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CRUPINA VULGARIS CASS. IN VITRO CULTURE OBTAINING

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The *in vitro* cultivation conditions for obtaining the isolated culture of *Crupina vulgaris* were firstly developed. Callus culture was obtained on MS (Murashige and Skoog) basal medium, supplemented with 2.0 mg/L indole 3 acetic acid (IAA), 0.2 mg/L kinetin, but further growth was supported on both MS medium and it`s modification, containing 6-*benzylaminopurine* (BAP) 2mg/l, IAA - 0.5mg/l.

Crupina vulgaris- callus culture

The *Crupina* genus plants species belonging to the Asteraceae family which contain secondary metabolic products with high biological activity are widely used in medicine, cosmetic and food industry. Plant materials from Asteraceae family might be bioactive secondary metabolites that have the potential to treat different afflictions. Examples of these compounds include phenols, phenolic glycosides, unsaturated lactones, sulfur compounds, saponins, cyanogenic glycosides and glucosinolates [7, 9]. A number of species are considered invasive. Different plants contain different bioactive compounds and these vary with area, climate and mode of agricultural practice if they are not present in wild environment. Herbivory, pathogens and competition are the driving forces that induce plant species to develop chemical defense compounds. These plant origin compounds are good models for elucidation of their functional roles in medication and treatment of different afflictions. Poisonous plants exposed to frequent grazing by animals are commonly rich in alkaloids which have many biological activities including anticancer potential [5]. However, the growth regulatory properties of some plant metabolites allow them to act as chemotherapeutical agents. Flavonoids from *Scutellaria baicalensis* act on cyclin-dependent kinases to inhibit cancer cell proliferation [1]. Members of the Asteraceae family produce a large number of various secondary metabolites that show biological activity. Among them, sesquiterpene lactones and flavonoids are the most interesting ones from the pharmacological point of view. These substances are known for their reported medical efficacy e.g. strong anti-inflammatory, antimalarial, antioxidant, antitumor activity, as well as for the fact that they increase immunity and decrease the risk of atherosclerosis, arthritis and gastrointestinal disorders [7]. In many cases, the α-methylene group in the lactone ring of sesquiterpene lactones, being potentially able to bind the nucleophile sites of biomolecules by conjugate addition, manifests their biological activity [4].

The genus *Crupina* comprises three species: *vulgaris, crupinastrum*, and *intermedia*. Ranges of *C. vulgaris* and *C. crupinastrum* overlap in Spain, Italy, Greece, and Turkey, although in each case, *C. vulgaris* grows on more northern or more mesic sites than *C. crupinastrum* [2]. The range of *C. vulgaris* extends northward to the dry valleys of the Alps in Switzerland, France and Italy, where it is restricted to open grasslands on steep south-facing slopes [10].

C. vulgaris normally grows on calcareous loam or clay loam soils of limestone parent material, and is rarely found, usually as an accidental introduction, on sandy soils or over siliceous (acidic) parent material [3].

Material and methods

Plant material. The investigated plant *C. vulgaris* Cass. was collected from Aragatsotn region (Mughni) in Armenia (1500-1600 m above sea level, N 40° 22.085′, E 44° 22.815′) during the flowering period.

Obtaining of isolated cultures. The leaves and stems were used as explants. To isolate tissue cultures the explants (approx. diameters of explants - 0.8 to 1.2 cm) were sterilized with solution of 330 mg/L mercuric chloride and 660 mg/L cetylpyridinium chloride for 8-10 min followed by four rinses with sterile distilled water. The sterilized explants were individually aseptically placed into petri dishes, containing 20 mL Murashige and Skoog (MS) nutrient medium [6], supplemented with 2.0 mg/L indole 3 acetic acid (IAA), 0.2 mg/L kinetin, 0.1 g/L myo-inositol, 0.1 mg/L thiamine HCl, 0.5 mg/L pyridoxine HCl, 0.5 mg/L nicotinic acid, 2.0 mg/L amino acetic acid, 30 g/L sucrose and 8 g/L agar. The explants were placed in thermostat at 25°C (for the initiation of proliferation processes). Afterwards the formed primary callus tissues were placed in the flasks (50 mL) then replaced to the thermostat (in the same conditions).

To optimize medium composition for shoot multiplication, *in vitro* cultivation was performed in MS basal medium (including vitamins, sucrose and agar), supplemented with BAP-2mg/l, IAA-0.5 mg/l. This medium was conditionally named MR_{30}. Both of media were adjusted to pH 5.8 with 0.1 M NaOH before autoclaving for 20 min at 121°*C*. The cultures were placed in growth chamber at 25°C and illuminated with a 16-h photoperiod (natural daylight, supplemented with artificial light (approx. 150 μmol quanta m^{-2} s^{-1}) provided by a white fluorescent lamp (Philips Inc., 36 W). Callus tissue further stable growth was supported in both MR_{30} medium. *In vitro* micro-propagation of *C. vulgaris* was carried out using modified variant of nutrient medium.

