Polyhaploid production in hexaploid wheat crosses with stored pearl millet pollen

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Introduction

For last two decades a technique to produce polyhaploids of hexaploid wheat (*Triticum aestivum* L.) has been developed by use of wide crosses followed by chromosome elimination. Significant technical developments were attributed to pollen selection from subfamililial species and the application of plant growth regulators (for a review, Inagaki and Mujeeb Kazi 1994a). However, this kind of method always requires viable pollen at the time of pollination and influences the polyhaploid production duration. Pollen storage technique may resolve this disadvantage. Maize (*Zea mays* L.) pollen dried to an optimum water content range and stored at -80 C for three months produced polyhaploid embryos on wheat, but the frequencies decreased to the half in case of fresh pollen (Inagaki and Mujeeb-Kazi 1994b). Pearm millet (*Penisetum glaucum* (L.) R. Br.) is an alternative pollen source not only for polyhaploid production of hexaploid wheat (Ahmad and Comeau 1990, Inagaki and Mujeeb-Kazi 1995) but also for long-term pollen storage (Hanna 1990). This study presents the embryo formation frequencies in crosses of hexaploid wheat with stored pearl millet pollen.

Materials and Methods

The hexaploid wheat variety Norin 61 and the pearl millet line NEC 7006 grown in greenhouses (max./min.; 25/15 and 35/10°C respectively), were used as female and pollen parents. Five grams of pearl millet pollen were collected between 9:30 - 10:00 a.m., and dried with gentile ventilation at 35°C and 35-40% R.H. The dried pollen was distributed among 15 polyethylene tubes (1.5 ml vol.). The sealed tubes were immersed in liquid nitrogen (-196°C) for five minutes and placed in ultra-freezers set at -80 and -20°C. After thawing at 38°C for five minutes, pearl millet pollen was placed on hairy stigma of pearl millet to check the germination. Pearl millet pollen was also used for the pollination onto emasculated wheat florets to investigate the formation of polyhaploid embryos, according to the method of Inagaki and Bohorova (1995).

Results and Discussion

Pearl millet pollen collected from the ears at anthesis had water contents of 20-50% and germinated on the stigma at frequencies of 70 - 80%. Pearl millet pollen germinated at frequencies of 40 - 50% when the water contents decreased to 5 - 7% after two hours of drying. Dried pollen kept similar germination frequencies after the freezing process. With no freezing treatment, the germination frequencies of dried pollen decreased as time passed, and the pollen lost viability after one week. The frequencies of embryo obtained from crosses of hexaploid wheat with pearl millet pollen are shown in Table 1. In crosses using fresh pollen, embryos obtained at a frequency of 27.6%. In crossed with the pollen stored for one month, the embryo formation frequencies ranged from 17.4 to 27.5%. after three months of storage, the frequencies kept a range between 14.2 and 29.1%. No significant difference in embryo formation frequency was found among the pollen water contents at the time of collection and among the storage temperatures. Hence we infer that pearl millet pollen is more tolerant to drying and freezing than is maize pollen, and stored pearl millet pollen would be an efficient polyhaploid production source for hexaploid wheat.

References

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Table 1: Embryo formation frequencies in crosses of hexaploid wheat with stored pearl millet pollen

Water content (%)		Storage temperature (° C)	Storage period (month)		
Fresh	Dried		0	1	3
			--- Embryo formation frequency (%) ---		
36.8	--	--	27.6	--	--
54.7	6.6	-196	--	24.3	28.5
		-80	--	21.1	26.0
		-20	--	23.8	26.7
36.8	7.6	-196	--	25.4	29.1
		-80	--	22.1	26.4
		-20	--	17.4	14.2
22.0	5.7	-196	--	27.5	25.7
		-80	--	22.4	20.5
		-20	--	18.2	14.3