

- V. Bankova, S. Popov, N. Marekov; "HPLC analysis of flavonoids from propolis", *J. Chromatogr.* 1982, **242**, 135 - 143.

[https://doi.org/10.1016/S0021-9673\(00\)87255-6](https://doi.org/10.1016/S0021-9673(00)87255-6)

Abstract: A simple high-performance liquid chromatographic procedure using a reversed- phase column and an internal standard has been developed for the qualitative and quantitative analysis of flavonoids from the bee-hive product propolis. The method can be applied even to flavonoids that produce overlapping peaks

1. V. Nenov, N. Mollova, P. Demirev, V. Bankova, S. Popov; "Computer assisted mass-spectral investigation of flavonoid mixtures", *Int. J. Mass Spectrom. Ion Phys.*, 1983, **47**, 333 - 336. [https://doi.org/10.1016/0020-7381\(83\)87202-7](https://doi.org/10.1016/0020-7381(83)87202-7)

Abstract: A flavonoid mixture is investigated by means of direct introduction and the spectra are processed according to an established algorithm in order to achieve pure spectra.

2. V. Bankova, S. Popov, N. Marekov; "A study on flavonoids from propolis", *J. Nat. Prod.*, 1983, **46**, 471 - 474. <http://pubs.acs.org/doi/abs/10.1021/np50028a007?journalCode=jnprdf>

Abstract: Seven known propolis flavonoids have been isolated and identified. A selective elution, used in hplc analysis of propolis flavonoids have permitted the detection of three new flavonoids in this object. Structural assignments for two of them {3,7-dihydroxy-5-methoxyflavanone (**8**) and 2,5-dihydroxy-7-methoxyflavanon(**9**) } have been given.

3. N. Marekov, V. Bankova, S. Popov; "The practical value of the polyphenols taken from propolis", *IMPACT of Science on Society* 1984, **136**, 396 - 374. <http://unesdoc.unesco.org/images/0006/000625/062504eo.pdf#nameddest=62523>

6. N. Manolova, V. Maximova, G. Gegova, Y. Serkedjieva, S. Usunov, N. Marekov, V. Bankova; "On the antiinfluenza action of fractions from propolis", *Compt rend. de l'Acad. Bulg. Sci.* 1985, **38**, 735 - 738.

7. V. Bankova, Al. Dyulgerov, S. Popov, N. Marekov; "A GC/MS study of the propolis phenolic constituents"; *Z. Naturforsch.* 1987, **42c**, 147 - 151. <https://www.degruyter.com/view/j/znc.1987.42.issue-1-2/znc-1987-1-224/znc-1987-1-224.xml>

Abstract: Silylated phenolic fraction from two propolis samples was investigated by capillary GC/MS. Fifteen phenolic acids (seven of them new for propolis), two ketones, new for propolis, eight esters of phenolic acids, new for propolis, six flavanones and two flavonols, were tentatively detected.

8. H. Neychev, V. Dimov, V. Vuleva, L. Shirova, E. Slavcheva, G. Gegova, N. Manolova, V. Bankova; "Immunomodulatory action of propolis. II. Effect of water soluble fraction on influenza infection in mice", *Acta Microbiologica Bulgarica* 1988, **23**, 58 - 62.

9. V. Bankova, S. Popov, N. Marekov; "Isopentenyl cinnamates from poplar buds and propolis", *Phytochemistry* 1989, **28**, 871 - 873.
[https://doi.org/10.1016/0031-9422\(89\)80133-5](https://doi.org/10.1016/0031-9422(89)80133-5)

Abstract: From *Populus nigra* buds, *P. italica* buds and propolis, two esters of caffeic acid and two esters of ferulic acid with isomeric pentenyl alcohols have been isolated and their structures elucidated.

10. V. Bankova; "Synthesis of natural esters of substituted cinnamic acids", *J. Nat. Prod.* 1990, **53**, 821 - 824.

<http://pubs.acs.org/doi/abs/10.1021/np50070a007?journalCode=jnprdf>

Abstract: Natural esters of substituted cinnamic acids, found in propolis and poplar buds, have been synthesized by the Wittig reaction from (carbalkoxymethyl)-triphenylphosphonium halides and unprotected phenolic aldehydes in solid-liquid heterogeneous medium under sonochemical conditions. The synthetic products confirm the previously described structures of the natural products and allow testing of their biological activities.

11. V. Dimov, N. Ivanovska, N. Manolova, V. Bankova, N. Nikolova, S. Popov; "Immunomodulatory activity of propolis. Influence of anti-infectious protection and macrophage function", *Apidologie* 1991, **22**, 155 - 162.

https://www.apidologie.org/articles/apido/pdf/1991/02/Apidologie_0044-8435_1991_22_2_ART0008.pdf

Abstract: The immunomodulatory action of a water-soluble derivative (WSD) of natural propolis was investigated. The oral and parenteral administration of the WSD enhanced the survival rate and the mean survival time in experimental bacterial (*Klebsiella pneumoniae*, *Staphylococcus aureus*) and fungal (*Candida albicans*) infections in mice. An increased resistance was observed also in *Klebsiella pneumoniae* infection induced after cyclophosphamide treatment. The WSD stimulated peritoneal macrophages to produce in vitro interleukin-1, which

corresponded to their elevated total protein secretion. In addition, WSD failed to trigger lymphocyte proliferation as determined by popliteal lymph node assay. The WSD was suggested to augment non-specific host defense via macrophage activation.

12. V. Bankova, Al. Dyulgerov, S. Popov, L. Evstatieva, L. Kuleva; "A study on the origin of Bulgarian propolis", *Apiacta* 1991, **26**, 13 - 17.

http://www.fiitea.org/cgi-bin/index.cgi?sid=&zone=download&action=download_file&file_id=696&ateg_id=182

13. V. Bankova, A. Dyulgerov, S. Popov, L. Evstatieva, L. Kuleva, O. Pureb, Z. Zamjansan; "Propolis produced in Bulgaria and Mongolia: phenolic compounds and plant origin", *Apidologie* 1992, **23**, 79 - 85.

https://www.apidologie.org/articles/apido/pdf/1992/01/Apidologie_0044-8435_1992_23_1_ART0009.pdf

Abstract: Phenolic composition of Bulgarian and Mongolian propolis and their possible plant sources (the resinous secretions of different tree buds) were investigated by gas chromatography-massspectrometry (GC/MS). The results obtained demonstrate that in both countries propolis is collected mainly from poplars: from *Populus nigra* and to some extent from *P italica* in Bulgaria, and from *P suaveolens* in Mongolia.

14. J. Serkedjieva, N. Manolova, V. Bankova; "Anti-influenza virus effect of some propolis constituents and their analogues (esters of substituted cinnamic acids)", *J. Nat. Prod.* 1992, **55**, 294 - 297.

<http://pubs.acs.org/doi/abs/10.1021/np50081a003?journalCode=jnprdf>

Abstract: The antiviral activity of six synthetic substances, esters of substituted cinnamic acids, identical with or analogous to some of the constituents of the Et20 fraction of propolis was studied in vitro. One of them, isopentyl ferulate, inhibited significantly the infectious activity of influenza virus NHong Kong (H3N2) in vitro and the production of hemagglutinins in ovo. By the use of diverse experimental patterns, it was found that the maximal inhibition of viral reproduction was observed when test substances were present in the medium during the whole infectious process.

15. V. Dimov, N. Ivanovska, V. Bankova, S. Popov; "Immunomodulatory action of propolis. IV. Prophylactic activity against Gram-negative infections and adjuvant effect of the water-soluble derivative", *Vaccine* 1992, **10**, 817 - 823.

<http://www.sciencedirect.com/science/article/pii/0264410X9290043J>

Abstract: The efficacy of the water-soluble derivative (WSD) of natural propolis (bee glue) was examined for augmentation of host resistance against experimental infections caused by Gram-negative pathogens (*Klebsiella pneumoniae*, *Proteus vulgaris*, *Escherichia coli*, *Pseudomonas aeruginosa*). The substance was found to induce significant non-specific protection, but did not inhibit the in vitro growth of the same strains. Pretreatment with WSD prior to the standard scheme for tumour necrosis factor (TNF) induction (BCG and two weeks later lipopolysaccharide (LPS)) provoked an interval-dependent reduction in the lytic capacity of serum against L 929 target cells. The replacement of the triggering or priming signal with WSD markedly increased TNF production. In vivo administration of WSD led to a rapid and route-dependent change in the alternative complement pathway haemolysis. The alteration in C1q complement component and total protein synthesis, and also in nitroblue tetrazolium reduction, suggests that macrophage activation makes a major contribution to the capacity of WSD to prevent infections.

16. V. Bankova, R. Christov, G. Stoev, S. Popov; "Determination of phenolics from propolis by capillary gas chromatography", *J. Chromatogr.* 1992, **607**, 150 – 153.

<http://www.sciencedirect.com/science/article/pii/0021967392870671>

Abstract: A procedure using capillary gas chromatography with an internal standard has been developed for the determination of the main biologically active phenolics of propolis (bee glue): the flavonoid aglycones pinocembrin and galangin, and caffeic acid and its β -phenylethyl ester.

17. R. Christov, V. Bankova; "Gas chromatographic analysis of underivatized phenolic constituents from propolis using an electron capture detector", *J. Chromatogr.* 1992, **623**, 182 - 185.

<http://www.sciencedirect.com/science/article/pii/0021967392870671>

Abstract: Underivatized phenolic constituents from propolis (flavonoid aglycones, phenolic acids and their esters) were analysed by capillary gas chromatography using an electron-capture detector. The analysis was possible because of the good electron-capture response of these compounds, which belong to the so-called "coagated electrophores".

18. A. Kujumgiev, V. Bankova, A. Ignatova, S. Popov; "Antibacterial activity of propolis, some of its components and their analogs"; *Pharmazie* 1993, **48**, 785 - 786. No link available.

19. V. Bankova, S. Popov, G. Bocari, E. Haxhalushi; "Phenolics from Albanian poplar buds and their relationship to propolis", *Fitoterapia* 1994, **65**, 326 - 330.