Results and discussion

The leaves and stems were used as explants in this investigation. Callus formation was occurred only on the explants of leaf origin. The proliferation begins on 7-8th days on MS medium and the primary callus was formed on 18-20th days of cultivation. On the MS medium *C. vulgaris* callus culture has darker color and growth was generally directed over the surface of the agar medium (Fig. 1a), whereas the growth on MR_{30} medium callus tissue was directed upward. Callus tissue of *C. vulgaris* had granular consistency and light yellow color growing in dark conditions (Fig.1b).

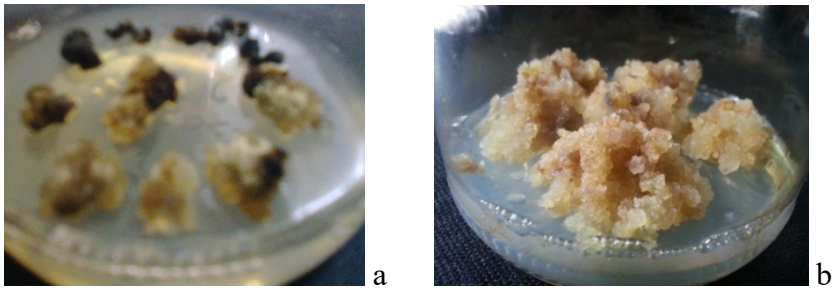

Fig. 1. Callus cultures of *C. vulgaris*, growing on MS medium (a), growing on MR_{30} medium (b).

After several passaging light color of callus culture has changed into a light brown with patches of white meristematic zones. Such tissue undergoes spontaneous organogenesis under the light (Fig. 2).

Fig. 2. Organogenesis of *C. vulgaris* on the MR_{30} medium.

On the same nutrient medium root formation occurs. The leaves and roots of *C. vulgaris* regenerated plants were placed on the MR_{30} fresh nutrient medium and callus culture was formed in these conditions.

Generally, the growth cycle of calli was ended on the 27-28th day, but in case with *in vitro* plantlets – on the 35-40th day of cultivation.

Conclusions

Summarizing data of our investigation might be possible to conclude that isolated culture of *C. vulgaris* can be obtained using MS medium modified variant in which the concentration of different auxins is prevalent on the cytokinin concentration of more than 7 fold.

Previous studies unveiled the high metabolic activity of isolated cultures of different plants growing in Armenia [8, 11, 12]. The metabolic activity of *C. vulgaris* on *in vitro* cultures is not reported. Since both *in vitro* cultures and intact plants of *C. vulgaris* have intensive growth rate, they can be used as model systems in biotechnological investigations to produce valuable metabolites.

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THE STUDY OF THE CHANGES IN ACTIVITY OF SOD IN PATHOPHYSIOLOGY OFGASTRIC CANCER DEPENDING ON THE STAGE OF DISEASE

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Superoxide dismutase (SOD) is an antioxidant enzyme that plays an important role in the defense system of the body. The aim of this study was to investigate the changes in activity of superoxide dismutase (SOD) in the plasma and tumor tissue of patients with gastric cancer depending on the stage (I-IV) of disease. It has been shown that SOD activity in the plasma of gastric cancer patients is significantly reduced compared with SOD activity in the plasma of healthy donors. In the same time tumor tissue SOD activity is increased compared with healthy (histologically examined) control tissue. The results obtained indicated that changes of SOD activity in plasma and tumor tissue of gastric cancer patients depend on the stage of disease.

Gastric cancer - superoxide dismutase - reactive oxygen species

Սուպերօքսիդդիսմուտազը (ՍՕԴ) հակաօքսիդանտային ֆերմենտ է, որը կարևոր դեր է խաղում օրգանիզմի պաշտպանողական համակարգում: Այս հետազոտության նպատակն էր ուսումնասիրել ՍՕԴ-ի ակտիվության փոփոխությունը ստամոքսի քաղցկեղով հիվանդների արյան պլազմայում և ուռուցքային հյուսվածքում կախված հիվանդության զարգացման փուլից (I-IV): Ցույց է տրվել, որ ստամոքսի քաղցկեղով հիվանդների արյան պլազմայում ՍՕԴ-ի ակտիվությունը նշանակալիորեն նվազում է առողջ դոնորների արյան պլազմայում ՍՕԴ-ի ակտիվության համեմատ: Միևնույն ժամանակ, ուռուցքային հյուսվածքում ՍՕԴ-ի ակտիվությունն աճում է առողջ (հյուսվածաբանորեն զննված) ստուգիչ հյուսվածքում ՍՕԴ-ի ակտիվության համեմատ: Ստացված տվյալները ցույց են տալիս, որ ստամոքսի քաղցկեղով հիվանդների պլազմայում և ուռուցքային հյուսվածքում ՍՕԴ-ի ակտիվության փոփոխությունը կախված է հիվանդության փուլից:

Ստամոքսի քաղցկեղ – սուպերօքսիդդիսմուտազ – թթվածնի ակտիվ ձևեր

Супероксиддисмутаза (СОД) - антиоксидантный фермент, который играет важную роль в системе защиты организма. Целью данной работы было исследование изменений активности супероксиддисмутазы (СОД) в плазме и опухолевой ткани пациентов с раком желудка в