

THE GROUPING OF SOME *ANGELICA ARCHANGELICA* L. FAMILIES WITH HELP OF DENDROGRAM OBTAINED BY RAPD TECHNIQUE

Mihai Radu POP*

*"Lucian Blaga" University of Sibiu, Department of Biology, Romania
mihaipop76@yahoo.com

Abstract: *Angelica archangelica* L. species represents a floristic rarity, is declared endangered species, its harvesting from spontaneous flora not being permitted [4]. For obtaining vegetal material for industrialization, this is realized by cultivation of the species [8].

Modern ameliorating has the tendency to enrich scientific , using methods that will permit the acceleration of process and accuracy by diminution of his probabilistic character. Using of molecular markers is inscribed in the same tendency, wich consists in one of the most efficient methodes wich offers an important opportunity in analyzing, administrating and using of genetic variability through RAPD technique.

Keywords: *Angelica archangelica* L., RAPD, dendrogram.

INTRODUCTION

Regarding anterior research of *Angelica archangelica* L species, some authors consider that the more extended origin zone is situated in Eurasian boreal zone, to Central Russia, from where the greatest part of Europe was naturalized (LARWLESS, 1992). Other authors consider it originative from Syria, from where it spreaded in cold european climate [3]. PĂUN et al (1986) think that *Angelica archangelica* L. is an Eurasian species, met from Siberia to Island, and POTLOG și VINȚAN (1983) consider it originating from north of Europe.

The introduction in culture of *Angelica archangelica* L. species will be an important accomplishment in the line of durable developpement, offering an income source to the farmers in the mountain area and maintaining its biodiversity and charm, responding to european demands in this field, which Romania will have to respect after joining European Union [2].

The used biological material, the population "De Cristian", especially heterogeneous, with big amplitudes of variation under the aspect of morphological characteres, made the effectuation of some selection works regarding this genotype possible.

Within improvement works, developed in Brasov and Sibiu, 10 important families of *Angelica archangelica* L., were identified, noted with G₁-G₁₀, under cantitative and calitatrive aspect, compared with populația "De Cristian" population (POP, 2006) [7].

The reasearch pointed out the fact that biological characteristics of *Angelica archangelica* L. species have major impact both on the improvement method and on the culture technique and production of seed. Because of this we set the goal of studying the genetic variability of selected families.

MATERIALS AND METHODS

For obtaining vegetal material from wich DNA was prelevated, we took seeds from the 10 selected families

and from local population „De Cristian” from wich little plants used for prelevating ADN were obtained.

DNA prelevation was realized after the protocol described by AUSUBEL et al. (1990), using the CTAB method.

For appling RAPD technique, the amplification of DNA was produced. Very good results were obtained with decameric fuses (with ten nucleotides) wich were purchased from Promega – MADISON WI USA company.

The amplification was realized with a termocycler wich was programated for an initial cycle of 94°C for 5 minutes, followed by 40 cycles of 1 minute at 94°C, 1 minute at 36°C and 2 minutes at 72°C, followed by the last cycle at a temperature of 72°C, for 8 minutes and then preserving the samples at 4°C until the moment of the electrophoresy in gel [6].

The electrophoresy was programmed at 2.5 V/cm for 180 minutes. The gel, after migration, was maintained in a solution of bromură de etidiu, with a concentration of 0.5 μl/ml, on an agitating apparatus with a continuous movement of 150 rot./min. for 20 minutes. The gel prepared in this way was examined in an UV transiluminator, scheduled with a photo camera for preserving the image, using the programme Imager Appligene (Imager Appligene, Chester-Le-Street, UK)

RESULTS AND DISCUSSIONS

The results followed after the amplification of DNA, obtained from the ten selected families of *Angelica archangelica* L compared with the local „De Cristian” population, through utilisation of primers, materialized in the apparition of polimorphic bands. The studied families were grouped favorable to their degree of genetic approach or distance.

Analyzing the polimorphic bands obtained through RAPD technique the dedrogram was made (**Fig. 1**) wich shows the way the analized families can be grouped after the distance or approach of the analized families that shows clearly the difference between the DNA resulted from the families of *Angelica archangelica* L and the DNA coming from "De Cristian" population.

