

## THE INFLUENCE OF ULTRAVIOLET UV B RADIATIONS ON THE PROPOLIS AND POLLEN COLLECTED FROM DIFFERENT GEOGRAPHIC REGIONS FROM THE WESTERN PART OF THE COUNTRY

Orodan Maria<sup>\*</sup>, Paul Atyim<sup>\*</sup>, Glisici Mihaela<sup>\*</sup>, Toderescu Corina Dalia<sup>\*\*</sup>, Negrei Carolina<sup>\*\*\*</sup>, Morgovan Claudiu<sup>\*</sup>, Pallag Annamaria,<sup>\*\*\*\*</sup> Osser Gyongyi<sup>\*</sup>

<sup>\*</sup>“Vasile Goldiș” Western University of Arad, Romania, Faculty of Medicine, Pharmacy and Dentistry, Address: 31, Henri Coandă, Arad, 310429, Romania, e-mail: [gyongyiosser@ymail.com](mailto:gyongyiosser@ymail.com)

<sup>\*\*</sup> “Carol Davila” University of Medicine and Pharmacy, Department of Pharmaceutical Technology and Biopharmacy, 6, Traian Vuia Street, Bucharest, Romania

<sup>\*\*\*</sup> “Carol Davila” University of Medicine and Pharmacy, Department of Toxicology, 6, Traian Vuia Street, Bucharest, Romania

<sup>\*\*\*\*</sup> University of Oradea, Faculty of Medicine and Pharmacy, Department of Pharmacy, 29, N. Jiga, Oradea, Romania

### Abstract

*The paper shows the test results of the propolis and pollen samples which have been collected as raw material by the beekeepers from various regions of western Romania. The samples were extracted with ethanol and the final samples were analyzed with the HPLC method.*

*Results lead to the conclusion that the enzyme activity of the propolis and pollen samples is modified under the effect of the ultraviolet UV-B type radiations on the plants, as well as their influence on the apiarian products resulted from the beekeepers from these geographical areas, in comparison with the investigated biological material.*

**Key words:** propolis, pollen, HPLC method

### INTRODUCTION

Propolis and pollen samples have been collected as raw material by the beekeepers from various parts of western Romania. The samples were extracted with ethanol, and the final samples were analyzed with the HPLC method.

Propolis is considered as being a traditional natural medicine that is gaining more and more popularity with alternative therapies. Propolis is also called “bee clay” or “Romanian penicillin” (Ambasht et al, 1998).

Propolis is a resin-like material from the buds of poplar and cone-bearing trees.

Propolis has a long history of medicinal use, dating back to 350 B.C., the time of Aristotle.

Greeks have used propolis for abscesses; Assyrians have used it for healing wounds and tumors; and Egyptians have used it for mummification (Neacșu, 2002). It still has many medicinal uses today, although its

effectiveness has only been shown for a couple of them (Srawhi et al, 2012, Rajendiran et al, 2004, Markham et al, 1996, Simionocivi et al, 1983).

Propolis is collected from tree sap and various other plants by honeybees. It contains a wide range of vitamins and minerals and is believed to be an antioxidant as well. Propolis may also be useful as an antibiotic and antifungal supplement with several potential health benefits (Villanueva et al, 2002).

Many people ingest propolis tincture, a mixture of propolis extract and food-grade alcohol, because they believe it can ward off a number of illnesses from the common cold to cancer. The tincture can also be applied topically as a means to treat cuts, acne, and even scars (Marcucci, 1995, Strid et al, 1994, Strid et al 1992). The pollen is composed of a multitude of microscopic corpuscles, contained in the pollen sacs of the flower's stamens' arteries (Ahn et al, 2007).