<http://kdb.kew.org/kdb/detailedresult.do?id=115912>

20. V. Bankova, R. Christov, S. Popov, O. Pureb, G. Bocari; "Volatile constituents of propolis", *Z. Naturforsch.* 1994, **49c**, 6 - 10.

<https://www.degruyter.com/downloadpdf/j/znc.1994.49.issue-1-2/znc-1994-1-202/znc-1994-1-202.pdf>

Abstract: Volatile oils from Albanian, Bulgarian and Mongolian propolis have been investigated.

21. V. Bankova, R. Christov, A. Kujumgiev, M. C. Marcucci, S. Popov. "Chemical Composition and antibacterial activity of Brazilian propolis", *Z. Naturforsch.* 1995, **50c**, 167 - 172.

<https://www.degruyter.com/downloadpdf/j/znc.1995.50.issue-3-4/znc-1995-3-402/znc-1995-3-402.pdf>

Abstract: Four samples of Brazilian propolis were investigated by GC/MS of different fractions. 32 volatile compounds, (10 of them new for propolis), as well as 12 more polar compounds (one of them new for propolis) were identified. Antibacterial activity was found in some propolis fractions.

22. N. D. Ivanovska, V. B. Dimov, S. Pavlova, V. S. Bankova, S. S. Popov. "Immunomodulatory action of propolis. V. Anticomplementary action a of water-soluble derivative", *J. Ethnopharmacol.* 1995, **47**, 135 - 143.

<http://www.sciencedirect.com/science/article/pii/037887419501273G>

Abstract: The effect of a water-soluble derivative (WSD) of propolis on the classical pathway (CP) and the alternative (AP) complement activity has been investigated. The in vitro experiments show that WSD inhibits both pathways and the effect depends on the source of complement. The suppression of complement-mediated haemolysis proves to be time- and temperature-related. High WSD concentrations cause direct damage of the target erythrocytes. The estimation of C3-residual activity indicates that the preparation diminishes C3 functional activity.

23. N. D. Ivanovska, V. B. Dimov, V. S. Bankova, S. S. Popov. "Immunomodulatory action of propolis. VI. Influence of a water soluble derivative on complement activity in vivo", *J. Ethnopharmacol.* 1995, **47**, 145 - 147.

<http://www.sciencedirect.com/science/article/pii/037887419501272F>

Abstract The water soluble derivative (WSD) of propolis in a dose of 150 mg/kg was administered intravenously (i.v.), intraperitoneally (i.p.) and orally (p.o.) to mice. The alteration of serum alternative pathway (AP) complement level was observed. The WSD also influenced the process of acute inflammation provoked by zymosan in mice. The effect was strongly dependent on the route of WSD administration.

24. N. Ivanovska, H. Neychev, Z. Stefanova, V. Bankova, S. Popov. Influence of cinnamic acid on lymphocyte proliferation, cytokine release and *Klebsiella* infection in mice", *Apidologie* 1995, **26**, 73 - 81.

https://www.apidologie.org/articles/apido/pdf/1995/02/Apidologie_0044-8435_1995_26_2_ART0001.pdf

Abstract: The effect of a complex of propolis constituents, cinnamic acid and L-lysine, on host defence mechanisms was investigated. After intraperitoneal (ip) application of cinnamic acid lysine complex (CN·Ly) to mice at a dose of 3 mg/kg for 3 consecutive days prior to inoculation with *Klebsiella pneumoniae*, a protective effect was observed. The action of CN·Ly was associated with its ability to provoke proliferation of thymic and splenic lymphocytes and to augment mitogen-induced proliferation, and the release of interleukin-1 (IL-1) and interleukin-2 (IL-2). In all experiments, a solution of L-lysine in a concentration equivalent to the lysine content in the complex was used for comparison. The results indicated that free lysine acted in a mitogen-like manner predominantly on T-lymphocytes. In the complex, lysine played the role of a carrier and exhibited a less pronounced effect.

25. V. Bankova, M. C. Marcucci, S. Simova, N. Nikolova, A. Kujumgiev and S. Popov; "Antibacterial diterpenic acids from Brazilian propolis". *Z. Naturforsch.* 1996, **51c**, 277 - 280.

<https://www.degruyter.com/downloadpdf/j/znc.1996.51.issue-5-6/znc-1996-5-602/znc-1996-5-602.pdf>

Abstract: Four labdane-type diterpenic acids and syringaldehyde were isolated and identified from Brazilian propolis. All the compounds exhibit antibacterial activity. The diterpenes, found for the first time in propolis, are typical for some *Araucaria* species and thus indicate a possible plant source of Brazilian propolis.

26. V. Bankova, N. Nikolova, M. Marcucci; "A new lignan from Brazilian propolis". *Z. Naturforsch.* 1996, **51b**, 735 - 737.

<https://www.degruyter.com/downloadpdf/j/znc.1996.51.issue-9-10/znc-1996-9-1019/znc-1996-9-1019.pdf>

Abstract: The known flavonoid kaempferid 4 and a new benzofurane lignan 1 were isolated from Brazilian propolis. The new lignan is a suitable taxonomic marker and gives new possibilities for identification of propolis plant sources in the tropical regions

27. P. Georgieva, N. Ivanovska, V. Bankova, S. Popov; "Anticomplement activity of lysine complexes of propolis phenolic constituents and their synthetic analogs". *Z. Naturforsch.* 1997, **52c**, 60 - 64.

<https://www.degruyter.com/downloadpdf/j/znc.1997.52.issue-1-2/znc-1997-1-211/znc-1997-1-211.pdf>

Abstract: Several phenolic constituents of propolis and their synthetic analogs were derivatized with L-lysine. The ability of these complexes to alter complement activity was estimated in vitro in human serum. The influence of selected complexes on C3 hemolytic activity via classical pathway (CP) and alternative pathway (AP) and on zymosan-induced AP activation was determined. The results suppose that the anticomplement effect of the complexes might be related to the interaction with C3 complement component.

28. G. Boudourova-Krasteva, V. Bankova, J. M. Sforcin, N. Nikolova and S. Popov, "Phenolics from Brazilian propolis". *Z. Naturforsch.* 1997, **52c**, 676 - 679.

<https://www.degruyter.com/downloadpdf/j/znc.1997.52.issue-9-10/znc-1997-9-1016/znc-1997-9-1016.pdf>

Abstract: The main phenolic constituents from Brazilian propolis, originating from Sao Paulo State, were isolated and identified: three flavonoids, a prenylated coumaric acid and two new benzopyranes, E and Z 2,2-dimethyl-6-carboxyethenyl-8-prenyl-2H-benzopyranes

29. M. C. Marcucci, J. Rogriguez, F. Ferreres, V. Bankova, R. Grotto and S. Popov. Chemical composition of propolis from Sao Paulo State. *Z. Naturforsch.* 1998, **53c**, 117 - 119.

<https://www.degruyter.com/downloadpdf/j/znc.1998.53.issue-1-2/znc-1998-1-220/znc-1998-1-220.pdf>

Abstract: Two propolis samples from Sao Paulo State were investigated by GC/MS. 39 compounds were identified, 8 being new for propolis. Both samples showed some similarities in their qualitative composition. In one of them, coumaric acid and its prenylated derivatives predominated, while in the other one triterpenic alcohols were the main constituents.

30. R. Christov, V. Bankova, A. Hegazi, F. Abd El Hady, S. Popov. Chemical composition of Egyptian propolis. *Z. Naturforsch.* 1998, **53c**, 107- 200.

<https://www.degruyter.com/downloadpdf/j/znc.1998.53.issue-3-4/znc-1998-3-409/znc-1998-3-409.pdf>

Abstract: A sample from Egyptian propolis was investigated by TLC and GC/MS. 39 compounds were identified, 8 being new for propolis. Partial structures of four new esters of caffeic acid have been proposed.

31. V. Bankova, R. Christov, C. Marcucci and S. Popov. Constituents of Brazilian geopropolis. *Z. Naturforsch.* 1998, **53c**,402 - 406.

<https://www.degruyter.com/downloadpdf/j/znc.1998.53.issue-5-6/znc-1998-5-616/znc-1998-5-616.pdf>

Abstract: Three samples of Brazilian geopropolis, collected by three indigenous bee species, were investigated by GC/MS. More than 50 compounds were identified, mainly terpenoids and phenolics. The chemical composition of propolis gathered by different bee species was different.

32. V. Bankova, G. Boudourova-Krasteva, S. Popov, J. M. Sforcin, S. R. Cunha Funari. Seasonal variations of the chemical composition of Brazilian propolis. *Apidologie* 1998, **29**, 361 - 367.

https://www.apidologie.org/articles/apido/pdf/1998/03/Apidologie_0044-8435_1998_29_4_ART0006.pdf

Abstract: The seasonal variations in the chemical composition of Brazilian propolis, collected by two bee subspecies, Africanized *Apis mellifera* and European *Apis mellifera ligustica*, have been investigated by GC and GC-MS. The main components of the samples were phenolic compounds, especially cinnamic acid derivatives, the only exception being the autumn sample from *Apis mellifera ligustica*, where diterpenes predominated. In propolis from both subspecies, diterpenes appeared in summer and reached maximum percentage in autumn, but were absent during the other seasons. The results obtained indicated that both bee subspecies collect propolis from among the same group of plants, and that there are at least two important plant sources, but these remain unidentified.

33. V. Bankova, R. Christov, A. Delgado Tejera. Lignans and other constituents of propolis from Canary Islands. *Phytochemistry* 1998, **49**, 1411 - 1415.

<http://www.sciencedirect.com/science/article/pii/S0031942298001083>

Abstract: Two propolis samples, collected at different location on Gran Canaria (Canary Islands) were analysed by TLC and GC-MS. In volatiles sesquiterpenoids predominated, while

alcoholic extracts mainly contain furofuran lignans and carbohydrates. Partial structures of two new lignans are proposed.

34. V. Bankova, G. Boudourova-Krasteva, S. Popov, J. Sforcin, S. C. Funari. Seasonal variations in essential oil from Brazilian propolis, *J. Essent. Oil Research* 1998, **10**, 693-696.

