### **Stage I/2007**

To be executed by 15.12.2007;

Stage value: 150,000 lei of which 30,000 lei from Co-funding

Documents: intermediate report on the activity;

## Objective of stage I/2007

Studies on the preliminary phytochemical characterisation of some medicinal plants with prophylactic properties in cardiovascular conditions

## 1<sup>st</sup> Stage Activities

**Activity 1.1** Study on some medicinal plants and classes of active principles acting as antioxidants, vasoprotector, reducing cholesterol and lipid concentrations;

Activity 1.2 Phytochemical screening of Avena sativa and Cynara scolymus species;

**Activity 1.3** Drafting technical specifications for plant products.

#### **Abstract**

Cardiovascular diseases are conditions of the heart and blood vessels such as atherosclerosis, ischemic cardiopathy (pectoral angina, myocardial infarction), and chronic venous deficiency.

The main risk factors are represented by an increase in the blood pressure (i.e. hypertension) and oxidative stress.

A large number of ingredients or natural components of our food may have either a positive or a negative impact on various markers of cardiovascular conditions. Such components are represented by lipids or fats, a series of which are found in food, mainly the triacylglycerols (TAG) and phorpholipids; sterols and stanols in plants which lower total cholesterol and LDL and reduce the risk of coronary condition onset as well; B-group vitamins (B9, B6, B12), vitamins E, A, and, which can be associated to reducing the risk of cardiovascular conditions as well owing mainly to their antioxidant potential; simple phenols, flavonoids, lignin, and condensed tannins; beta-glucans, soluble fibres that act by reducing or delaying absorption in increasing viscosity and lowering the rate of stomach discharge. The outcome is represented by a low level of LDL cholesterol, the after-meal level of glucose and the reduced insulin response; proteins, peptides.

The phytotherapy applied in cardiovascular conditions focus on more directions such as phytotherapy in ischemic cardiopathy (dilatation of coronaries, antispastic, sedatives, diuretics, lowing blood pressure), phytotherapy in myocardial infarction (analgesic, antispastic, anxiolytic (sedative), anticoagulant, stimulator of collagen biosynthesis, suppressors of collagen degradation), phytotherapy cardiac deficiency (cardiotonic heteroside, active principles with diuretic action), phytotherapy in high blood pressure (indolic alkaloids, vincamines, viscotoxins, sedatives, diuretics), anti-arrhythmia phytotherapy (cardiotonic heteroside, alkaloids, pro-anthocians), phytotherapy in low blood pressure (ephedrine, purine bases, camphor); phytotherapy in cerebral and peripheral vascular, phytotherapy in venous and lymphatic system conditions (flavonoids, coumarin derivatives and saponins).

Plant species that can be used in the phytotherapy of cardiovascular conditions are represented by *Cynarae folium, Avena sativa herba / fructus, Sorbus aucupariae fructus, Allii ursini herba.* 

Leaves of *Cynara scolymus* contain caffeoil - quinine acids, flavonoids and flavones derivatives, sesquiterpene lactones, sterols, tannin, mucilage, pectins, amino acids, carbohydrates, organic acids, caffeic acid, enzymes, mucilage, pectins and lab enzymes. Extracts of *Cynarae folium* have lowering cholesterol and lipids and antiatherogenic properties.

The fact that *Avena sativa* contains some active principles with antioxidant potential (e.g. polyphenols, E vitamin), as well as carbohydrate indigestible polymers justifies the proposition to value both its caryopses and the aerial part with a view to obtain plant preparations that are efficient in cardiovascular conditions prophylaxis.

The high concentration of some antioxidant active principles makes the fruit of *Sorbus aucuparia* represent an important raw matter with a view to obtaining efficient plant preparations in cardiovascular conditions prophylaxis.

The composition of *Allii ursini herba* is similar to the garlic's. Allicin induces dilatation of vessels; ajoene and allicin reduce cholesterol synthesis by 20-70%.

The plant products selected underwent phytochemical analysis in order to determine the flavonoids and polyphenol concentrations in *Cynarae folium* and *Avena sativa fructus*.

In order to identify the major groups of active principle successive extractions of plant matter s were performed with various polarity solvents (e.g. (dichloromethane, ethanol, water). By means of dichloromethane, lipid soluble substances were extracted from the plant matter. The ethanol extracted active principles such flavonosides, pentacyclic triterpene saponosides, amino acids. Macromolecular active principles (e.g. polyholosides and proteins) were extracted in water as well as small molecule substances, such as oses, flavonosides, triterpene saponosides and some amino acids.

With a view to acquiring a preliminary orientation on the chemical composition of the two plant species a chromatographic study was conducted aiming at determining the active principle classes that are representative for the phytochemical study: flavonoids compounds, polyphenol carboxylic acids / revealing their antioxidant activity; amino acids. Moreover, extractive solutions of *Cynara scolymus* aerial parts, e.g. ethylic alcohol 50% v/v and water, and of *Avena sativa* caryopses were scanned within UV/VIS (190-850) nm domain.

