

ASPECTS OF „IN VITRO" MULTIPLICATION AND ACCLIMATISATION IN *DROSERA ROTUNDIFOLIA* (FAM. DROSERACEAE)

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1. Introduction

Drosera rotundifolia L. (roua cerului) is a carnivorous plant from *Droseraceae* family, met in mountain peat-bog. She is until 26 cm in height, with basal leaves disposed in rosette, specialised in capture and digestion of insects, with adaptation to carnivorous digestion. The plants from *Drosera* genus present a pharmaceutical value, because they present a content in naphtoquinone (droserone, oxidroserone, plumbagone, ramentaceone a.o.) Through „in vitro" culture, it is possible to obtain a big quantity of matter for the extraction of bioactive substances.

2. Material and method

The „in vitro" culture initiation at *Drosera rotundifolia* L. has used plants from subculture, originating from Mrs. Dr. Dorina Cachita - Cosma (Institute of Biological Research from Cluj). Segments of 5 mm length from leaves or petiole were inoculated on a MS basal medium, or on a MS basal medium supplied with different bioactive substances: putrescine in different quantity: IAA (indolyl acetic acid) and KIN (kinetine) ± magnetic fluids on petroleum basis (LMP) and active coal (Table 1).

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Chemical composition of the culture media used for „in vitro” multiplication in *Drosera rotundifolia* L.

Component	Culture media							
	13	65	66	63	64	61-1	61-2	61C-2
Macroelements	MS	MS	MS	MS	MS	50% MS	50% MS	50% MS
Microelements	MS	MS	MS	MS	MS	MS	MS	MS
Thiamine-HCl (mg/l)	0.1	0.1	0.1	0.1	0.1	0.3	0.3	0.3
Piridoxyne-HCl (mg/l)	0.5	0.5	0.5	0.5	0.5	-	-	-
Nicotinic acid (mg/l)	0.5	0.5	0.5	0.5	0.5	-	-	-
Mioinositol (mg/l)	100	100	100	100	100	100	100	100
IAA (mg/l)	-	-	-	-	-	1.8	1.8	1.8
KIN (mg/l)	-	-	-	-	-	0,011	0,011	0,011
Active coal (g/l)	-	-	-	-	-	-	-	20
Putrescine (mg/l)	-	0.5	1.0	1.0	3.0	-	-	-
LMP (mg/l)	-	-	-	-	-	30	1.380	60
Succrose (g/l)	30	30	30	30	30	30	30	30
Agar-agar (g/l)	8	8	8	8	8	8	8	8

30 days after inoculation, phenotypical and biometrical observations (diameter of basal leaves rosette and rhysogenesis process) were effected. The explants were maintained in a growth room at $23^{\circ} \pm 2^{\circ}\text{C}$, and at 16 h light per day (2200 lx light intensity).

3. Results and method

The explants of *D. rotundifolia* have presented a different response dependent on nature and concentration of bioactive substances from culture media (growth hormones or other substances as putrescine or magnetic fluid). They determine different aspects of the organogenesis processes.

Basal leaves rosette diameter

The observations recorded at 20 and 30 days after inoculation, do present a big difference in the plants response, depending on nature and concentration of bioactive substances from culture media.

On media supplied with putrescine (1.0 and 3.0 mg/l), the diameter of basal leaves is greater in comparison with control (Fig. 1) at 20 and 30 days after inoculation. Supplementation of culture media with magnetic fluid on petroleum basis (30, 60 and 1380 mg/l), has presented an inhibitory effect on the development of basal leaves, in comparison with control (Fig. 1).