<http://dx.doi.org/10.1080/10412905.1998.9701012>

Abstract: Seasonal variations in the chemical composition of essential oil from Brazilian propolis were investigated using GC and GC/MS. Seasonal differences were not very significant and were predominantly quantitative. The main components of the oil were spathulenol (3.0–13.9%), (2Z,6E)-farnesol (1.6–14.9%), benzyl benzoate (0.3–18.3%) and prenylated acetophenones (3.4–17.1%). On the basis of the results obtained some suggestions are made concerning the plant origin of the oils.

35. A. Kujumgiev, I. Tsvetkova, Yu. Serkedjieva, V. Bankova, R. Christov and S. Popov. Antibacterial, antifungal and antiviral activity of propolis from different geographic origin. *J. Ethnopharmacol.* 1999, **64**(3), 235 - 240.

<https://pdfs.semanticscholar.org/26ce/cdc1cc51562aecb6176a3ad63b22b54b34bc.pdf>

Abstract: Propolis samples from different geographic origins were investigated for their antibacterial (against *Staphylococcus aureus* and *Escherichia coli*), antifungal (against *Candida albicans*) and antiviral (against Avian influenza virus) activities. All samples were active against the fungal and Gram-positive bacterial test strains, and most showed antiviral activity. The activities of all samples were similar in spite of the differences in their chemical composition. In samples from the temperate zone, flavonoids and esters of phenolic acids are known to be responsible for the above mentioned activities of bee glue; tropical samples did not contain such substances but showed similar activities. Obviously, in different samples, different substance combinations are essential for the biological activity of the bee glue. It seems that propolis has general pharmacological value as a natural mixture and not as a source of new powerful antimicrobial, antifungal and antiviral compounds.

36. R. Christov, V. Bankova, I. Tsvetkova, A. Kujumgiev and A. Delgado Tejera. Antibacterial furofuran lignans from Canary Islands propolis. *Fitoterapia* 1999, **70**, 89 - 92. (No abstract available – short communication)

<http://www.sciencedirect.com/science/article/pii/S0367326X98000446>

37. V. Bankova, R. Christov, S. Popov, M.C. Marcucci, I. Tsvetkova and A. Kujumgiev. Antibacterial activity of essential oils from Brazilian propolis. *Fitoterapia* 1999, **70**, 190 - 193. (No abstract available – short communication)

<http://www.sciencedirect.com/science/article/pii/S0367326X98000458>

38. V. Bankova, G. Boudourova-Krasteva, J. M. Sforcin, X. Frete, A. Kujumgiev, R. Maimoni-Rodella and S. Popov. Phytochemical evidence for the plant origin of Brazilian propolis from Sao Paulo state. *Z. Naturforsch.* 1999, **54c**, 401 - 405.

<https://www.degruyter.com/downloadpdf/j/znc.1999.54.issue-5-6/znc-1999-5-616/znc-1999-5-616.pdf>

Abstract: Propolis and plant secretions from three species, most frequently mentioned as botanical sources of the bee glue in Brazil (*Baccharis dracunculifolia*, *Araucaria angustifolia* and *Eucalyptus citriodora*) have been investigated using GC-MS. Based on chemical evidence, *B. dracunculifolia* was shown to be the main propolis source in São Paulo state. The antibacterial and antifungal activities of all four materials were also tested, the most active being propolis and *Baccharis* leaf exudate.

39. V. Bankova, S. L. de Castro, M. C. Marcucci. Propolis: recent advances in chemistry and plant origin. *Apidologie* 2000, **31**, 3 - 15.

<https://www.apidologie.org/articles/apido/pdf/2000/01/M0105.pdf>

Abstract: New information published since 1995 about propolis constituents is reviewed. The available information on the biological action of new found components is presented. Recent publications are reviewed on propolis of native South American stingless bees. The plant sources of bee glue are discussed, taking into consideration data based on reliable chemical evidence including comparisons between propolis samples and plant material. Some aspects of the chemical standardization of propolis are discussed.

40. M.C. Marcucci, F. Ferreres, A. R. Custodio, M. M. C. Ferreira, V. S. Bankova, C. Garcia-Viguera, W. A. Bretz. Evaluation of phenolic compounds in Brazilian propolis from different geographic regions. *Z. Naturforsch.* 2000, **55c**, 76 - 81.

<https://www.degruyter.com/downloadpdf/j/znc.2000.55.issue-1-2/znc-2000-1-215/znc-2000-1-215.pdf>

Abstract: Chemometrics has been shown quite efficient to uncover relationships between chemical composition of a sample and its geographical origin. Forty propolis samples originated from the the South and South East of Brazil were analyzed by HPLC and 18 compounds of interest were studied which included: caffeic, p-coumaric and ferulic acids, and some of their derivatives, pinobanksin, a derivative of kaempferol and five phenolic compounds (assigned as 3-prenyl-4-hydroxycinnamic acid (PHCA); 2,2-dimethyl-6-carboxyethnyl-2H-1-benzopyran (DCBE); 3,5-diprenyl-4-hydroxycinnamic acid (DHCA); compound E (still unknown) and 6-propenoic-2,2-dimethyl-8-prenyl-2H-1-benzopyran acid (DPB). Principal Component Analysis (PCA) indicated three different groups of propolis samples, having the same typical chromatogram, evaluated by HPLC. Samples from the South East group were rich in derivatives of kaempferol. Samples from the South group I had a high content of DPB compound, but a low concentration of kaempferol derivatives and of DCBEN compound. Samples from the South group II were characterized by a high concentration of DCBEN , DHCA , p-coum-aric and DPB compounds. Therefore, the identification of new compounds in Brazilian propolis can give useful information about the plant sources of a given geographic region.

41. M. C. Marcucci, V. S. Bankova. Chemical composition, plant origin and biological activity of Brazilian propolis. *Current Topics in Phytochemistry* 1999, Vol. 2, pp. 115 - 123.

<http://www.researchtrends.net/tia/abstract.asp?in=0&vn=2&tid=24&aid=3945&pub=1999&type=3>

Abstract: The chemical composition of Brazilian propolis has been reviewed, with special attention to the components specific for Brazilian samples. The plant sources of Brazilian bee glue are discussed, taking into consideration data based on reliable chemical evidence, including comparison between propolis samples and plant material. The available information on the biological action of Brazilian propolis and its components is presented.

42. V. Bankova. Determining quality in propolis samples. *Bee Informed* 7(2), 11 – 13 (2000). (No abstract available)

<http://utahcountybeekeepers.com/Other%20Files/Information%20Articles/Determining%20Propolis%20Quality.pdf>

43. R. O Orsi, S. R. C. Funari, A. M. V. C. Soares, S. A. Calvi, S. L. Oliveira, J. M. Sforcin, V. Bankova. Immunomodulatory action of propolis on macrophage activation. *J. Venom. Anim. Toxins*, 6 (2), 205 – 219 (2000).

http://www.scielo.br/scielo.php?pid=S0104-79302000000200006&script=sci_arttext

Abstract: Propolis has been the subject of several recent studies, with the aim of elucidating its biological and pharmacological properties. Propolis has a well-known antimicrobial activity as well as antioxidant, antitumoral, antiinflammatory, and regenerative properties, but literature about its effects on the immune response is scarce. The goal of this work was to evaluate the propolis effect on macrophage activation by oxygen (H₂O₂) and nitrogen (NO) metabolite determination. Propolis was produced by africanized honeybees and hydroalcoholic solutions were prepared at different concentrations. Peritoneal macrophages were obtained from male BALB/c mice and culture cells were stimulated *in vitro* with propolis or interferon-gamma (IFN- γ). In the *in vivo* assay, the animals were sacrificed after propolis treatment and cells were stimulated with IFN- γ . We also investigated the co-stimulant action of propolis associated with IFN- γ on macrophages. The results show that propolis induces a discreet elevation in H₂O₂ release and a mild inhibition of NO generation, depending on concentration. Propolis had no co-stimulant activity, diminishing IFN- γ action on H₂O₂ and NO production. Data suggest that propolis acts on host non-specific immunity by macrophage activation.

44. J. M. Sforcin, A. Fernandes, C. A. Lopes, V. Bankova, S. R. Funari. Seasonal effect on Brazilian propolis antibacterial activity. *J Ethnopharmacol.* **73**(1-2), 243-249 (2000)

[https://doi.org/10.1016/S0378-8741\(00\)00320-2](https://doi.org/10.1016/S0378-8741(00)00320-2)

Abstract: The behavior of microorganisms towards the antibiotic action of propolis has been widely investigated. Since reports dealing with seasonal effect on propolis activity are not available, this assay was carried out aiming to observe the *in vitro* antimicrobial activity of propolis, collected during the four seasons, on bacterial strains isolated from human infections. Dilution of ethanolic extract of propolis (EEP) in agar was the method performed, with serial concentrations ranging from 0.4 to 14.0% (% v/v). The behavior of some bacteria was analysed according to the incubation period in medium plus propolis, and the survival curve was plotted. It was verified that the growth of Gram-positive bacteria is inhibited by low propolis concentrations (0.4%) whereas Gram-negative bacteria were less susceptible to this substance, the minimal inhibitory concentration ranging from 4.5 to 8.0%. There was no significant difference with regards to the seasonal effect on the survival curve of *Staphylococcus aureus* and *Escherichia coli*; after incubation with propolis, there was an efficient antimicrobial action, mainly towards Gram-positive bacteria.

45. M. Velikova, V. Bankova, M.C. Marcucci, I. Tsvetkova and A. Kujumgiev. Chemical composition and biological activity of propolis from Brazilian Meliponinae. *Z. Naturforsch.* **55c**, 785 – 789 (2000).

<https://www.degruyter.com/downloadpdf/j/znc.2000.55.issue-9-10/znc-2000-9-1018/znc-2000-9-1018.pdf>

Abstract: Twenty-one propolis samples produced by 12 different Meliponinae species were analyzed by GC-MS. Several chemical types of stingless bees' propolis could be grouped, according to the prevailing type of compounds like: "gallic acid", "diterpenic" and "triterpenic" types. The results confirm that neither the bee species nor the geographical location determine the chemical composition of Meliponinae propolis and the choice of its plant source, respectively. This could be explained by the fact that Meliponinae forage over short distances (maximum 500 m) and thus use as propolis source the first plant exudate they encounter during their flights. The antibacterial, antifungal and cytotoxic activities of the samples were also investigated. Most samples had weak or no activity against *E. coli*, weak action against *Candida albicans*. Some of them showed significant activity against *St. aureus*., presumably connected to the high concentration of diterpenic acids. Samples rich in diterpenic acids possessed also high cytotoxic activity (*Artemia salina* test).

