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## Production of double haploids in oat (*Avena* sativa L.) by pollination with maize (*Zea mays* L.)

**Research Article** 

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Abstract: The aim of the study was to optimize the method of oat haploid production by pollination with maize. Seventeen oat genotypes were used in the experiment. Various factors influencing the growth and development of ovaries and embryo production were investigated: genotype, time of pollination, growth regulators and time of their application. Emasculated before anthesis, oat florets were pollinated with maize pollen after 0, 1 or 2 days. Next, one of two auxins analogues (2,4-D or dicamba) were applied to oat pistils. These auxins had no significant influence on the number of enlarged ovaries and embryos. The time of application of these growth regulators had a significant influence on embryo production. Haploid embryos were obtained from all used genotypes, although the frequency of enlarged ovaries and obtained embryos did not differ markedly between the genotypes. On average, 85% of ovaries were enlarged and 11.7% of them produced haploid embryos. Depending on the regeneration medium, 24–41% of embryos were germinated, of which 12% had developed into green plants. A strong significant difference in the number of germinating embryos and haploid plants was observed between the kind of regenerating medium used. There were no albino plants and all the obtained plants were haploid.

Keywords: Wide crossing • 2,4-D • Dicamba • Oat haploid embryos

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## **Abbreviations**

2,4-D- 2,4-dichlorophenosyacetic acid;dicamba- 3,6-dichloro-2-methoxybenzoic acid;DH- double haploid.

## 1. Introduction

Modern crop breeding has a number of advantages when using DH plants, such as shortening of the production time of new varieties and the possibility of using them in research. Classical methods of production of new varieties, based on a selection in every generation, take six to ten years. Biotechnological methods allow for a shortening of this time, even to one vegetative season. Breeders can make new cultivars with features from DH lines and be confident about completely homozygous plants. Moreover, DH lines can be used in the production of genetic maps, which help in the search for quantitative traits loci (QTL), conjugated with molecular markers and connected with many physiological processes, such as receptivity to the effectiveness of androgenesis or tolerance to biotic and abiotic stresses [1,2]. Haploids can be obtained using a number of methods, namely: from male gametophyte by androgenesis, using anther cultures or isolated microspores cultures [3-8], or from female gametophytes by gynogenesis or wide crossing, usually with maize (a method also known as chromosomes elimination) [1,9,10]. In comparison to anther culture, pollination with maize has an advantage because all regenerated haploid embryos are green, whereas in anther culture many of them are albino.

The method of chromosome elimination that has been used in these experiments includes the following stages: after emasculation of cereal flowers the stigma is pollinated by maize, the pollen then germinates, a pollen tube is formed [11,12] and grows into the cereal embryo sac, where the cereal egg is fertilised by the maize sperm nuclei. A hybrid zygote is produced. The hybrid zygotes