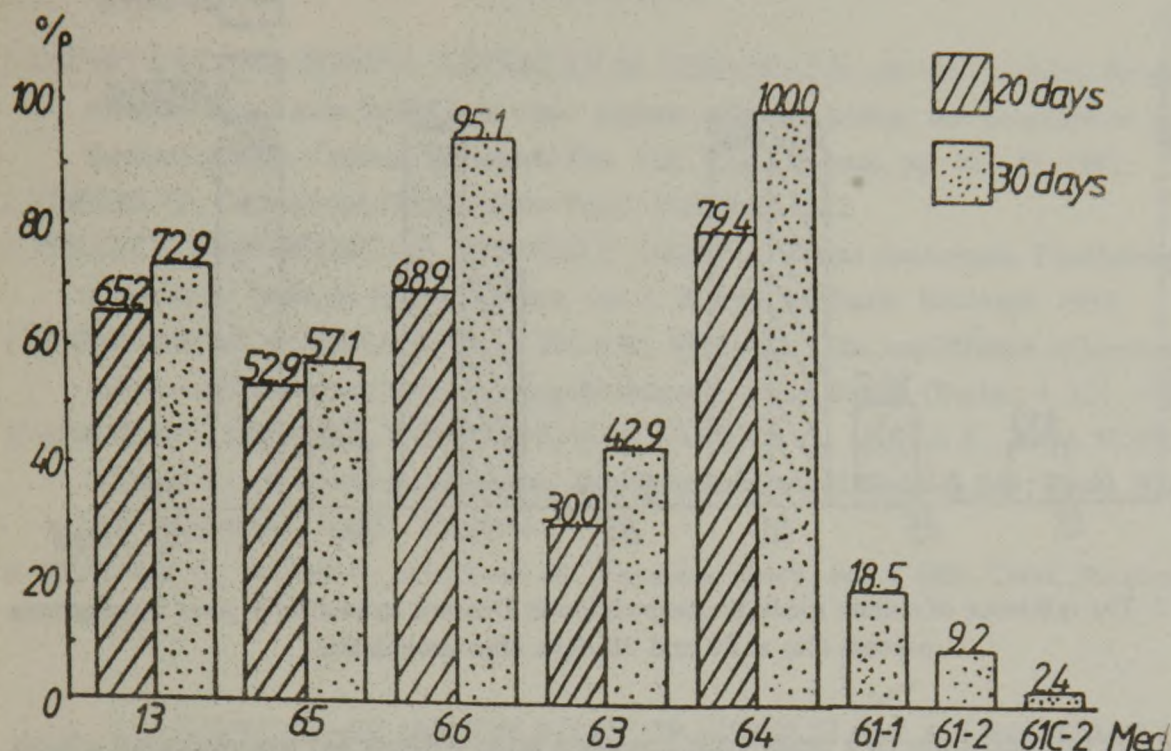


Fig. 1. The influence of culture medium composition at *Drosera rotundifolia* over basal leaves rosette diameter (%) at 20 and 30 days after inoculation.

Rooting process

At 20 days after inoculation, the rooting process was absent on control medium, as well as on media supplied with magnetic fluid. The high percent of rooting registered on media supplied with 1.0 mg/l putrescine (+ 60% in comparison with control), or with 3.0 mg/l putrescine (+ 37.5% in comparison with control).

At 30 days inoculation, at control the rooting percent was 10%, while on media supplied with putrescine (1.0 or 3.0 mg/l) or with magnetic fluid of LMP type (1380 mg/l), the rooting percent was 60%, 62.5% respectively 60% (Fig. 2).

The rooting process was absent on media supplied with 0.5 mg/l putrescine, as well as on media supplied with 60 mg/l LMP and active coal. In this case, the presence of active coal presents an inhibitory effect on the rhysogenesis process.

Acclimatisation

The plants with well developed radicular system were used for acclimatisation. The roots were fashioned until a length 2 times greater in comparison with the diameter of basal leaves rosette. They were planted on a mixture formed from rot peat and perlite at a pH of 6.8. The planting vessels

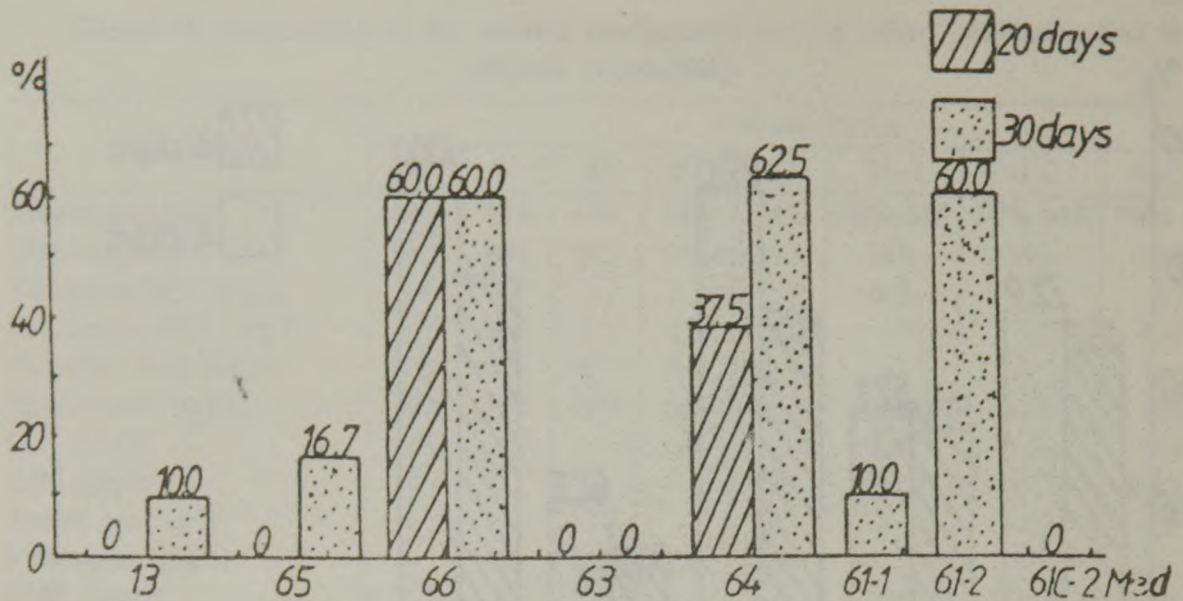


Fig. 2. The influence of culture medium composition at *Drosera rotundifolia* over rhylogenesis process (%) at 20 and 30 days after inoculation.