46. M. Velikova, V. Bankova, K. Sorkun, S. Houcine, I. Tsvetkova and A. Kujumgiev. Propolis from the Mediterranean region: chemical composition and antimicrobial activity. *Z. Naturforsch.* **55c**, 790 – 793 (2000).

<https://www.degruyter.com/downloadpdf/j/znc.2000.55.issue-9-10/znc-2000-9-1019/znc-2000-9-1019.xml>

Abstract: The chemical composition of propolis from Bulgaria, Turkey, Greece and Algeria was investigated by GC-MS. All of them contained mainly flavonoids and esters of caffeic and ferulic acids, which indicated that their main source are buds of poplars of the taxonomic section *Aegieros*. Some Turkish samples contained a low percent of diterpenic acids, while in Algerian samples significant amounts of a hydroxyditerpenic acid (M=322, its structure not determined by its MS) were found. All samples showed significant antibacterial and weak to moderate antifungal activity.

47. M. Velikova, V. Bankova, I. Tsvetkova, A. Kujumgiev, M. C. Marcucci. Antibacterial *ent*-kaurene from Brazilian propolis of native stingless bees. *Fitoterapia* **71**,693 - 696 (2000)

[https://doi.org/10.1016/S0367-326X\(00\)00213-6](https://doi.org/10.1016/S0367-326X(00)00213-6)

Abstract: Three *ent*-kaurene diterpenoids, not previously described as constituents of propolis, were isolated from a sample collected by Brazilian native bees *Melipona quadrifasciata anthidioides*. One of them, kaurenoic acid, as well as the total extract, displayed moderate antibacterial activity.

48. V. Bankova, M. C. Marcucci. Standardization of propolis: Present status and perspectives. *Bee World* **81**(4), 182 - 188 (2000).

<http://www.tandfonline.com/doi/abs/10.1080/0005772X.2000.11099492>

Abstract: Propolis has been known for its healing properties for centuries, and today many propolis products are available commercially. However, propolis collected from different regions of the world will have different compositions and will vary in activity. It is therefore important to establish standards for the composition of propolis, where the presence and amount of important constituents, and absence of contamination can be determined.

49. J. M. Sforcin, A. Fernandes Junior, C. A. M. Lopes, S. R. C. Funari, V. Bankova. Seasonal effect of Brazilian propolis on *Candida albicans* and *Candida tropicalis*. *J. Venom. Anim. Toxins*, **7** (1), 139 - 144 (2001).

<http://www.tandfonline.com/doi/abs/10.1080/0005772X.2000.11099492>

Abstract: Propolis has been known for its healing properties for centuries, and today many propolis products are available commercially. However, propolis collected from different regions of the world will have different compositions and will vary in activity. It is therefore important to establish standards for the composition of propolis, where the presence and amount of important constituents, and absence of contamination can be determined.

50. M.C. Marcucci, F. Ferreres, C. García-Viguera, V.S. Bankova, S.L. De Castro, A.P.Dantas, P.H.M. Valente, N. Paulino. Phenolic compounds from Brazilian propolis with pharmacological activities. *J. Ethnopharmacology* **74** (2) 105 - 112, (2001).

[https://doi.org/10.1016/S0378-8741\(00\)00326-3](https://doi.org/10.1016/S0378-8741(00)00326-3)

Abstract: Four compounds were isolated from Brazilian propolis. They are identified as: (1) 3-prenyl-4-hydroxycinnamic acid (PHCA), (2) 2,2-dimethyl-6-carboxyethenyl-2H-1-benzopyrane (DCBEN), (3) 3,5-diprenyl-4-hydroxycinnamic acid (DHCA), and (4) 2,2-dimethyl-6-carboxyethenyl-8-prenyl-2H-1-benzopyran (DPB). The structures of the compounds were determined by MS and NMR techniques. All compounds were assayed against *Trypanosoma cruzi* and the bacteria *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus faecalis*. Compounds (1) to (4) were active against *T. cruzi*. Except (1), all compounds presented activity against the bacteria tested. When compounds (1)–(3) were tested in the guinea pig isolated trachea, all induced a relaxant effect similar to propolis extract.

51. Velikova, V. Bankova, K. Sorkun, S. Popov, A. Kujungiev. Chemical composition and biological activity of propolis from Turkish and Bulgarian origin. *Mellifera* **1** (1), 57 - 59 (2001)

<http://eds.b.ebscohost.com/eds/pdfviewer/pdfviewer?vid=1&sid=08a25b90-a63f-4047-81b2-a62bfa0debe1%40sessionmgr103>

Abstract: The study investigated the chemical composition of propolis from one Bulgarian and two Turkish propolis samples by gas chromatography-mass spectrometry (GC-MS). Their chemical composition was similar and indicated their origin from buds of poplars of the section *Aegios*. They contained mainly flavonoids and esters of caffeic and ferulic acids. All samples showed significant antibacterial, antifungal and cytotoxic activities. The similarity in plant origin, chemical composition and biological activity is an indication that Turkish propolis can be taken into consideration in the future elaboration of European criteria for quality control of propolis.

52. Kortenska, M. Velikova, I Totzeva, V. Bankova, M. Marcucci. Antioxidant properties of cinnamic acid derivatives on the lipid autooxidation. *Compt. Rend. De l'Academie bulgare de Sciences* **54**(1), 47 - 50 (2001). (No abstract available – short communication)

<http://adsabs.harvard.edu/full/2001CRABS..54a..47K>

53. M. Popova, V. Bankova, S. Spassov, I. Tsvetkova, C. Naydenski, M. V. Silva, M. Tsartsarova. New Bioactive Chalcones in Propolis from El Salvador. *Z. Naturforsch.* **56c**, 593 – 596 (2001).

<https://www.degruyter.com/downloadpdf/j/znc.2001.56.issue-7-8/znc-2001-7-819/znc-2001-7-819.xml>

Abstract: 2',3'-Dihydroxy-4,4'-dimethoxychalcone (1) and 2',3',4-trihydroxy-4'-methoxychalcone, two new chalcones, were isolated from propolis from El Salvador. The compounds showed significant antibacterial and antifungal activity and moderate toxicity to *Artemia salina* nauplii

54. Popova, V. Bankova, I. Tsvetkova, C. Naydenski, M. V. Silva. The First Glycosides Isolated from Propolis: Diterpene Rhamnosides. *Z. Naturforsch.* **56c**, 1108 – 1111 (2001).

<https://www.degruyter.com/downloadpdf/j/znc.2001.56.issue-11-12/znc-2001-11-1230/znc-2001-11-1230.xml>

Abstract: Two diterpene glycosides, ent-8(17)-labden-15-O- α -L-rhamnoside and ent-8(17)-labden-15-O-(3'-O-acetyl)- α -L-rhamnoside (new natural compounds) were isolated from propolis from El Salvador. The compounds showed significant antibacterial activity and moderate toxicity to *Artemia salina* nauplii. These are the first glycosides reported in bee glue.

55. D. Kortenska, M. P. Velikova, N. V. Yanishlieva, I. R. Totzeva, V. S. Bankova, M. C. Marcucci. Kinetics of lipid oxidation in the presence of

cinnamic acid derivatives. *European Journal of Lipid Science and Technology*, **104** (1), 19-28 (2002).

[http://onlinelibrary.wiley.com/doi/10.1002/1438-9312\(200201\)104:1%3C19::AID-EJLT19%3E3.0.CO;2-Q/full](http://onlinelibrary.wiley.com/doi/10.1002/1438-9312(200201)104:1%3C19::AID-EJLT19%3E3.0.CO;2-Q/full)

Abstract: The effects of seven (prenyl- and methoxy-) derivatives of cinnamic acid (0.1 mM) on the kinetics of lipid (sunflower oil triacylglycerols, TGSO) bulk phase oxidation at 80 °C have been compared. Synthesis of prenyl cinnamic acid derivatives: 3-prenyl-4-hydroxy-cinnamic acid (PHC), 3,5-diprenyl-4-hydroxy-cinnamic acid (DPHC), 2,2-di-methyl-6-carboxy-ethenyl-2H-benzopyran (DMCB), 2,2-dimethyl-6-carboxy-ethenyl-8-prenyl-2H-benzopyran (DCEPB) present in Brazilian propolis has been performed. The monoprenyl derivative (PHC) has been found to exert a higher antioxidant activity as compared to the diprenyl derivative (DPHC). However, cinnamic acid derivatives DMCB and DCEPB have caused no change in the kinetics of TGSO oxidation. The results obtained have been compared with those on related compounds containing a cinnamic acid moiety as a structural feature, such as 4-hydroxy-cinnamic (p-coumaric), 3-methoxy-4-hydroxy-cinnamic (ferulic) and 3,5-dimethoxy-4-hydroxy-cinnamic (sinapic) acids, as well as with data on butylated hydroxytoluene (BHT) and α -tocopherol (α Toc). PHC has shown a stronger antioxidant efficiency than BHT, p-coumaric and ferulic acid, but a weaker antioxidant efficiency than α -Toc and sinapic acid. The observed antioxidant effect of DPHC was stronger than that of p-coumaric and ferulic acids and weaker than that of α -Toc, BHT and sinapic acid.

56. J.M. Murad, S. A. Calvi, A.M. V. Soares, V. Bankova, J. M. Sforcin. Effect of propolis from Brazil and Bulgaria on fungicidal activity of macrophages against *Paracoccidioides brasiliensis*. *J. Ethnopharmacology* **79** (3), 331 - 334 (2002).

[https://doi.org/10.1016/S0378-8741\(01\)00404-4](https://doi.org/10.1016/S0378-8741(01)00404-4)

Abstract: Paracoccidioidomycosis is the most important systemic mycosis in Latin America. Its etiological agent, *Paracoccidioides brasiliensis*, affects individuals living in endemic areas through inhalation of airborne conidia or mycelial fragments. The disease may affect different organs and systems, with multiple clinical features, with cell-mediated immunity playing a significant role in host defence. Peritoneal macrophages from BALB/c mice were stimulated with Brazilian or Bulgarian propolis and subsequently challenged with *P. brasiliensis*. Data suggest an increase in the fungicidal activity of macrophages by propolis stimulation, independently from its geographic origin.

