

# Genotypic and environmental variation in production of $2n$ -gametes of Oriental x Asiatic lily hybrids

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## Introduction

Introgression breeding between Oriental (O) and Asiatic (A) hybrid lilies is hampered by the (male) sterility of OA  $F_1$  hybrids. This crossing barrier can be overcome by means of polyploidisation. Meiotic polyploidisation (using unreduced gametes) is an attractive approach for this purpose because it allows homoeologous recombination between the Oriental and Asiatic chromosomes.

The number of  $2n$  gamete producing genotypes among OA-hybrids is naturally very low. A careful screening method was used to detect  $2n$  pollen producing genotypes in a large collection of OA  $F_1$  hybrids. Furthermore an attempt was made to induce the production of  $2n$  pollen in sterile OA-hybrids using heat shock treatment.

## Materials & Methods

The collection of OA  $F_1$  hybrid genotypes were grown in both heated (causing an environment with a more or less stable temperature) and unheated greenhouses (creating an environment with temperature fluctuation due to the daily changes in outside temperature). Additionally plants of four normally sterile genotypes were grown in a fytotron for 6 weeks and exposed daily to an extreme temperature fluctuation regime: 4 alternating periods of 10 °C and 30 °C each day.

Upon flowering all genotypes were screened for  $2n$  pollen production using a pollen germination test: Pollen was cultured on artificial bacteriological agar medium (100 g sucrose, 5 g bacteriological agar, 20 mg boric acid and 200 mg calcium nitrate per litre) over night at 25 °C. After 24 hours the pollen germination (i.e. viable  $2n$  pollen) percentage was scored.

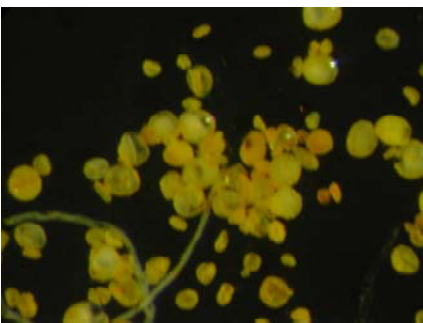


Figure 1.  $F_1$  OA pollen on pollen germination medium.

## Results

In total 12 genotypes were found to produce  $2n$  pollen in notable frequencies. Considerable variation in germination percentages was observed. Not only variation between the different genotypes but also variation between clones of the same genotype growing in different environments. There was even considerable variation among different clones of one genotypes within one environment and even among the different flowers of one clone.

Four of these twelve genotypes had already been identified as  $2n$  pollen producers in previous years. The other eight were all detected in the unheated greenhouse. Up until then they had always been grown in heated greenhouses and had always been tested sterile.

Figure 2.  $F_1$  OA hybrids used in the induction experiment.



A. 951301-5

B. 951914-1



C. 953508-1

D. 951462-1

All flowers of the plants from the heat shock treatment were tested for pollen germination. In total 2% of the flowers responded to the treatment. But among the four genotypes the response as well as the pollen germination percentage varied.

## Discussion & Conclusion

These results suggest that the variation in  $2n$  pollen production is of both environmental and genetic origin. Temperature fluctuation both natural (the unheated greenhouse) and artificial (the fytotron) might be an agent that stimulates the production of  $2n$  gametes. But not all results corroborate with this assumption: some genotypes showed higher pollen germination percentages in the heated greenhouse. Because of the large variation in  $2n$  pollen frequencies and the inconsistent nature of this variation it is advisable to repeat screening and screen under different environments. It is also demonstrated that heat shock treatment can be used successfully to induce  $2n$  pollen production for breeding purposes.