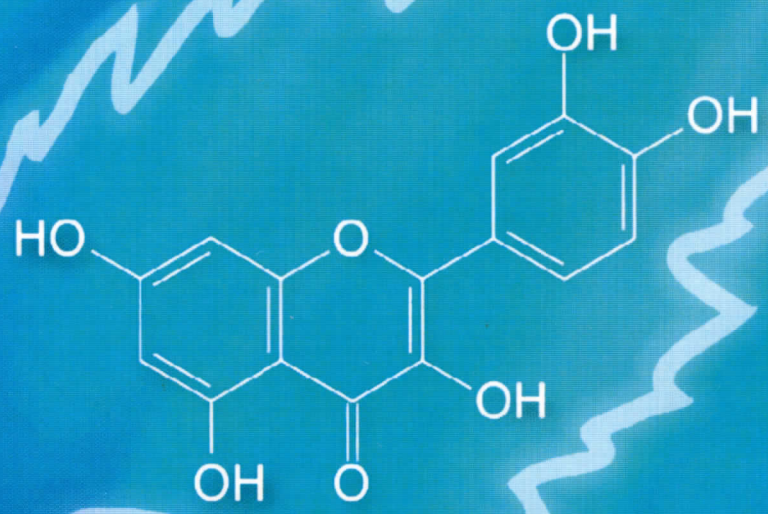
РОССИЙСКАЯ АКАДЕМИЯ НАУК

ФЕНОЛЬНЫЕ СОЕДИНЕНИЯ:

ФУНКЦИОНАЛЬНАЯ РОЛЬ В РАСТЕНИЯХ



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В сборнике представлены результаты исследований по изучению полифенолов, регуляции их образования и распределения в клетках и тканях растений. Рассматриваются вопросы их участия в растительной экофизиологии (биотические и абиотические стрессы, патогенез, устойчивость, сигналинг). Сообщается об использовании методов биоинформатики при изучении фенольного метаболизма.

Для широкого круга специалистов по физиологии и биохимии растений, биотехнологии, агротехнологий, защиты растений, а также студентов и аспирантов высших учебных заведений.

Материалы публикуются в авторской редакции с согласия авторов.

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FLAX CELL CULTURES FOR THE PRODUCTION OF BIOACTIVE COMPOUNDS

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Abstract. Different flax species have been exploited as starting material to obtain in vitro cell cultures for the improvement of lignans production. Stable cell lines were established from leaf explants of Linum usitatissimum and Linum austriacum. The results highlight the differences in phenol content between cell suspensions deriving from two flax species belonging to different clades.

Flax is one of the oldest and highly cultivated oil seed crop in

Europe and is regarded as a functional food due to the presence of high quality omega-3 fatty acids (α-linolenic acid), proteins, lignin, phenolic acids (lignans), and dietary fibers [1]. Besides, industrial and nutritional importance, flax has a long history of medicinal uses due to the presence of biologically active components mainly belonging to the class of lignans, which exhibit antioxidant, cytotoxic, antifungal, antiviral and phytoestrogenic properties [2]. A thousand of studies have demonstrated the role of phytoestrogens as health promotors, in cancer prevention and in tumors treatments. [3]. Considering the growing need of these medicinally and nutritionally important molecules, there is a great interest in finding alternative platforms for their production other than conventional plant cultivation in order to meet their worldwide demand. Among different techniques, plant *in vitro* cultures offer an attractive alternative method for production of these valuable metabolites.