57. M. Popova, V. Bankova, A. Chimov, M. S. Vides. A scientific note on the high toxicity of propolis that comes from *Myroxilon balsamum* trees.

Apidologie **33**, 87 - 88 (2002). (No abstract available – short communication)

<https://www.apidologie.org/articles/apido/pdf/2002/01/Popova.pdf>

58. V. Bankova, M. Popova, S. Bogdanov, and A.G. Sabatini. Chemical composition of European propolis: expected and unexpected results. *Z. Naturforsch.* **57c**, (5/6) 530 - 533 (2002).

<https://www.degruyter.com/downloadpdf/j/znc.2002.57.issue-5-6/znc-2002-5-622/znc-2002-5-622.xml>

Abstract: Ten propolis samples from Bulgaria, Italy and Switzerland were analyzed by GC-MS. As expected, most samples displayed the typical chemical pattern of “poplar” propolis: they contained pinocembrin, pinobanksin and its 3-O-acetate, chrysin, galangin, prenyl esters of caffeic and ferulic acids. Two samples differed significantly: one from the Graubünden Alpine region, Switzerland, rich in phenolic glycerides, and one from Sicily which contained only a limited number of phenolics and was rich in diterpenic acids.

59. V.D. Kortenska, V.A. Roginski, V.S. Bankova, M.P. Popova, T.K. Barsukova. Study of the antioxidative efficiencies of new chalcones from propolis of El Salvador during methyl linoleate oxidation in micellar solutions. *La Rivista Italiana di Sostanze Grasse* LXXX, 323 – 327, (2003).

<http://cat.inist.fr/?aModele=afficheN&cpsidt=15267684>

Abstract: The antioxidant efficiency for two new chalcones isolated from El Salvador's propolis: 2',3',4-trihydroxy-4'-methoxy-chalcone (BH₂) and 2',3'-dihydroxy-4,4'-diethoxy-chalcone (AH₂) were determined by analyzing the kinetics of oxygen consumption in SDS micellar systems. The results demonstrated that AH₂ and BH₂ showed moderate chain-breaking activities higher than naringenin, but lower than caffeic acid, butylated hydroxy toluene and DL- α -tocopherol.

60. M. Popova, V. Bankova, D. Butovska, V. Petkov, B. Damyanova, A.G. Sabatini., G.L. Marazzan, S. Bogdanov. Poplar type propolis and analysis of its biologically active components. *Honeybee Science* **24**(2), 61 – 66 (2003). (In Japanese)

61. F.C. Lopes, V. Bankova, J.M. Sorcin. Effect of three vegetal sources of propolis on macrophages activation. *Phytomedicine* **10**(4), 343 (2003). (No abstract available – short communication)

<https://doi.org/10.1078/094471103322004848>

62. Prytyk E, Dantas AP, Salomao K, Pereira AS, Bankova VS, Castro SL, Neto FR. Flavonoids and trypanocidal activity of Bulgarian propolis. *J Ethnopharmacol.* **88**(2-3),189-193 (2003).

<http://www.sciencedirect.com/science/article/pii/S0378874103002101>

Abstract: Acetone and ethanol extracts of two Bulgarian propolis samples (**Bur** and **Lov**) were investigated by high temperature high resolution gas chromatography coupled to mass spectrometry (HT-HRGC-MS), and their activity against *Trypanosoma cruzi* was evaluated. The ethanol extracts—**Et-Bur** and **Et-Lov**—showed similar composition, with a high content of flavonoids, and strong inhibitory activity against *T. cruzi* proliferative epimastigotes, which were more susceptible than trypomastigotes. In the presence of blood, the activity of **Et-Bur** or **Et-Lov** against trypomastigotes was similar to that of the standard drug, crystal violet. Both extracts also showed similar and significant activity against *Staphylococcus aureus* and *Candida albicans*, while being inactive against *Escherichia coli*. The acetone extract, **Ket-Bur**, was more active than **Et-Bur** against both forms of *T. cruzi*.

63. N. Paulino, A. P. Dantas, V. Bankova, D. Taggliari Longhi, A. Scremin, S.Lisboa de Castro, J. B.Calixto. Bulgarian Propolis Induces Analgesic and Anti-inflammatory Effects in Mice and Inhibits In Vitro Contraction of Airway Smooth Muscle. *J. Pharmacol. Sci.* **93**, 307 – 313 (2003)

https://www.jstage.jst.go.jp/article/jphs/93/3/93_3_307/pdf

Abstract: Propolis is a bee product, which has long been used in folk medicine for the management of different diseases. In this study we evaluated the analgesic and anti-inflammatory effects of a standard ethanolic extract of Bulgarian propolis (Et-Blg) in mice and its in vitro effect on airway smooth muscle. Et-Blg inhibited acetic acid-induced abdominal contortions with an $ID_{50} = 7.4 \pm 0.7 \text{ mg} \cdot \text{kg}^{-1}$. In the formalin test, the extract caused a significant reduction in pain in mice treated with $100 \text{ mg} \cdot \text{kg}^{-1}$ Et-Blg during the neurogenic phase and for the inflammatory phase with all doses of the extract, with an $ID_{50} = 2.5 \pm 0.4 \text{ mg} \cdot \text{kg}^{-1}$. Et-Blg inhibited also the capsaicin-induced ear edema in mice; however, this extract was ineffective when assessed in the tail-flick and hot-plate thermal assays. The analgesic effect of Et-Blg was associated with the inhibition of inflammatory responses and not to a simple irritation of nervous terminals. In vitro, this extract inhibited the contraction of trachea smooth muscle induced by histamine ($IC_{50} = 50 \pm 5 \mu\text{g} \cdot \text{mL}^{-1}$), capsaicin ($IC_{50} = 26.8 \pm 3 \mu\text{g} \cdot \text{mL}^{-1}$), 80 mM KCl ($IC_{50} = 27.8 \pm 3 \mu\text{g} \cdot \text{mL}^{-1}$), and carbachol ($IC_{50} = 54 \pm 2 \mu\text{g} \cdot \text{mL}^{-1}$).

64. B. Trusheva, M. Popova, V. Bankova, I. Tsvetkova, C. Naydensky, A.G. Sabatini. A new type of European propolis, containing bioactive labdanes. *Rivista Italiana E.P.P.O.S.* **36**, 3 – 7 (2003)

https://www.tib.eu/en/search/id/BLSE%3ARN144194777/A-new-type-of-European-propolis-containing-bioactive/?tx_tibsearch_search%5Bsearchspace%5D=tn

Abstract: From a new type European propolis from Sicily, four labdane diterpenes were isolated and their structure determined by spectral methods: E/Z communic acid **1**, isocupressic acid **2**, acetylisocupressic acid **3** and labd-8(17),13E-diene-15-ol **4**. The latter was found for the first time in natural source. All compounds showed significant antibacterial activity and toxicity to *Artemia salina* nauplii.

65. I.B. da Silva Cunha, K. Salomao, M. Shimizu, V. S. Bankova, A. R. Custodio, S. Lisboa de Castro, M. C. Marcucci. Antitrypanosomal Activity of Brazilian Propolis from *Apis mellifera*. *Chem. Pharm. Bull.* **52**(5) 602—604 (2004)

https://www.jstage.jst.go.jp/article/cpb/52/5/52_5_602/_pdf

Abstract: Extracts from different samples of Brazilian propolis were obtained by Soxhlet extraction or maceration at room temperature using ethanol, water, and combination of both solvents. Analysis of their composition using HPLC revealed that no major differences were seen when a propolis sample was subject to different extraction methods. The activity of the 15 extracts was assayed against bloodstream trypomastigotes of *Trypanosoma cruzi*, the etiologic agent of Chagas' disease. Multivariate analysis was applied to evaluate the efficiency of the different extracts and the trypanocidal activity. The extracts could be divided into two groups. In the first, in which, extracts were obtained by reflux in Soxhlet using 100% ethanol, there was a lower content of bioactive compounds and consequently lower trypanocidal activity. Extract 136-Et100 stands out in this group, since it had the highest levels of bioactive compounds together with highest activity against the parasite when compared with all other extracts. The second group comprises extracts with intermediate levels of bioactive compounds and higher activity against *T. cruzi*.

66. A.C. Sawaya, D.M. Tomazela, I.B. Cunha, V.S. Bankova, M.C. Marcucci, A.R. Custodio, M.N. Eberlin. Electrospray ionization mass spectrometry fingerprinting of propolis. *Analyst.* **129**(8) 739-44 (2004)

<http://repositorio.unicamp.br/bitstream/REPOSIP/64619/1/WOS000222997200014.pdf>

Abstract: Crude ethanolic extracts of propolis, a natural resin, have been directly analysed using electrospray ionization mass (ESI-MS) and tandem mass spectrometry (ESI-MS/MS) in the negative ion mode. European, North American and African samples have been analyzed, but emphasis has been given to Brazilian propolis which displays diverse and region-dependent chemical composition. ESI-MS provides characteristic fingerprint mass spectra, with propolis samples being divided into well-defined groups directly related to their geographical origins. Chemometric multivariate analysis statistically demonstrates the reliability of the ESI-MS fingerprinting method for propolis. On-line ESI-MS/MS tandem mass spectrometry of characteristic $[M - H]^-$ ion markers provides an additional dimension of fingerprinting selectivity, while structurally characterizing the ESI-MS marker components of propolis. By comparison with standards, eight such markers have been identified: *para*-coumaric acid, 3-methoxy-4-hydroxycinnamaldehyde, 2,2-dimethyl-6-carboxyethenyl-2H-1-benzopyran, 3-prenyl-4-hydroxycinnamic acid, chrysin, pinocembrin, 3,5-diprenyl-4-hydroxycinnamic acid and dicaffeoylquinic acid. The negative mode ESI-MS fingerprinting method is capable of discerning distinct composition patterns to typify, to screen the sample origin and to reveal characteristic details of the more polar and acidic chemical components of propolis samples from different regions of the world.

