

**UNIVERSITY OF MEDICINE AND PHARMACY OF TÎRGU
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DOCTORAL SCHOOL

Abstract of the PhD thesis

**Research regarding the phytochemistry of the red wine
and its biological effects**

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The wine, a part of the human history, has been considered a medicinal product since ancient times. However, its beneficial effects on the human body, even if proven in some scientific studies are still a challenge for the researchers. The effects and the mechanism of action of the red wine in certain pathological conditions are not fully understood.

The antioxidant property (spin trapping effect) of the red wine is considered the main biological activity and is linked to the high level of polyphenol compounds. This claim can be proved by measuring several markers of the oxidative stress in the biological fluids. We can consider endogenous molecules as markers of the oxidative stress able to trap free radicals or molecules that results after the interaction of several free radicals with biological compounds, a process that leads to destruction of the biological compounds and formation of measurable degradation products. Most of the time the reactions induced by the reactive oxygen species (ROS) are monitored.

The introductive part of the thesis describes the recent knowledge in this field and is used as a starting point for further research. In this part detailed information about the oxidative stress, its biomarkers, defense mechanisms of the organism against free radicals, kinetic and pharmacologic data about polyphenols are presented.

Four large experiments represent my original contribution to this field of knowledge.

The first part contains several analytical methods: HPLC-DAD/ESI-MS and HPLC-DAD/APCI(-)-MS usable for measuring flavonoid and non-flavonoid

compounds in several types of wines; Folin-Ciocalteu method for measuring the total phenolic compounds; the pH-differential method for measuring monomeric antocyanins; GC-FID method for measuring the volatile compounds present in wine and the Q-ICP-MS method for the metal content measurement. These methods can be successfully used for obtaining data about the quality of the wines and eventual counterfeiting procedures.

The beneficial effects of the red wines were measured *in vitro* using the DPPH free radical scavenging method, the ORAC assay (oxygen radical absorbance capacity), chemiluminescence technique (Photochem) and the ability to reduce the activity of the angiotensin-converting enzyme (ECA).

Results showed that there is a positive correlation between the antioxidant activity (speed rate of DPPH neutralization) of the red wine and its total polyphenol content. The most active polyphenols in the reaction with the peroxy free radical were: resveratrol, p-coumaric acid and myricetin. The most active polyphenols in the ECA inhibition reaction were: p-coumaric acid, myricetin and quercetin.

The third objective of this work was the development and validation of analytical methods for *in vivo* measurement of reduced (GSH) and oxidized (GSSG) glutathione and plasma malondialdehyde levels as oxidative stress markers.

In vivo experiments using positive and negative blanks (red wine, red wine without alcohol, resveratrol, 13 % hydroalcoholic solution, antocyanins) proved the positive effects of red wine or its constituents against the oxidative stress during our experiment.

The **innovative ideas** of the research presented in this thesis can be summarized as follows: measurement of DPPH free radical scavenging reaction rate constant, measurement of ECA inhibition ability using the principal compound analyze in obtaining data about the antioxidant ability of the red wine and its constituents. Obtaining new analytical methods for assessment of oxidative stress or significant improvement of the existing ones (malondialdehyde and glutathione), reduction of analysis time, sample size reduction and especially the increase of sensitivity can also be mentioned.

Key words: oxidative stress, red wine, HPLC-MS, Folin-Ciocalteu, GC-MS, Q-ICP-MS, GS-FID, DPPH, ORAC, Photochem, ECA, malondyaldehyde, glutathion, rat experimental models.