In contemporary analytical chemistry, more and more interest in biologically active substances, especially isolation from plants increases. Most of these are secondary metabolites, which play an important functions in the interaction of plants with the external environment. Many of them exhibit show biological activity, therefore they can have a positive effect on human health. In recent years, products of natural origin focus more and more attention, due to the chemical diversity of the isolated compounds. A large group of these compounds are substances with antioxidant activity. Antioxidants are the compounds which have the ability to neutralize free radicals, to damage the body's cells, thus increasing the risk of many diseases and accelerate the aging process. The occurrence of low concentrations of bioactive compounds in plants forces on the modern science the development of effective extraction methods.

In recent years, supercritical fluid extraction (SFE) has become an alternative to conventional solvent extraction methods to isolate valuable substances, mainly through the use of solvents generally regarded as safe (GRAS). The most commonly used is carbon dioxide, which is the ideal solvent for the isolation of natural products because it is non-toxic, cheap and easy to remove from the extracted products. Due to the fact that carbon dioxide is non-polar, the addition of a small amount of polar solvent allow for the extraction of polar compounds, e.g. polyphenols and flavonoids. A key role in using supercritical carbon dioxide extraction has an appropriate selection of extraction parameters. The solubility of the isolated compounds has the greatest impact on the efficiency of the extraction process. However, the mechanism which determine the efficiency of extraction is the transport of the mass of the substance extracted from the interior of the plant and then from the contact surface of the phases to the solvent. An important role in the extraction process is the resistance of mass transfer associated with the structure of the raw material and the specific location of the extracted compounds. An interesting approach in the crossing of this barrier is the use of enzymes degrading the cell wall of plants. The destruction of cell wall integrity by the use of different enzymes aims to increase the isolation of bioactive plant components and thus improve the extraction efficiency.

Therefore, the aim of our project will be the development of new methods of isolation natural of plant-based non-nutritive substances with a high concentration of antioxidant compounds. Opposite it challenge is using of the supercritical CO_2 extraction combination with flash chromatography, which is an innovative approach to improving the selectivity of extraction. In order to optimize the extraction conditions, a statistical response surface method will be used. The stage of characterization of the isolated extracts and their fractions will contain the use of chromatographic techniques, including supercritical chromatography (SFC), which is a "greener and safer" alternative to conventional preparative chromatography. What's more, the use of plant cell wall degrading enzymes will facilitate the extraction of bioactive compounds, so that the isolated compounds will be released more easily, which significantly improve the extraction efficiency.

Plant extracts currently available on the cosmetic or medical market differ in the content of active substances and thus in the quality. Today, in the pursuit of profits the companies producing plant extracts often overlook the quality of their products. Nevertheless, quality problems seem to outweigh the potential health benefits of plant extracts, and the main cause of these problems seems to be the lack of reliable analytical techniques and methodologies for chemical analysis of plant materials. Results obtained from the implementation of this project may play a key role in obtaining valuable products of high biological activity and potential application in pharmaceutical and cosmetic industry.