67. M. Popova, V. Bankova D. Butovska, V. Petkov, B. Nikolova-Damyanova, A.G. Sabatini, G.L. Marcazzan, S. Bogdanov. Validated methods for the quantification of biologically active constituents of poplar-type propolis *Phytochem Anal.* **15**(4):235-40 (2004).

<http://onlinelibrary.wiley.com/doi/10.1002/pca.777/full>

Abstract: The validation of rapid, low-cost spectrophotometric procedures for the quantification of the three main groups of bioactive substances (flavones and flavonols, flavanones and dihydroflavonols, and total phenolics) in poplar-type propolis has been performed. A spectrophotometric assay based on the formation of an aluminium chloride complex was applied for the quantification of total flavones and flavonols using galangin as standard. Because of the high amount of flavanones and dihydroflavonols in “poplar type” propolis, the introduction of a distinct procedure for their quantification was considered of special significance and the DAB9 colorimetric method was applied for the purpose. Total phenolic content was measured by the Folin–Ciocalteu procedure using a mixture of pinocembrin and galangin as a reference. The procedures were validated using a model mixture of compounds representing the poplar-type propolis composition as found in previous studies. The accuracy (recovery) varied in the range 84–109%, and the relative standard deviation was 0.5–6.2%. The developed spectrophotometric procedures were applied to six poplar type propolis samples. The results were verified independently by a HPLC procedure. The two sets of results agreed satisfactory, as proven by Student's t-test.

68. V. Bankova, M. Popova. Standardizzazione e controllo di qualità della propoli: una breve sintesi. *APOidea* 1, 19 – 23 (2004). (In Italian)

http://agris.fao.org/agris-search/search.do;jsessionid=59837DA8BF6E64207DD2E582A4C492BD?request_locale=fr&recordID=IT2006601930&query=&sourceQuery=&sortField=&sortOrder=&agrovocString=&advQuery=¢erString=&enableField=

Abstract: Propolis has been known for its healing properties for centuries and today many propolis products are available commercially. However, propolis collected from different geographic regions of the world may have different composition and may vary in activity. The lack of commonly accepted standardization and quality control procedures is an obstacle to the wide use of propolis in the mainstream medicinal practice. It is therefore important to establish standards for the composition of propolis, where the presence and amount of important constituents can be determined. Knowledge of propolis plant sources in specific regions could help in solving this problem. Many studies have shown that in the temperate zone bees almost exclusively collect propolis from the bud exudates of poplar trees (*Populus* spp.). The main propolis constituents in these regions are the typical poplar phenolics: flavonoids, phenolic acids and their esters, known to be responsible for the essential pharmacological activities of bee glue. The identification of poplar propolis can be performed by chromatographic identification of several important compounds - taxonomic markers. After that, the amount of the main groups of biologically active compounds has to be measured and spectrophotometric methods are especially suitable, rapid and easy to perform (the AlCl₃ method for total flavones and flavonols, the 2,4-dinitrophenylhydrazine method for total flavanones and dihydroflavonols and the Folin Ciocalteu method for total phenolics). It is also important to determine the antibacterial activity of the samples (MIC against *S. aureus*) and the absence of contaminants such as acaricides and heavy metals.

69. M. Popova, V. Bankova, H. Naydenski, I. Tsvetkova, A. Kujumgiev. Comparative study of the biological activity of propolis from different geographic origin: a statistical approach. *Macedonian Pharmaceutical Bulletin* 50 (1), 9 – 14 (2004).

<http://bulletin.mfd.org.mk/>

Abstract: Propolis samples from different geographic origin and from different bee species (honeybee *Apis mellifera* and stingless tropical bees, subfamily Meliponinae) were analyzed for their antibacterial activity against *Staphylococcus aureus* and toxicity to *Artemia salina* nauplii (potential cytotoxicity). Statistical methods (Student's *t*-test and ANOVA) were applied to find out correlation between biological activity and geographic origin of propolis samples, and also

the influence of the species of the collecting bees on propolis activity. Meliponinae propolis was found to be of statistically significant lower antibacterial activity than the Brazilian and European ones. ANOVA was used also to study the correlation between the antibacterial activity of honeybee propolis from Europe, Brazil and El Salvador: propolis from Europe and Brazil had similar activity despite the well-known drastic differences in chemical composition; this activity was higher than that of propolis from El Salvador. On the other hand, there was no significant correlation between geographic origin and potential cytotoxicity. These results demonstrate that the statistical approach is useful with respect to propolis standardization. This approach could help in finding the best practical application for any specific type of propolis according to its plant source, the chemistry determined by this source and the corresponding biological activity

70. B. Trusheva, M. Popova, H. Naydenski, I. Tsvetkova, J.G. Rodriguez, V. Bankova. New polyisoprenylated benzophenones from Venezuelan propolis. *Fitoterapia*, **75**(7-8), 683-689 (2004)

<https://doi.org/10.1016/j.fitote.2004.08.001>

Abstract: Two new polyisoprenylated benzophenones, 18-ethyloxy-17-hydroxy-17,18-dihydroscrobiculatone A and 18-ethyloxy-17-hydroxy-17,18-dihydroscrobiculatone B, together with the known scrobiculatones A and B, were isolated from Venezuelan propolis. The scrobiculatones A and B showed significant antibacterial activity and moderate toxicity to *Artemia salina* nauplii.

71. R. O. Alves de Lima, A. P. Bazo, R. A. Said, J. M. Sforcin, V. Bankova, B. R. Darros, D. M.F. Salvadori. Modifying Effect of Propolis on Dimethylhydrazine-Induced DNA Damage But Not Colonic Aberrant Crypt Foci in Rats. *Environmental and Molecular Mutagenesis* **45**, 8-16 (2005).

https://www.researchgate.net/profile/Roueda_Said2/publication/8125328_Modifying_Effect_of_Propolis_on_Dimethylhydrazine-Induced_DNA_Damage_but_Not_Colonic_Aberrant_Crypt_Foci_in_Rats/links/57bc859608ae3fbb8643ffd4.pdf

Abstract: Propolis is a honeybee product with several biological and therapeutic properties, including antimutagenic and anticarcinogenic activities. The effects of an aqueous extract of propolis (AEP) were evaluated on the formation of 1,2-dimethylhydrazine (DMH)-induced aberrant crypt foci (ACF) and DNA damage in the colon of male Wistar rats by the ACF and Comet assays, respectively. AEP was administered orally at 0.01%, 0.03%, 0.1%, and 0.3% in the drinking water, which resulted in doses of approximately 12, 34, 108, and 336 mg/kg body weight/day. Animals were also given a single subcutaneous injection of 40 mg/kg DMH and

sacrificed 4 hr later for evaluating DNA damage, or 4 doses of 40 mg/kg DMH, administered 2 doses/week for 2 weeks, and sacrificed 12 weeks after the last injection for evaluating ACF development in the distal colon. Administration of AEP either simultaneously with or after the DMH treatment resulted in no statistically significant reduction of ACF. In contrast, 0.01%, 0.03%, and 0.3% AEP, given simultaneously with DMH, reduced DNA damage induction in the mid and distal colon. However, 0.3% AEP alone increased DNA damage in the colon. In conclusion, AEP had no effect on the formation of DMH-induced ACF in rat colon, but it modulated DMH-induced DNA damage in colon cells. Further investigations are recommended in order to establish the conditions under which propolis produces either protective or deleterious effects.

72. R. O. Orsi, J. M. Sforcin, S. R.C. Funari and V. Bankova. Effects of Brazilian and Bulgarian propolis on bactericidal activity of macrophages against *Salmonella typhimurium*. *International Immunopharmacology*, **5**(2), 359-368 (2005)

<https://doi.org/10.1016/j.intimp.2004.10.003>

Abstract: Propolis has been used in folk medicine since ancient times due to its many biological properties, such as antimicrobial, antiinflammatory, antioxidant, immunomodulatory activities, among others. Macrophages play an important role in the early phase of *Salmonella* infection. In this work, macrophages were prestimulated with Brazilian or Bulgarian propolis and subsequently challenged with *Salmonella Typhimurium* at different macrophage/bacteria ratio. After 60 min of incubation, cells were harvested with Triton-X to lyse the macrophages. To assess the bactericidal activity, the number of colony-forming units (CFU) of *S. typhimurium* was determined by plating 0.1 mL in Mueller Hinton agar. After 24 h, CFU were counted, and the percentage of bactericidal activity was obtained. Propolis from Brazil and Bulgaria enhanced the bactericidal activity of macrophages, depending on its concentration. Brazilian propolis seemed to be more efficient than that from Bulgaria, because of their different chemical composition. In Bulgaria, bees collect the material mainly from the bud exudate of poplar trees, while in Brazil, *Baccharis dracunculifolia* DC. was shown to be the main propolis source. Our data also showed that the increased bactericidal activity of macrophages involved the participation of oxygen (H₂O₂) and nitrogen (NO) intermediate metabolites.

73. V. Bankova. Recent trends and important developments in propolis research. *Evidence Based Complementary and Alternative medicine (eCAM)* **2**(1), 29 – 32 (2005).

<http://downloads.hindawi.com/journals/ecam/2005/108379.pdf>

Abstract: The newest developments in propolis pharmacological research are summarized. The problem regarding biological studies, caused by the chemical variability of propolis, is

discussed. The most important trends and developments in recent propolis research are outlined: biological studies performed with chemically characterized samples, bioassay-guided studies of active principles and comparative biological studies of propolis of different origin and chemical composition. These types of studies are extremely valuable with respect to propolis standardization and practical applications in therapy. They will allow scientists to connect a particular chemical propolis type to a specific type of biological activity and formulate recommendations for practitioners.