Between the ten families of selected angelica, reading the dendrogram, lead to their separation in four classes, one with 4 families and three with 2 families

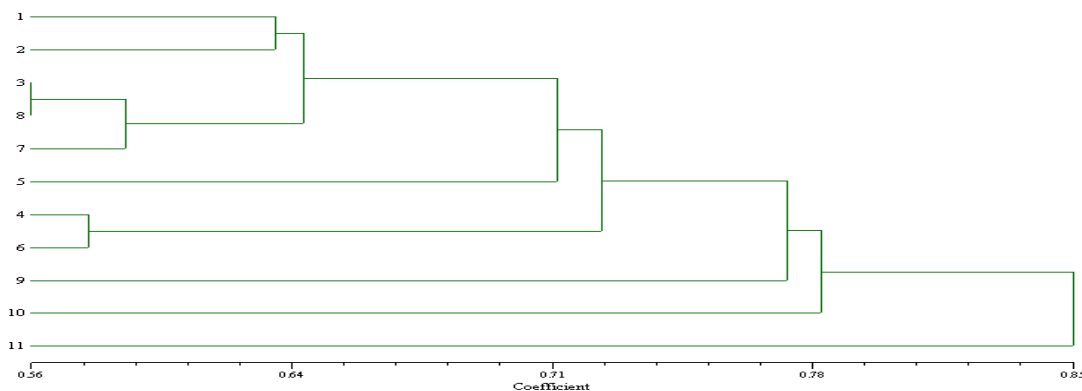


Figure 1. Dendrograma familiilor și populației de *Angelica*, analizate cu ajutorul tehnicii RAPD

The built dendrogram shows that the analysed samples can be grouped in five separate sub-groups:

- the sub-group of 1 and 2 families
- the sub-group of 3, 8, 7 and 5 families;
- the sub-group of 4 and 6 families;
- the sub-group of 9 and 10 families
- the sub-group of „De Cristian” population;

Based on the dendrogram analysis we can say that between the families of the species *Angelica archangelica* L an important degree of genetical polymorphism exists.

Due to the fact that in our analysis we used DNA from the selected families of *Angelica archangelica* L. And also from the „De Cristian” population, the waited results were supposed to lay between the two poles, which is in the proximity between the local population and any other family, theoretically, being the least in this case. This thing can be noticed from the dendrogram constructed on the laboratory results, fact that confirms in a way the veracity of the RAPD technique, and on the other way the correctness of the application of the work protocols.

The dendrogram shows clearly the difference made between the DNA resulted from the *Angelica archangelica* L. families and the DNA coming from „De Cristian” population.

CONCLUSIONS

- In the present study, it is distinguished that the ten selected families are totally different from the De Cristian population, the last being placed to a significant distance from the rest of samples taken into study.
- It can be foreseen, interpreting these results, that the cross-breeding between the families of *Angelica archangelica* L. species won't encounter difficulties, creating the possibility of genetical diversification of the improvement material.
- The dendrogram analysis makes us conclude that for distinguishing the cross-breeding possibilities at the *Angelica archangelica* L. species, an aid consists in applying the RAPD technique.

each. This separation shows the existence of one remarkable genetical polymorphism.

- The maximum genetical diversity, resulted from intraspecific cross-breeding in the *Angelica archangelica* L. species, can be expected in case of using as genitors families belonging to different sub-groups, with a coefficient of genetical distance as big as possible.
- The best example can be expressed through the G₅ și G₁₀ families on one way, and through the G₈ și G₉ families on the other way.

REFERENCES

- [1] Ausubel, F.M., Brent, R., Kingston, R.E., Moore, D.D., Seidman, J.G., Smith, J.A., Struhl, K., (1990). Current Protocols in *Molecular Biology*, John Wiley & Sons.
- [2] Bobiț, D., Sand, C., Drumea, V., (2002). Rezultatele cercetărilor privind identificarea genotipurilor valoroase la specia *Angelica archangelica* L., in: *Lucrările Conferinței Internaționale „Științe, Procese și Tehnologii Agro- Alimentare”*, Sibiu, 31.10-01.11.2002, Editura Universității „L. Blaga” din Sibiu.
- [3] Bown, D., (1995). *Encyclopaedia of Herbs and their Uses*, Dorling Kindersley, London, Great Britain.
- [4] Crăciun, F., Bojor, O., Alexan, M. (1976). *Farmacia naturii*, Editura Ceres, București.
- [5] Păun, E., Mihalea, A., Dumitrescu, A., Verzea, M., Coșocariu, O., (1986, 1988). *Tratat de plante medicinale și aromatice cultivate*, vol. I-II, Editura Academiei RSR, București.
- [6] Pop, M.R., Sand, C., Savatti, M., Cătană, C., Barbu, H., Power, J.B., Davey, M.R., (2003). Testing of 20 OPC primers for the RAPD analyze of ten genotypes of *Angelica archangelica* L., *Buletinul Universității de Științe Agricole și Medicină Veterinară-Cluj-Napoca*, Seria Zootehnie și Biotehnologii, Vol. 59, pp.306, ISSN 1454-2382.
- [7] Pop, M., (2006). Identificarea unor genotipuri de *Angelica archangelica* L., prin metode convenționale și neconvenționale în vederea ameliorării, *Teză de doctorat*, Cluj-Napoca.
- [8] Potlog, A.S., Vințan, A.G.H., (1983). *Plante medicinale*, Editura Științifică și Enciclopedică, București.