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Transregional Network for Innovation and Technology Transfer to Improve Health Care

Biochemical and immunochemical similarities among mammalian bilitranslocase and a plant flavonoid translocator

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Abstract — Flavonoids are a large class of plant secondary metabolites, belonging to polyphenol family, which possess pharmacological and nutritional properties. Their synthesis takes place only in plants, while mammals can acquire them only with diet. It has been demonstrated that flavonoid uptake occurs in rat also by the activity of bilitranslocase, a carrier that is involved in anion transport in liver cell, vascular endothelium and gastric mucosa. A sequence of bilitranslocase interacting with flavonoid moieties is already known and characterized. Antibody raised against such protein epitope were shown to exhibit cross-reactivity against plant membrane proteins in tissues involved in flavonoid transport and accumulation, such as teguments of carnation petals and skin of grape berries. Further immunolocalization studies allowed to demonstrate the presence of cross-reacting protein not only at the level of tegumental tissues, but also associated to sieve elements and seed teguments in grape berries.

Index Terms — flavonoid accumulation, plant bilitranslocase homologue, protein biochemical and structural characterization, secondary metabolite transport.

1 UNIVERSITY OF UDINE

It is a recent University at the heart of Europe. Established by popular demand, the University of Udine has its reference point at the center of a region that has historically been a meeting place and crossroad of different worlds and cultures. Since its origin, the University is strongly committed to the education of students

ready and able to face the challenges of a world, characterized by a global economy, that increasingly requires skills to be used at international level.

The internationalization represents the fourth mission of the University of Udine, alongside teaching, research and technology transfer. It grew through the constant increase and improvement of working relationships and partnerships with universities of Europe and other international institutions. This led, in particular, to the creation and offering of several graduate courses with international recognition of the certificate, masters, doctoral schools and European internships, as well as great development opportunities within the international mobility of students and teachers.

According to these aims, besides the traditional activities of higher education and scientific research, the University of Udine combines intensive transfer of innovative technologies and knowledge by serving the society and the world production and economics.

2 RESEARCH DEPARTMENT (UNIT)

The research unit consists of teachers/researchers involved in educational and research activities in Plant Biology. The most relevant field concerns some biochemical and plant physiology subjects focused on cell transport, mitochondrial respiration also in relation to oxidative stress and PCD manifestation. Approaches imply both membrane fraction as well as plant cell culture, grape in particular. The main task of such researches points to the acquisition of information about some fundamental metabolic plant process, as well as their role during stress responses. The second field concerns taxonomical studies on higher plants and geobotany, as well as algal biology and ecology, with the aim to study both species diffusion and their dynamics in natural and anthropic environments.

3 ACTIVITIES AND ROLES IN TRANS2CARE PROJECT

Several components of the grape berry have a straight influence on wine quality. Among these, polyphenols and other secondary metabolites are the major compounds affecting taste and flavour. Polyphenols are currently under a lively attention by researchers, particularly flavonoids, because they are relevant to grape and wine quality as well as for their pharmacological properties protecting human tissues against activated oxygen species (AOS), thus reducing cardiovascular and cancer risk [1]. These compounds, particularly anthocyanins and proanthocyanidins, are synthesized through the biosynthetic pathway of flavonoids [2].

There are several experimental evidences about the existence of a mammalian protein, named bilitranslocase (BTL), capable of a secondary active transport. This carrier transports not only plasmatic catabolites, but also anthocyanins [3,4]. In a previous work [5], it was reported that antibodies raised against mammalian BTL epitopes do cross-react with proteins obtained from a microsomal fraction of carnation petals (*Dianthus caryophyllus* L.). Moreover, some kinetic studies showed remarkable

similarities between bilitranslocase-mediated transport activity in hepatocytes and gastric epithelial cells and the plant carrier, although the latter has shown a different pattern of affinity for the substrates tested. Preliminary data, obtained in previous studies [6,7], have shown that a cross-reaction also occurs between antibodies raised against mammalian BTL and the microsomal fraction from grape berries. By means of immunochemical techniques, the presence of a translocator in white and red grape thin sections has been demonstrated. Carrier localization together with its expression profile, in different developmental stages, have been evidenced in both skin and pulp tissues. Therefore, it is relevant to verify, in *Vitis vinifera* berries of red cultivars, if proteins similar to the mammalian BTL mediate the accumulation of polyphenols, especially anthocyanins and tannins, and which effect on transport activity could be exerted by some environmental factors such as water deprivation.

4 CONCLUSIONS

In this framework the purposes of the project could be widespread in several topics. First, the study of the phylogenetic links between mammalian bilitranslocase and similar plant proteins.

Second the transport mechanisms regarding secondary metabolites, such as flavonoid and chlorophyll degradation products (biline-like compounds) would be characterized. Third, investigation on metabolic and environmental factors able to increase flavonoid production in plant cell cultures, would be also necessary.

Fourth, research on plants, as a model living organisms, will represent an advantageous alternative to animal use, ethically acceptable in biological studies.

To obtain these goals, it is essential to establish a wide network among different partners able to share their specific skills and knowledge, in agreement with the main task of Trans2Care project.

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