stabilizers, foaming agents, and shampoo additives. In addition, *V. hispanica* seeds

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**Table 1**  
Soil classification and descriptions for each site-year.

Site-year	Soil classification	Soil descriptions				
		pH	OM <sup>a</sup> (%)	Sand (%)	Silt (%)	Clay (%)
Carman 2006	Black Chernozem	5.9	7.5	36	24	30
Scott 2006	Dark brown Chernozem	6.0	3.5	38	40	22
Scott 2007	Dark brown Chernozem	6.0	3.5	38	40	22
Scott 2009	Dark brown Chernozem	6.0	3.5	38	40	21
Edmonton 2010	Black Chernozem	5.7	8.4	34	40	27

<sup>a</sup> Organic matter.

contain a high content of a unique, small grained starch that is approximately 0.3–1.5  $\mu\text{m}$  in size and may have potential use in the cosmetic industry (Mazza et al., 1992; Biliaderis et al., 1993).

Initial studies have found cow cockle to have attractive agronomic characteristics such as medium stature, early maturity, and ease of seed threshing (Mazza et al., 1992). Nevertheless, *V. hispanica* is susceptible to the disease *Alternaria saponariae* (Koike and Henderson, 1999), and yield can also be limited by the redbacked cutworm (*Euxoa ochrogaster* Guenée) (Willenborg and Dossdall, 2011). Although it is grown for its commercial value as an ornamental flower in California and other parts of the world (Koike and Henderson, 1999), cow cockle is likely to have potential benefits when grown in rotation with other small grain cereal, oilseed, and pulse crops on the Northern Great Plains. However, very limited agronomic information exists regarding the cultivation of cow cockle as a crop (Efthimiadou et al., 2012).

Two of the most important management factors determining crop productivity are optimal seeding date and seeding rate. Increasing seeding rates in many instances increases profitability, yield, and competitiveness of the crop (Mohler, 2001). Likewise, early planting of crops on the Northern Great Plains often leads to positive outcomes including higher yields and improved crop quality (McKenzie et al., 2005; De Bruin and Pedersen, 2008; O'Donovan et al., 2012). Specific knowledge of seeding date responses at various locations is necessary to determine whether early seeding should be a general recommendation (De Bruin and Pedersen, 2008). Interactions between crop types, genotypes, poor early season growth conditions, and soilborne pathogens conceivably could reduce any benefits associated with early seeding (Grau et al., 1994). There is currently no recommended optimal seeding date or rate for cow cockle. Thus far, cow cockle has been seeded in early to mid-May at a rate of 5.5–8  $\text{kg ha}^{-1}$  or to achieve a target plant population density of 200 plants  $\text{m}^{-2}$  (Balsevich, 2008; Willenborg and Dossdall, 2011). The objective of this study was to determine how seeding date and rate affected stand establishment, seed yield, and seed weight of cow cockle at various sites across western Canada.

## 2. Materials and methods

Field experiments were conducted at three locations across western Canada from 2006 to 2010. The locations were Scott, SK (52.36° N, 108.83° W), Carman, MB (49.68° N, 97.11° W), and Edmonton, AB (53.25° N, 113.33° W), Canada. Soil classifications and descriptions are presented in Table 1. The study was conducted at Scott, SK in 2006, 2007, and 2009, at Carman, MB in 2006, and at Edmonton, AB in 2010. All plots were established on conventionally tilled fields that had been fallowed the year prior.

Factorial combinations of cow cockle density and seeding date treatments were arranged in a randomized complete block design with four replications. Seeding dates consisted of planting in early and mid May as well as in early and mid June. Actual planting dates and accumulated thermal time in growing degree-days (base temperature 0°C) can be found in Table 2. Seeding rate treatments

were initially comprised of 50, 100, 200, 400 seeds  $\text{m}^{-2}$  in 2006; thereafter, seeding rate treatments of 800 and 1600 seeds  $\text{m}^{-2}$  were added to the experiment in all subsequent site-years. Plot size was 2 m  $\times$  6 m in Edmonton and Carman and 2 m  $\times$  5 m at Scott. A semi-domesticated genotype of cow cockle ('Scott') was used as the seed source for the entire study.

Prior to the sowing of each seeding date treatment, a single pre-seeding application of glyphosate (900 g a.e.  $\text{ha}^{-1}$ ) was applied to control emerged weeds at all sites. At Scott in 2009 and Edmonton in 2010, isoxaflutole (160 g a.i.  $\text{ha}^{-1}$ ) was also applied pre-emergence to provide selective residual control of dicotyledonous weed species. Planting was carried out with minimum disturbance plot seeders equipped with side band (knife or hoe) openers at Scott and Edmonton, and disk openers at Carman. Fertilizer was side-banded during seeding at Scott and Edmonton, but was broadcast and immediately incorporated with a tillage pass at the Carman site. Fertilizer was applied at recommended rates based on soil tests. Row spacing on the seed drills varied from 15 to 22 cm, with seed placed at approximately 1–1.5 cm in depth. All sites received an in-crop application of clethodim (56 g a.i.  $\text{ha}^{-1}$ ) with a crop oil concentrate (0.5%, v/v) at the 2- to 4-leaf stage to control weedy graminoid species. Any weeds that were not controlled by the herbicide applications were removed manually.

Site and environmental data were recorded at each site. Weather data were recorded from weather stations available at each site. Soil organic matter content, pH, and texture were determined to a depth of 30 cm. Cow cockle stand density was determined by counting the number of emerged plants in two individual 1-m row lengths 3–4 wk after sowing. Cow cockle grain yield was harvested with a small plot combine from the central six rows of each 8-row plot. Samples were then dried to a constant moisture, cleaned, and the weight of the grain recorded. From this sample, the weight of 250 seeds was determined and multiplied by a factor of four to provide an estimate of thousand seed weight (TSW). Thousand seed weights were not taken at the Scott site in 2006.

Data were tested and transformed (stand establishment) as required to meet the assumptions of analysis of variance. Stand establishment data were Ln transformed and consequently, back-transformed means and standard errors are presented. Stand establishment, yield, and TSW were subjected to a two-way analysis of variance using the MIXED model procedure of SAS (SAS Institute Inc., 2004). Seeding rate and date were considered fixed effects in the model. Year by location combinations (site-year), block (within site-year), and the site-year interaction with treatments were considered random effects. Treatment effects were declared significant at  $P \leq 0.05$ . Although seeding rate data could be combined across site-years, seeding date data could not be combined across site-years due to significant treatment by site-year interactions and therefore, these analyses were conducted within site-years. Linear regression was used (PROC REG; SAS Institute Inc., 2004) to describe the relationship between seeding rates and stand establishment.

A non-linear regression model was fitted to cow cockle seed yield means using the NLIN procedure of SAS (SAS Institute Inc.,

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