74. M. Popova, S. Silici, O. Kaftanoglu, V. Bankova. Antibacterial activity of Turkish propolis and its qualitative and quantitative chemical composition. *Phytomedicine* **12**, 221–228, (2005)

<https://doi.org/10.1016/j.phymed.2003.09.007>

Abstract: The antibacterial activity of propolis from different regions of Turkey was studied, accompanied by TLC and GC-MS analyses of its chemical composition and spectrophotometric quantification of the most important active principles. All six samples were active against the bacterial test strains used; however, samples 1 (Yozgat), 2 (Izmir) and 3 (Kayseri) were more active than samples 4 (Adana), 5 (Erzurum) and 6 (Artvin). By TLC comparison all samples were found to contain poplar taxonomic markers but in samples 4 (Adana), 5 (Erzurum) and 6 (Artvin), different substances were observed, which were not present in *P. nigra* L. bud exudate. The typical poplar samples 1 (Yozgat), 2 (Izmir) and 3 (Kayseri) displayed very similar phenolic and flavonoid content. Samples 4 (Adana), 5 (Erzurum) and 6 (Artvin) were characterized by low phenolic and very low flavonoid concentrations. Qualitative analysis by GC-MS revealed that sample 4 (Adana) contained diterpenic acids and high percent of cinnamyl cinnamate, sample 5 (Erzurum)—significant amounts of hydroxy fatty acids and triterpenic alcohols, and sample 6 (Artvin)—phenolic glycerides, characteristic for the bud exudate of *Populus euphratica* Oliv. The results confirm the importance of phenolics for propolis antibacterial activity, and the significance of *P. nigra* L. as a propolis source, which provides the hive with the best defense against microorganisms.

75. J.M. Sforcin, R.O. Orsi and V. Bankova. Effect of propolis, some isolated compounds and its source plant on antibody production. *J. Ethnopharmacology*, **98**,(3), 301-305 (2005)

<https://doi.org/10.1016/j.jep.2005.01.042>

Abstract: Propolis is a beehive product with a very complex chemical composition, widely used in folk medicine because of its several therapeutic activities. Its biological properties and chemical composition may vary according to the geographic location and to the different plant sources. The possible mechanism of action of propolis as well as of its active compounds has been the subject of researchers in recent years. In this work, first we reported the results of our

study on the seasonal effect of the immunomodulatory action of propolis on antibody production in bovine serum albumin (BSA)-immunized rats. Then, we compared the effect of Brazilian and Bulgarian propolis, some isolated compounds and *Baccharis* extract on anti-BSA antibody levels. Based on the results, we conclude that propolis stimulates antibody production, independently of the season and geographic origin. Caffeic acid, quercetin and *Baccharis* extract had no effect on antibody production, although the importance of isolated compounds is well reported in other biological assays. Propolis action is a consequence of plant-derived products with synergic effects, while isolated compounds or extracts from its plant sources had no effect in this assay.

76. R. Christov, B. Trusheva, M. Popova, V. Bankova, M. Bertrand. Chemical Composition of propolis from Canada, its antiradical activity and plant origin. *Natural Product Research* **20**(6), 531 – 536 (2006).

www.tandfonline.com/doi/abs/10.1080/14786410500056918

Abstract: The chemical composition of propolis from two regions in Canada was studied: Boreal forest and Pacific coastal forest that lay outside the area of distribution of *Aigeiros* poplars, the usual propolis source plants. In the sample from Victoria, *p*-hydroxyacetophenone, benzyl hydroxybenzoate and cinnamic acid were the major components, accompanied by significant amounts of dihydrochalcones, which allowed the identification of its plant source: *Populus trichocarpa* of Section *Tacamahaca*. Three dihydrochalcones were new for propolis. The sample from Richmond was characterized by large amounts of *p*-coumaric and cinnamic acid, typical for poplars of Section *Leuce*, subsection *Trepidae*, its plant source was identified as *P. tremuloides*. Both samples showed good radical scavenging activity against DPPH. Obviously, Northern type propolis is a promising potential source of biologically active substances and deserves further investigation.

77. V. Bankova. Chemical diversity of propolis and the problem of standardization. *J. Ethnopharmacol.* **100**(1-2), 114 – 117 (2005).

<http://www.sciencedirect.com/science/article/pii/S0378874105003223#>

Abstract: Chemical variability of propolis is discussed with respect to the problem of standardization. Several chemical types of propolis are formulated, based on their plant source. Reliable criteria for chemical standardization of different propolis types are needed but such generally accepted criteria do not yet exist. The chemical profile of “poplar” propolis, typical for the temperate zone, can be characterized by the following parameters: total flavone and flavonol content, total flavanone and dihydroflavonol content, and total phenolics content. These parameters correlate better with the biological activity and are more informative than the quantification of individual components. There is still a lot of work to be done to achieve standardization of other propolis types. Working with standardized material will allow scientists

to connect a particular chemical propolis type to a specific type of biological activity and formulate recommendations for mainstream practitioners.

78. B. Trusheva, M. Popova, V. Kortenska, I. Totseva, J. G. Rodrigues, V. Bankova. Chemical composition and antioxidative activity of propolis from Venezuela. *Oxidation Communications* **28**(2), 490 – 496 (2005).

https://www.researchgate.net/publication/267031181_Chemical_composition_and_antioxidant_activity_of_propolis_from_Venezuela

Abstract: The antioxidant activity of three propolis samples from Venezuela (V-1, V-2, and V-3) was studied and their chemical composition was analysed by GC-MS. We proved that the extracts from the propolis samples acted as effective inhibitors during triacylglycerols of lard autoxidation at 100°C. The effect of V-2 and V-3 was higher (1.5-fold) than that of V-1. The antioxidative activity might be connected, at least partially, to the presence of prenylated benzophenones in the samples, indicated by GC-MS.

79. V.D. Kortenska-Kancheva, V.S. Bankova, M.P. Popova. Antioxidant capacity of new chalcones from propolis of El Salvador during methyl linoleate oxidation in micellar solution. *Oxidation Communications* **28**(3), 525 – 523 (2005).

https://www.researchgate.net/publication/267031233_Antioxidant_capacity_of_new_chalcones_from_propolis_of_El_Salvador_during_methyl_linoleate_oxidation_in_micellar_solutions

Abstract: Combination of kinetic and quantum chemistry methods have been applied to study the antioxidant potency of two new chalcones isolated from El Salvador's propolis: 2',3'-dihydroxy-4,4'-dimethoxy-chalcone (AH 2) and 2',3',4-trihydroxy-4'-methoxy-chalcone (BH2) (Chemical Equation Presented) The antioxidant efficiency of AH2 and BH2 was determined by analysing the kinetics of oxygen consumption in sodium dodecyl sulphate (SDS) micellar systems. The results demonstrated that AH2 and BH2 showed moderate chain-breaking activities higher than naringenin, but lower than caffeic acid, butylated hydroxy toluene and DL- α -tocopherol. The determination of the hyperfine coupling constants was carried out with the density-functional theory (DFT) calculations, using the B3LYP hybrid function, the CPCM option for considering the solvent effect for the mostly dissociated aroxyl radicals and the basic sets 6-311G* and 6-311G** of the Gaussian98 program

80. B. Trusheva, M. Popova, V. Bankova, S. Simova, M. C. Marcucci, P. L. Miorin, F. da Rocha Pasin, I. Tsvetkova. Bioactive Constituents of

Brazilian Red Propolis. *Evidence Based Complementary and Alternative medicine (eCAM)*, **3**(2), 249 – 254 (2006).

<http://downloads.hindawi.com/journals/ecam/2006/934842.pdf>

Abstract: In a new propolis type, red Brazilian propolis, 14 compounds were identified (six of them new for propolis), among them simple phenolics, triterpenoids, isoflavonoids, prenylated benzophenones and a naphthoquinone epoxide (isolated for the first time from a natural source). Three of the major components demonstrated significant antimicrobial activity, and two (obtained as inseparable mixture) possessed radical scavenging activity against 1,1-diphenyl-2-picrylhydrazyl (DPPH).

81. R.O. Orsi, S.R.C. Funari, R. Barbattini, C. Giovani, F. Frilli, J.M. Sforcin, V. Bankova. Radionuclides in honeybee propolis (*Apis mellifera* L.). *Bul. Environ. Contam. Toxicol.* **76**, 637 – 640 (2006) (No abstract available)

<https://rd.springer.com/content/pdf/10.1007%2Fs00128-006-0967-1.pdf>

82. V. Kortenska Kancheva, V. Bankova. A review on the antioxidant activity of propolis and propolis constituents from different geographic origin. *Recent progress in medicinal Plants vol. 14 - Biopharmaceuticals*. Studuim Press, Houston 2006, pp. 81 – 96.

<https://www.amazon.com/RECENT-PROGRESS-MEDICINAL-PLANTS-BIOPHARMACEUTICALS/dp/0976184966>

Abstract: Many articles have been published, focused on the antioxidative properties of propolis and some of its constituents. However, there is no comprehensive review covering this important propolis activity and no generally accepted concept of its practical value as antioxidant and of the role of individual propolis components for its antioxidative properties. One of the reasons for this absence is the striking variability of propolis chemical composition depending on the site of collection, because in different ecosystems different plant exudates and secretions could serve as a source of propolis. In this article we summarize the up-to-date information published on the antioxidative action of propolis extracts and individual propolis components.

83. R. De Olivera Orsi, J.M. Sforcin, S.R.C. Funari, A.Fernandes Junior, V. Bankova. Synergistic effect of propolis and antibiotics on the *Salmonella* Typhi. *Braz. J. Microbiol.* **37**, 108 – 112 (2006).

<http://www.scielo.br/pdf/bjm/v37n2/arg02.pdf>

Abstract: The goal of this work was to investigate a possible synergistic effect between ethanolic extracts of propolis from Brazil and Bulgaria and some antibiotics (Amoxicillin,

Ampicillin and Cefalexin) against *Salmonella* Typhi. Brazilian and Bulgarian propolis showed an antibacterial action, but the sample from Bulgaria was shown to be more efficient. Both samples showed a similar synergistic effect with these antibiotics. One may conclude that the propolis samples show an important antibacterial action, as well as a synergistic effect with antibiotics against *Salmonella* Typhi.

84. V. Bankova, M. Popova, B. Trusheva. Plant sources of propolis: an update from a chemist's point of view. *Nat. Prod. Commun.* **1**(11), 1023 – 1028, (2006).

https://www.researchgate.net/publication/267031234_Plant_sources_of_propolis_An_update_from_a_chemist%27s_point_of_view

Abstract: The newest developments in research on propolis plant sources are summarized. Special attention is paid to data based on reliable chemical evidence including comparison between propolis samples and plant material, and on well documented bee behavior. A number of new proved propolis source plants are listed. Hypothetic sources, suggested as a results of comparison of propolis chemical composition and literature data about particular plants are also discussed.