The plant products used for the phytochemical study were quantitatively evaluated for the active principle classes likely to be responsible for the pharmaceutical action under study: *flavonoids*, *polyphenols*, *tannins*, *amino acids*, *micro and macro elements*. Moreover, the antioxidant activity was determined.

#### **CONCLUSIONS**

- 1. In order to identify the major groups of active principles, successive extractions were conducted with various polarity solvents, e.g. dichloromethane, ethanol, water.
  - the qualitative chemical analysis of the extracts obtained from the artichoke leaves and oat caryopses lead to the identification of the active principles groups: lipid soluble substances, water soluble potassium salts of some substances such as fat acids, pentacyclic triterpene acids, flavonosides, amino acids, flavone aglycons, polyholosides and proteins, small molecule substances such as oses.

- ethanol extract in artichoke leaves are noticeable owing to the rich concentrations of polyphenol substances, i.e. polyphenol carboxylic acids, flavonosides) whereas the aqueous extract in the oat caryopses contain polyholosides, reducing compounds and amino acids.
- 2. With a view to acquiring a preliminary orientation on the chemical composition of the two plant species a chromatographic study was conducted aiming at determining flavonoid compounds, polyphenol carboxylic acids / revealing their antioxidant activity and amino acids.

Moreover, extractive solutions of *Cynara scolymus* aerial parts, e.g. ethylic alcohol 50% v/v and water, and of *Avena sativa* caryopses were scanned within UV/VIS (190-850) nm range.

- hence, dark-grey spots displayed on the chromatographic plate viewed at 254 nm before spraying with the identifying reactive, reveals the presence of flavonoid compounds; blue spots displayed on the chromatographic plate viewed at 366 nm before spraying with the identifying reactive reveals the presence of polyphenol carboxylic acids; following viewing at 366 nm, after spraying with the identifying reactive, the presence of flavonoid compounds and polyphenol carboxylic acids was recorded. The spots characteristic to flavonoid compounds and polyphenol carboxylic acids that were identified in the artichoke and oat samples undergo a discolouration as a result of in DPPH solution spraying, hence indicating the presence of antioxidant activity.
- the presence of brick-tinted spots indicated the presence of amino acids.
- **3.** The extractive solutions e.g. hydroalcoholic and aqueous) of *Cynarae folium* and *Avena sativa fructus* were scanned within the UV/VIS (190-850) nm range. The analysis of UV/VIS absorption spectre diagrams of the hydroalcoholic and aqueous extracts of *Cynarae folium* and *Avena sativa fructus* revealed the presence of some *chemical structures shared* by the two extract types as well as some *chemical structures specific* to each type of extract within the UV and visible ranges.
  - **4.** The plant products used for the phytochemical study were quantitatively evaluated for the active principle classes likely to be responsible for the pharmacological action under research: *flavonoids*, *polyphenols*, *tannins*, *amino acids*, *micro and macro elements*.
    - for both plant extract, records prove that the highest total flavone concentration is to be founding the methanolic extract;
    - total polyphenols expressed in caffeic acid were determined via two methods: with Arnow reactive and with Folin one. The latter method allowed us to obtain the highest value for total polyphenols in both plant products.
    - as to artichoke leaves (*Cynarae folium*), out of the phenol compound analysed (flavones, polyphenols, tannins), the highest value was recorded by total flavones expressed in rutoside. Total amino acids expressed in glutamic acid records the highest value in aqueous extract.

- as to oat caryopses, the 50% EtOH hydroalcoholic extract records the highest amino acid concentration expressed in glutamic acid and in total fructosans expressed in fructose.
- With respect to the content of micro and macro elements, *Cynarae folium* displays higher values that those recorded in *Avena sativa fructus*, except for manganese.
- **5.** The antioxidant action of *Cynarae folium* samples expressed in the suppressing capacity of the DPPH free radical records the highest value in maximum concentration of total polyphenols expressed in caffeic acid, of plant product respectively.

To conclude, the flavonoid and total polyphenol content in *Cynarae folium* as well as the polysaccharide and amino acids content cum in *Avena sativa fructus* justifies the use of these products in order to obtain antioxidant, vasoprotector, and cholesterollowering preparations that can be used in cardiovascular condition prophylaxis.

# **Description/presentation of various events carried out within the project** Editing and publishing a book specialized in the field:

**Miron, A.** Plantele medicinale în profilaxia și tratamentul unor afecțiuni cardiovasculare, Editura «Gr. T. Popa», Iași, 2007, ISBN 978-973-7682-31-4. [Title in English: Medicinal Plants in Some Cardiovascular Condition Prophylaxis and Treatment]