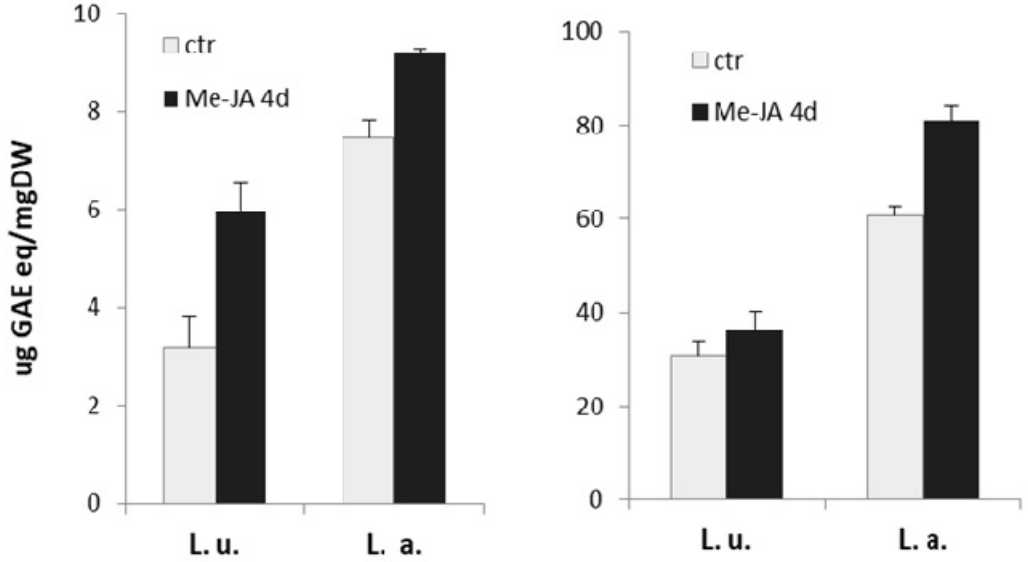

Fig.1. Total phenol content from (at left) cell suspensions of Linum usitatissimum (L. u.) and Linum austriacum (L. a.) in control (ctr) and 4 days Me-JA treatment and (at right) the corresponding media. The results are expressed as µg of Gallic acid equivalent (GAE) per mg of dry weight (DW)

In this work, different flax species have been exploited as starting material to obtain *in vitro* cell cultures for the improvement of lignans production. The flax species have been selected considering that lignan profile correlated with the genus systematic division of flax species. So far, three clades of linum species have been described on the basis of the type of lignan accumulated: aryltetralin-(AT)-lactone type, arylnaphthalene-(AN) and dibenzylbutanediol-type

lignans [4]. We have obtained cell suspensions from a few flax species belonging to each clade and we are analyzing their ability to produce bioactive compounds with the aim to select the flax species with the highest level of secondary metabolites. To this purpose the total phenol content, the antioxidant capacity and the metabolite profile of the cell extracts have been analyzed. Moreover, elicitors such as methyl jasmonate (Me-JA) have been used to increase metabolites production.

Results. Stable cell lines were established from leaf explants of *Linum usitatissimum* (cv Valoal) and *Linum austriacum*. Cell suspensions obtained from these lines were analized for their total phenolic content (Fig. 1). A significative difference was present between control and four days Me-JA elicited cells (Fig.1a). Moreover, *L. austriacum* extracts showed a higher phenolic content than *L. usitatissimum* in both control and treated samples. The same analyses were performed on culture media in order to verify if cells were able to secrete the phenolic compounds into the medium. As shown in Fig. 1b, the phenol content of the media from *L. austriacum* suspensions was almost double that of *L. usitatissimum*.

To obtain the metabolite profiling of flax cell suspensions the ^{1}H-NMR spectra were recorded at 14.09 T. The aromatic region of the ^{1}H-NMR spectra of the two species analized is shown in Fig. 2. The spectra highlighted that the two species contain different aromatic molecules and also that *L. austriacum* samples produce a higher number of compounds. On the basis of the resonances assignments the molecules belong to the lignan class.

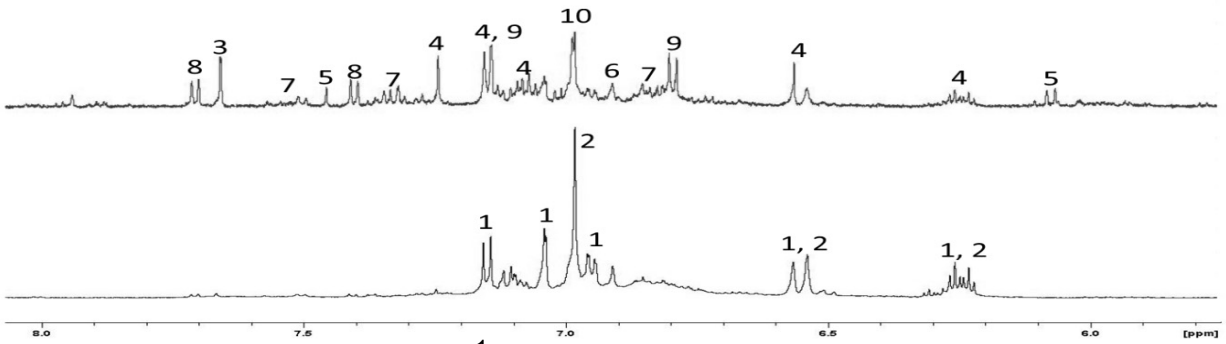

Fig. 2. Aromatic region of ^{1}H-NMR spectrum of extracts from flax cell suspensions: Linum austriacum (upper panel, red), Linum usitatissiumum (lower panel, blue)

Conclusions.

The results highlight the differences in phenol content between cell suspensions deriving from two flax species belonging to different clades.

Although only preliminary, these data clearly show that cell cultures from the two species investigated produce different amounts of aromatic compounds and that the elicitation treatment enhance the level of these molecules. NMR analyses reveal that the two species produce different compounds belonging to lignan class. Moreover, it seems that the two species are able to secrete part of the produced metabolites into the medium; in particular, the higher concentration of secreted metabolites was found in *L. austriacum* medium.

These results suggest that the cell suspension technology and elicitation could represent a good system for bioactive compound production. These findings are particularly interesting since lignans have phytoestrogenic and cancer chemopreventive properties [5].

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