Propolis supports the immune system in various ways. First, the antimicrobial properties suppress harmful bacteria and infections. Further, it actually stimulates the immune system and raises the body's natural resistance (Ambasht, Agrawal, 1998)

The effect of the UV ultraviolet type radiations that influence the growth and development of the plants from different geographical regions has been investigated, so that the bees harvest pure and ecological products with a high content of vitamins (Bungău et al, 2011) and minerals (Crocì et al, 2010, Bungău, 2015, Bungău et al, 2011, Haro et al, 2000, Joean, 1997, Torabinejad et al, 1998).

We wanted to highlight the effect of the UV B ultraviolet type radiations on the plants, as well as their influence on the apiarian products resulted from the beekeepers from these geographical areas (Băra et al, 2003).

The technical details are described in the article and the results conclude that the samples from the high mountainous area show the highest concentration of ascorbic acid and therefore have the best antioxidant effect (Alscher et al, 2002).

## **MATERIALS AND METHODS**

Propolis usually contains a variety of chemical compounds, such as: polyphenols (flavonoids, phenolic acids and their esters), terpenoids, steroids and amino acids. The composition of the propolis depends on the vegetation from the collection areas

The bee pollen contains the following substances: vitamins (provitamin A, vitamin A, vitamin B1, B2, B3, B5, B6, B12, vitamin C, D, E, F, H, K, PP and folic acid), minerals (calcium, phosphorus, potassium,

iron, copper, iodine, zinc, sulfur, sodium, chlorine, magnesium, molybdenum, selenium, bromine, silicium and titanium), amino acids, carbohydrates, fatty acids, enzymes, co-enzymes and various fats, minerals necessary to the body, even those minerals that are not usually found in dietary supplements sold in pharmacies (Antofie, 2010, Bungău, 2015, Bungău et al, 2011, Bungău et al, 2011, Bungău et al, 2003).

The pollen and propolis samples were collected as raw materials by the beekeepers from: Arieșeni, Gurahonț, Miniș and Arad.

The raw propolis was extracted with ethanol at a temperature of 65°C, during 24 hours, by default methods (Apel et al, 2004).

The propolis must be ground well, the pollen must be put each in a mortar (to increase the contact surface with the alcohol and to aid the extraction process) and ethylic alcohol must be added of 80-90°, 3 parts propolis (i.e. pollen), 10 parts of ethylic alcohol must be added. These substances were left in the water bath at a temperature of 65 degrees, for 24 hours (Băra et al, 2006, Băra et al, 2003).

The ethanol suspension was separated by centrifugation for 10 minutes at 1000 rpm, after which the obtained solution was vacuum filtered and analyzed on the HPLC.

## RESULTS AND DISSCUSIONS

One sample of pollen and propolis, from each of the three different regions of Romania, were analyzed through the HPLC method (Chen et al, 2004), to highlight the ascorbic acid content. The obtained results can be found in Table 1.

Table 1.

Results obtained from the products collected from different areas			
<i>The mountainous area (Gurahonț, Arieșeni)</i>			
Samples	From the device [µg/L]	By mass of the sample [µg/kg]	Final result [mg/kg]
pollen	1122.524	149669.9	149.6699
propolis	1093.34	145778.7	145.7787
<i>Hillside area (Miniș)</i>			
pollen	1112.01	149336.2	149.3362
propolis	1022.11	144885.6	144.8856
<i>Lowlands (Arad)</i>			
pollen	1001.01	148995.1	148.9951
propolis	1013.02	144793.3	144.7933

The effects of the ultraviolet UV radiations differ slightly, depending on the geographical region, due to the fact that at high altitudes (mountains) the ultraviolet radiations reaching the plants are smaller thus increasing the

ascorbic acid concentration for the pollen and propolis; a decrease in the ascorbic acid concentration, regarding the pollen and propolis collected by the bees was also observed.

It can be noticed in the case of the samples collected from the hillside region, that the UV radiations have an increased effect on the plants; an even greater decrease in the ascorbic acid concentration can be noticed when analyzed samples were collected from the lowlands region, where the ultraviolet radiations have very intense effect on the plants. The color indicates its vegetal origin or its specific geographical area.

