Highly efficient rye androgenesis – impact of genotype and various stress factors

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Homozygosity of breeding lines is supposed to be obtained in a short time through the doubled haploid (DH) system. After many years of experiments, regeneration of doubled haploids of rye is still a challenge and it is a strongly genotype-dependent process. The development of DHs is more problematic in out-crossing than in self-pollinating species.
The struggle for researchers has been always to develop an efficient method of rye doubled haploids production, but still, many problems remain unsolved. One of them is that the genotype was in the past and remains at present the primary factor that influences the efficiency of regeneration from microspores.
The switch of microspore development from gametophytic to sporophytic pathway has been stimulated by various stress factors. Such stress treatment not only stops pollen development but reprograms the microspore towards embryo formation. Moreover, the type of stress influences the number of regenerated albino and green plants, and also the efficiency of spontaneous chromosomes doubling.
In the present study we investigated the effects of various treatments on viability of microspores. We applied nine different stress combinations in order to induce androgenesis.
Experiments were performed on microspores and anthers of winter rye breeding lines. The results showed a correlation between the genotype and stress applied, with the level of androgenesis induction and mortality of microspores in the early culture stage. The survival of microspores was the highest for two genotypes, after cold pretreatment of tillers at 4°C for two weeks followed by preculturing of anthers in mannitol solution at 4°C for seven days. Cooling of tillers for three weeks, for anthers, was equally effective. The use of heat shock (32°C) led to 100% cells death. For the first time we present high efficiency isolated microspore divisions of rye and regeneration of nearly 2 thousand of green anther derived rye plants.