were introduced then in a aquarium covered with a glass for maintaining a high humidity. The sunstroke regime of aquarium was moderate, with a temperature of 35° - 40°C (Summer, in interior). After 3 weeks, the plants were acclimatised and the were fed with *Drosophila melanogaster* from culture, followed by well development of nettled hairy.

4. Conclusions

- The supplementation of a MS basal medium with 3.0 mg/l putrescine, has presented the best stimulation development of the basal leaves rosette in *Drosera rotundifolia*.

- Rhylogenesis process was enhanced on a MS basal medium supplied with 1.0 or 3.0 mg/l putrescine (at 20 and 30 days after inoculation) or on a MS basal medium (with 50% macroelements) supplied with 1380 mg/l magnetic fluid of LMP type (with petroleum as dispersion phase).

- The presence of active coal together with 60 mg/l magnetic fluid of LMP type in culture medium, has presented an inhibitory effect on the rhylogenesis process.

- The plants with well developed radicular system, were planted in a mixture formed from rot peat and perlite (pH = 6.8) in a damp atmosphere, for 21 days.

REFERENCES

1. CACHIȚĂ-COSMA DORINA, ZĂPĂRȚAN M., CRIGORAȘ S., „In vitro” cultured *Drosera rotundifolia* - a new biotest. „In vitro” explant cultures - present and perspectives. (Ed. Dorina Cachiță - Cosma), Biological Res. Inst., Cluj - Napoca, pp. 42 - 43, 1991.
2. CHEERS G., Carnivorous Plants, Globe Press, Melbourne, 1983.
3. CIULEI I., GRIGORESCU E., STĂNESCU URSULA, Plante medicinale, Fitochimie și Fitoterapie. Tratat de Farmacognozie, vol. I, II, Edit. Medicală, București, 1993.
4. CORNEANU M., CORNEANU G. C., BICA D., VEKAȘ L., The amplification of hormones role in culture medium through using the magnetic fluids, SozEp. (Berlin), 4, 121, 1993.
5. CORNEANU MIHAELA, CORNEANU G. C., BĂDICĂ C., MINEA R., BICA DOINA, VEKAȘ L., „In vitro” organogenesis at *Aloe arborescens* (Liliaceae), Rev. Roum. Biol., Biol. Végét., 39 (1), 45 - 52, 1994.
6. CRĂCIUN F., BOJOR O., ALEXAN M., Farmacia naturii, vol. I, Edit. Ceres, București, 1976.

ASPECTE ALE MULTIPLICĂRII „IN VITRO” ȘI ALE ACLIMATIZĂRII LA *DROSERA ROTUNDIFOLIA* L. (FAM. DROSERACEAE)

Rezumat

Drosera rotundifolia L. este o plantă farmaceutică cu un conținut bogat în naftiquinone. Ea prezintă însă o utilizare limitată deoarece este întâlnită numai în turbării în cantitate mică. Cultura „in vitro” permite obținerea unei cantități mari de material vegetal pur, tot timpul anului, care poate fi utilizat cu succes în industria farmaceutică. Pentru cultura „in vitro” în acest experiment s-au utilizat fragmente de pețiol și frunze care au fost înoculate pe un mediu bazal MS suplimentat cu: (a) IAA 1,8 mg/l și KIN 0,011 mg/l + lichid magnetic de tip LMP (cu petrol de fază de dispersie) în cantitate de 30, 60 sau 1380 mg/l, sau (b) putresceină în cantitatea de 0.5 - 3.0 mg/l. Cele mai bune rezultate pentru dezvoltarea plantelor (diametrul rozetei de frunze bazale și procesul de înrădăcinare) au fost obținute pe un mediu bazal MS fără hormoni de creștere, suplimentat cu putresceină (1,0 - 3,0 mg/l), precum și pe un mediu bazal MS cu hormoni de creștere (1,8 mg/l IAA și 0,011 mg/l KIN) suplimentat cu 1380 mg/l LMP (pentru procesul de înrădăcinare). Este prezentat, de asemenea, procesul de aclimatizare la *Drosera rotundifolia*, în cazul culturii „in vitro”.