## CONCLUSIONS

The physiological and developmental processes of the plants are affected by the UVB radiations, even by the current UVB radiation levels.

Despite the mechanisms to reduce or mitigate these effects and a limited ability to adapt to a high level of UVB radiations, the plants' development may be directly affected (Bogdanov, 2006).

High quality pollen and propolis must not contain impurities, must not be collected from areas polluted with pesticides, insecticides, fertilizers, industrial pollutants, areas with high ultraviolet radiations, must not be contaminated radioactively, must not come from a hive where there have been used synthetic drugs or antibiotics (Brunchon-Harti et al, 1994).

The pollen and propolis, due to the substances present in their composition, exhibit a wide biological activity, although not very high, and in order to obtain a highly therapeutic result, we must have quality products.

The effects of the ultraviolet radiations differ slightly, depending on the geographical region (Caldwell et al, 1989, Beggs et al, 1985), the values obtained from the measurements that were performed revealed that the samples collected from the highest area, namely the mountainous region Gurahonț – Arieșeni has the highest content of ascorbic acid, respectively the best antioxidant activity.

## REFERENCES

1. Ahn, M. R., Kumazawa, S., Usui, Y., Nakamura, J., Matsuka, M., Zhu, F. And Nakayama T., 2007, Antioxidant activity and constituents of propolis collected in various areas of China, Food Chemistry, 101, pp. 1383-1392
2. Alscher, R. G., Erturk, N., Heath, L. S., 2002, Role of superoxide dismutases (SODs) in controlling oxidative stress in plants, J. Exp. Biol.; 531(1), pp. 1331–1341
3. Ambasht, N.K., Agrawal, M., 1998, Physiological and biochemical responses of *Sorghum vulgare* plants to supplemental ultraviolet-B radiation, Can. J. Bot. 76, pp.1290–1294
4. Antofie M., Constantinovici, M.D., Pop, M.R., Iagaru, P., Sand, C., Cirotea, G., 2010, Theoretical methodology for assessing the status of conservation of crop landraces in

