Hand Emasculations and Induced Male Sterility to Improve Field Pennycress (Thlaspi arvense L.) Breeding

W. Hoffman, K. Roskamp, and W.B. Phippen

School of Agriculture, Western Illinois University, Macomb, IL 61545, USA

INTRODUCTION

Field Pennycress (Thlaspi arvense L.) is a winter annual oilseed crop that is currently being studied as a new source of industrial products and biodiesel. Conducting controlled crosses between small flowered pennycress varieties is difficult and time consuming. The objective of this study was to develop a reliable method of inducing pollen sterility in order to conduct controlled crosses in this self-pollinated crop.

A total of 146 controlled crosses between winter and spring lines were conducted during the morning (8-10am) and afternoon (3-5pm) hours resulting in 32.1% and 44.6% fertilization, respectively. The morning to winter line crosses resulted in 46% fertilization. For winter to spring crosses, only 30% were successful. A secondary experiment was initiated to investigate the role of sulfonurea herbicide, tribenuron-methyl, as a potential aid to induce male sterility in pennycress. Pennycress seedlings were grown in a controlled environment. When all plants reached reproductive stage, applications of 0.1, 0.2, 0.3, and 0.4 µg/ml tribenuron-methyl per plant were applied to the leaves and repeated 10 days later. An application consisted of a single 1 ml mist spray to each of 8 replicated plants in a treatment group. All applications of the herbicide resulted in severe stunting of the plants and a delay in flowering. Control plants flowered within 2 days, while the 0.1 µg/ml application flowered in 10 days. Plants treated with applications of 0.2, 0.3, and 0.4 µg/ml tribenuron-methyl had severe yellowing of young tissue and did not set seed after 20 days of treatment.

The development of a reliable induced male sterility protocol will greatly improve the efficiency of breeding and will help lead to the development of new varieties of pennycress.

MATERIALS AND METHODS

Hand-Emasculations: Field pennycress (Thlaspi arvense) breeding lines 'NY10’, ‘W.12’, ‘OD1’, and spring line ‘Spring 32’ were used throughout these experiments. To synchronize flowering of winter lines to spring lines, the winter varieties were emasculated prior to removing the winter lines from the cold. ’Spring 32’ seeds were germinated under warm conditions and transplanted to individual pots. Within 30 days, both winter and spring lines were initiating floral buds. Hand-emasculations were performed in the morning (8 to 10am) and in the afternoon (3 to 5pm) for 2 weeks (Figure 2). All open flowers and immature buds were removed from the apical meristem leaving only partial expanded floral buds (Figure 3). Sepals, petals, and anthers were removed from the female plant utilizing magnifiers and sterilized forceps. Pollen was placed on the exposed stigma by a small paint brush or from a donor flower. Plants were labeled and covered with a glycine bag (Figure 4).

Induced Male Sterility: Seeds from pennycress line ‘Spring 32’ were germinated as described above and transferred to individual pots and allowed to grow until the reproductive stage under warm (24°C) and long day (18hr) conditions. Applications of 0.1, 0.2, 0.3, and 0.4 µg/ml tribenuron-methyl per plant were applied to the leaves of 8 plants per treatment and repeated 10 days later on only 4 plants per treatment. An application consisted of a single 1 ml mist spray. Tribenuron-methyl is the active ingredient at 50% in Express® herbicide from Dupont.

RESULTS AND DISCUSSION

The objective of this study was to determine the success of conducting hand-emasculations between winter and spring lines of pennycress grown under controlled environmental conditions. Experiments on a similar oilseed crop, Camelina have been shown to be successful (Lessman, 1990). Conducting emasculations and subsequent pollinations appears to be highly dependent on time of day. Literature suggests early morning crosses tend to be much more successful than late afternoon (Sahbwarw and Doležel, 1993). A second objective was to investigate the role of sulfonylurea herbicide, tribenuron-methyl, as a potential aid to induce male sterility in pennycress. Previous research has suggested the application of sulfonylurea herbicides have been effective in creating male sterility in Brassica species and is now widely used in canola hybrid production (Yu et al. 2006). The development of a reliable induced male sterility protocol will greatly improve the efficiency of pennycress breeding and will help lead to the development of new varieties of pennycress.

LITERATURE CITED


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