

## EVALUATION OF CUCUMBER ACCESSION HOMOGENEITY BY USING RAPD MOLECULAR MARKERS

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### Summary

The method for high quality DNA isolation from the cucumber seeds was developed. Sib- and inbred lines created from the old Latvian cucumber variety 'Dindoņa Zaļie Ķekaru' were evaluated for homogeneity by RAPDs. Five lines were selected for submission in the Latvian Gene Bank of Cultivated Plants based both on molecular and field evaluation data.

### Keywords

Cucumber, molecular markers, RAPD, homogeneity, DNA isolation

### Introduction

Latvia is close to the northern boundary of cucumber cultivation. Therefore cucumbers of Latvian origin may contain genes of tolerance to frost and broad temperature fluctuations. Till the middle of the 1990s, PGR activities in Latvia were not conducted. Therefore evaluation of the available varieties and their entry to the gene bank became important and immediate tasks. In 1997, the Latvian Gene Bank of Cultivated Plants was established in the framework of the Nordic–Baltic cooperation in PGR [1]. Several available cucumber samples were included in the list of accessions to be preserved. The problem of accession quality was raised, as genetic erosion of some of them during commercial seed production was observed. For example, only 3% of plants of the old Latvian variety 'Dindoņa Zaļie Ķekaru' grown from commercially available seeds were morphologically corresponded to the variety description. Such material was not valuable for entry in the gene bank. Therefore homogenisation of the variety was started with the aim to select homogeneous lines that best corresponded to the original trait complex of the variety 'Dindoņa Zaļie Ķekaru', for placement in the Latvian Gene Bank of Cultivated Plants.

### Material and Methods

Several inbred and sibilines of the cucumber variety 'Dindoņa Zaļie Ķekaru' were created. For creating inbredlines, the most typical plants from commercially available seeds were chosen for selfing. During six generations, plants most corresponding to the original description of the variety by a complex of traits were selected for further inbreeding. For creating sibilines, plants most corresponding to the variety were selected among plants grown from uncontrolled pollination. Next year progenies of each such plant were recognised as a sibline. Selection of the most typical plants and pollination in each following was made within sibilines. Phenology, morphological parameters (plant branching, flowering type, fruit development in clusters, fruit shape, cross section of fruits and size of ovary, fruit colour, drawing, density and size of warts, colour and type of prickles) and yield were scored for six generations of inbredlines and three generations of sibilines.

The method for DNA isolation from the cucumber seeds was developed by modification of the method used by Roder et. al. for wheat [2]. Molecular marker (RAPD) analysis was performed according to Staub and Meglic [3] for single plants of the 6<sup>th</sup> generation of inbredlines and the 3<sup>rd</sup> generation of siblines.

### **Results and Discussion**

High quality DNA was isolated from the cucumbers seeds. In total 26 oligonucleotide primers among 53 tested were polymorphic. The level of loci polymorphism for the amplified primers varied between 50 and 100%. The length of amplification products was between 250 and 3000 bp (base pairs), which is similar to that estimated earlier [3]. The calculated coefficient of similarity between all accessed plants varied between 0.04 and 0.91. There was a significant difference in average values of similarity coefficients between the lines. Average coefficient of similarity between the plants of the most homogeneous inbredline was 0.83, compared to far the least homogeneous line 0.46.

Some contrasting results were obtained for the phenotypic and molecular evaluation of genotypes. Genotypes ascertained as homogeneous by phenotypic evaluation were not the most homogeneous according to results of the RAPD analysis but the differences were not very large. Dendrograms developed from both phenotypic and molecular data were similar. Inbredlines were grouped mainly in the side clusters, and siblines mostly in the mid part of the dendrogram. As a result of inbreeding, lines are more distinct than after sibling.

Differences between results of phenotypic and molecular analyses could have resulted from interaction of environment and genotype in forming traits. Phenotypic evaluation can be insufficient for making the final decision about including an accession in a gene bank for long-term preservation. RAPD markers together with field evaluation are appropriate for detection of homogeneity and distinctiveness of accessions.

Three inbredlines and two siblines were placed in the Latvian Gene Bank of Cultivated Plants taking into account both phenotypic and genotypic evaluation. Lines most homogeneous and corresponding to the phenotypic description of the variety, and the most homogeneous lines according to the RAPD analyses were chosen. The lines were selected from different clusters of the dendrogram to cover the genetic diversity of the variety.

### **REFERENCE LIST**

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