- Romania, Analele Universității din Oradea, Fascicula Biologie, TOM XVII, Issue2, pp.313-317
5. Apel, K., Hirt, H., 2004, Reactive oxygen species: metabolism, oxidative stress, and signal transduction, *Annu Rev Plant Biol* 55, pp. 373–399
  6. Băra C. I., Crețu R. M., (2006). Biochemical effects induced by UV treatment on 5 romanian *Phaseolus vulgaris* L. cultivars, grown in field, Analele Științifice ale Universității “Al.I.Cuza” din Iași, Secțiunea I, a.Genetică și Biologie Moleculară, tom VII, p.145-150
  7. Băra, C., Artenie, V, Băra, I., 2003, Effects of UV-B radiation in higher plants, Analele Științifice ale Universității “Al.I.Cuza” din Iași, Secțiunea I, a.Genetică și Biologie Moleculară.
  8. Beggs, C.J., Wellmann, E., 1985, Analysis of light controlled anthocyanin formation in coleoptiles of *Zea mays*, the role of UV-B blue, red and far red light, *Photochem. Photobiol.*41, pp. 481-486.
  9. Bogdanov, S., 2006, Contaminants of bee products. *Apiology* 38: p. 1–18.
  10. Brunschön-Harti S., Fangmeier A., Jäger H., J., 1994, Effects of ethylendurea and ozone on the antioxidative system in beans *Phaseolus vulgaris*, *Environmental Pollution* vol. 90, pp. 95-103.
  11. Bungău, S., 2014, Vitamins and amino acids determination using kinetic methods, *Colecția Science&Technology*, Ed. Italian Academic Publishing, București
  12. Bungău S., Fodor, A., Szabo, I., Muțiu, G., 2011, Comparative studies on *Petroselinum crispum* folium ascorbic acid content using kinetic, spectrophotometric and iodometric methods, *Archives of the Balkan Medical Union*, 46(1), pp. 77-80
  13. Bungău, S., Fodor, A., Țiț, D. M., Szabó, I., 2011, Studies on citrus species fruits ascorbic acid content using kinetic, spectrophotometric and iodometric methods, *Analele Universității din Oradea, Fascicula Protecția Mediului*, Vol. XVI/A, Anul 16, ISSN 1224-6255, pp.212-217
  14. Bungău, S., Bâldea, I., Copolovici, L., 2003, Determination of ascorbic acid in fruit using a Landolt type method, *Revista de Chimie*, 54 (3), pp. 213-216
  15. Caldwell, M. M., Teramura, A. H., Tevini, M., 1989, The changing solar ultraviolet climate and the ecological consequences for higher plants. *Trends in Ecology and Evolution* 4: pp. 363-367
  16. Chen M., Chory J., Fankhauser Ch., 2004, Light signal transduction in higher plants, *An. Rev. Genet.* 38, pp. 87–117
  17. Croci, A. N., Cioroiu, B., Lazar, D., Corciova, A., Ivanescu, B., Lazar, M. I., 2010, HPLC evaluation of Phenolic and polyphenolic acids from propolis. *Pharmacy press*.
  18. Haro, A; López-Aliaga, I; Lisbona, F; Barrionuevo, M; Alférez, M J; Campos, M S., 2000, Beneficial effect of pollen and/or propolis on the metabolism of iron, calcium, phosphorus and magnesium in rats with nutritional ferropenic anemia, *Journal of Agriculture and Food Chemistry* 48: p. 5715–5722
  19. Joean D., 1997, Statistical analysis concepts and applications in pharmaceutical sciences, *the pharmacy*, p. 45(1), 55-63
  20. Marcucci, M. C., 1995, Propolis: chemical composition. biological properties and therapeutic activity, *Apiology*, p. 26: 83-99
  21. Markham, K. R., Mitchell, K. A., Wilkins, A. L., Daldy, J. A., Lu, Y., 1996, HPLC and GC-MS Identification of the major organic constituents in New Zealand propolis, *Phytochem*, p. 42: 205-211
  22. Neacșu C., 2002, *Apitherapy Compendium*, Technical Publishing House, Bucharest.
  23. Rajendiran, K., Ramanujam, M.,P., 2004, Improvement of biomass partitioning flowering and yield by triadimeton in UV-B stressed *Vigna radiata*(L), *Wilezek- Biol. Plant.* 48, pp.145-148.

24. Rawhi M.A.Abdalla, 2012, All about Apitherapy, All Publishing House, Bucharest, p.76-88, 94-111.
25. Simionovici M., Cârstea Al., Vlădescu C., 1983, Pharmacological research and drug prospecting, Medical Publishing House, Bucharest, p. 140, 228-231, 259-261, 414-429, 437-438
26. Singh S., Pankaj K., Ashwani K., 2006, Ultraviolet radiation stress: molecular and physiological adaptations in trees in A biotic stress tolerance in plants, Springer etherlands editer. 1007, pp. 4020-4389
27. Strid, Å., Chow, W., Anderson, J. M., 1994, UV-B damage and protection at the molecular level in plants, Photosynthesis Research 39: pp. 475-489.
28. Strid, Å., Porra, J. R., 1992, Alterations in pigment content in leaves of *Pisum sativum* after exposure to supplementary UV-B. Plant Cell Physiol. pp. 33-37.
29. Torabinejad, J. M., Caldwell, M., Flint, S.D., Urham, S. D., 1998, Susceptibility of pollen to UV-B radiation, An assay of 34 taxa. American Journal of Botany 85, pp. 360-369.
30. Villanueva, M T O; Marquina, A D; Serrano, R B; Abellán, G B., 2002 The importance of bee-collected pollen in the diet: a study of its composition, International Journal of Food Sciences and Nutrition 53(3): p. 217-224.