85. I.B.da Silva Cunha, M.L.T. Rodrigues, E.C. Meurer, V.S. Bankova, M.C. Marcucci, M.N.Eberlin, A.C.H.F. Sawaya. Effect of the maceration time on chemical composition of extracts of Brazilian propolis. *J. Apicult. Res.* **45**(3), 137 – 144 (2006).

<http://www.tandfonline.com/doi/abs/10.1080/00218839.2006.11101332>

Abstract: It is believed that longer maceration times change significantly the ethanolic extract composition of propolis, improving its pharmacological, nutritional and antimicrobial properties. To probe whether such superior composition is indeed obtained, 10 samples of propolis from several regions in Brazil and one sample from North America were left to macerate in 70% ethanol for periods between 20 days and one year. The resultant extracts were evaluated in terms of the yield of extracted material and also analyzed by electrospray ionization mass spectrometry (ESI-MS) fingerprinting. A small increase in the yield of the extracted material over the period analyzed was observed. ESI-MS fingerprints indicate qualitatively the same composition but with a greater incidence of the high-mass components after six months. The extracts of one sample of green Brazilian propolis were also quantified using both gas chromatography mass spectrometry and high performance liquid chromatography. No new components were identified after longer maceration times and the changes in the relative concentrations of the identified components were in line with the results of the yield and ESI-MS fingerprints. Therefore, prolonged extraction periods do not necessarily result in richer propolis extracts.

86. V. Bankova, M. Popova, B. Trusheva, S. Bogdanov, A.-G. Sabatini, M. Marcucci. Propolis: from folk medicine to modern preparations. *Honeybee Science* **27**(2), 63 – 67 (2006). (In Japanese)

<http://agris.fao.org/agris-search/search.do?recordID=JP2007003995>

Abstract: Propolis has been used as a remedy since ancient times, because of its remarkable healing properties. It attracts the attention of modern scientists with its diverse pharmacological activities and low toxicity. At present, there are two main directions in propolis research. One is aimed to the use of propolis in well defined pharmaceutical preparations, and the other one is based on propolis as a source of new biologically active molecules, mainly anticancer, antibacterial and antioxidative agents. Concerning the first line of research, chemical variability of propolis is a serious obstacle to its standardization and respectively, to its official acceptance into the main stream of the health care system. For standardization to be achieved, it is very important to realize that comparing propolis samples from different regions of the world might be the same as comparing extracts of two plants that belong to different plant families. We have proposed a reliable standardization procedure for poplar propolis, a propolis type which contains mainly flavonoids and phenolics. The application of this procedure to a number of samples (over 100) enabled us to describe the typical characteristics of poplar propolis in terms of bioactive components percentage.

87. V. Bankova, M. Popova. Propolis of stingless bees: a promising source of biologically active compounds. *Pharmacognosy reviews* **1**, 88 – 92., 2007

<http://www.phcogrev.com/article.asp?issn=0973-7847;year=2007;volume=1;issue=1;spage=88;epage=92;aui=Bankova;type=0>

Abstract: The information published about chemical composition and biological activity of propolis from stingless bees (Meliponinae) of different genera is reviewed. The available data on the biological action of individual constituents is presented. The plant sources of this propolis are also considered. The perspectives of research and therapeutic use of Meliponinae propolis are discussed.

88. M. Popova, V. Bankova, S. Bogdanov, I. Tsvetkova, C. Naydenski, G.- L. Marcazzan, A.-G. Sabatini. Chemical characteristics of poplar type propolis of different geographic origin. *Apidologie* **38** 306– 311 (2007)

<https://rd.springer.com/content/pdf/10.1051%2Fapido%3A2007013.pdf>

Abstract: Validated spectrophotometric procedures were used to quantify three main groups of bioactive substances (phenolics, flavones/flavonols, flavanones/dihydroflavonols) in 114

samples of poplar-type propolis from different geographic origins. From the results, we characterized raw poplar propolis in terms of minimum content of its bioactive components (antimicrobial and antioxidant) as follows: 45% resin, 21 % total phenolics, 4% total flavones/flavonols; 4% total flavanones/dihydroflavonols, and a maximum Minimum Inhibitory Concentration (MIC) against *S. aureus* of 250 µg/mL. A significant negative correlation was observed between the amount of total phenolics and MIC. The results indicate that measuring the concentrations of groups of active compounds, rather than individual components, is an appropriate approach in developing quality standards for propolis.

89. B. Trusheva, D. Trunkova and V. Bankova. Different extraction methods of biologically active components from propolis: a preliminary study. *Chemistry Central Journal*, **1**:13, doi:10.1186/1752-153X-1-13. (2007).

<https://rd.springer.com/content/pdf/10.1186%2F1752-153X-1-13.pdf>

Abstract: Background: Propolis is widely used in apitherapy, preparations, and food and beverage additives. Various extraction techniques were applied in the extraction of the biologically active constituents of poplar type propolis in order to compare their efficiency. The methods employed were: traditional maceration extraction, ultrasound extraction (UE), and microwave assisted extraction (MAE). **Results:** The total amounts of extracted phenolics and flavonoids were determined, and the effectiveness of the methods compared. MAE was very rapid but led to the extraction of a large amount of non-phenolic and non-flavonoid material. UE gave the highest percentage of extracted phenolics. **Conclusion:** Compared to the maceration extraction, MAE and UE methods provided high extraction yield, requiring short timeframes and less labour. UE was shown to be the most efficient method based on yield, extraction time and selectivity.

90. R. O. Orsi J.M. Sforcin, S.R.C. Funari, A. Fernandes-Jr., P. Rodrigues, V. Bankova. Effects of propolis from Brazil and Bulgaria on *Salmonella* serovars. *J. Venom. Anim. Toxins incl. Trop. Dis.* **13**, 4, 748 - 759 (2007)

<http://www.scielo.br/pdf/jvatitd/v13n4/a06v13n4.pdf>

Abstract: Propolis shows biological properties such as antibacterial action. This bee product has a complex chemical composition, which depends on the local flora where it is produced. *Salmonella* serovars are responsible for human diseases that range from localized gastroenteritis to systemic infections. The aim of the present study was to investigate the susceptibility of *Salmonella* strains, isolated from food and infectious processes, to the antibacterial action of Brazilian and Bulgarian propolis, as well as to determine the behavior of these bacteria, according to the incubation period, in medium plus propolis. Dilution of ethanolic extract of propolis in agar was the used method. Brazilian and Bulgarian propolis showed an antibacterial action against all *Salmonella* serovars. The minimal inhibitory concentrations (MIC) of propolis

were similar, although they were collected in different geographic regions. *Salmonella typhimurium*, isolated from human infection, was more resistant to propolis than *Salmonella enteritidis*.

91. V. Bankova, B. Trusheva., M. Popova. New developments in propolis chemical diversity studies (since 2000) In: N. Orsolich & I. Basic (Eds), *Scientific Evidence of Use of Propolis in Ethnomedicine*, 1-13, Transworld Research Network , Trivandrum, 2008.

<http://www.researchgate.net/publication/266111111>

92. V. Bankova. Chemical diversity of propolis makes it a valuable source of new biologically active compounds. *Journal of ApiProduct & ApiMedical Science* 1(2), 23 – 28, 2009.

<http://www.ibrabee.org.uk/component/content/article?layout=edit&id=3220>

Abstract: Propolis with its diverse pharmacological activities and low toxicity has attracted the attention of modern scientists since about 50 years. However, propolis, or bee glue, has demonstrated remarkable chemical variability, which is a serious obstacle to its standardization and consequently, to its official acceptance into the main stream of our healthcare system. On the other hand, the variation in the chemistry of propolis from different ecosystems has made it a source of new biologically active molecules, mainly antioxidative, antibacterial and anticancer agents. The most important recent findings concerning bioactive molecules isolated from propolis are reviewed and discussed here.

93. M. P. Popova, I. B. Chinou, I. N. Marekov, V. S. Bankova. Terpenes with antimicrobial activity from Cretan propolis. *Phytochemistry* 70(10), 1262-1271, 2009.

<http://www.sciencedirect.com/science/article/pii/S0031942209003008#>

Abstract: Five terpenes, the diterpenes: 14,15-dinor-13-oxo-8(17)-labden-19-oic acid and a mixture of labda-8(17),13E-dien-19-carboxy-15-yl oleate and palmitate as well as the triterpenes, 3,4-seco-cycloart-12-hydroxy-4(28),24-dien-3-oic acid and cycloart-3,7-dihydroxy-24-en-28-oic acid were isolated from Cretan propolis. Moreover, 18 known compounds were also isolated, seven of them for the first time as propolis components. All structures were established on the basis of spectroscopic analysis and chemical evidence. All isolated compounds were tested for antimicrobial activity against some Gram-positive and Gram-negative bacteria as well as against some human pathogenic fungi showing a broad spectrum of antimicrobial activity.

94. A.C.H.F. Sawaya, P. V. Abdelnur, M.N. Eberlin, S. Kumazawa, M.-R. Ahn, K. -S. Bang, N. Nagaraja, V. S. Bankova, H. Afrouzan. Fingerprinting of Propolis by Easy Ambient Sonic-Spray Ionization Mass Spectrometry. *Talanta* **81**, 100 – 108, 2010.

<http://www.sciencedirect.com/science/article/pii/S0039914009008923>

Abstract: Chemical profiles of a representative set of 49 propolis ethanolic extracts collected worldwide (North and South America, Europe, Asia and Oceania) were obtained via easy ambient sonic-spray ionization mass spectrometry (EASI-MS). This simple and easily implemented fingerprinting technique analyses directly (without any pre-separation or sample manipulation) a tiny droplet of the ethanolic extract placed on a inert surface under ambient conditions. Data acquisition took about a minute per sample with no substantial sample carry-over. Extraction of propolis with ethanol by using an ultrasonic bath method gave EASI-MS data similar to the traditional maceration method, reducing total analysis time for the crude propolis resin from a week to just ca 1 h. Principal component analysis of the EASI-MS data is shown to group samples according to the plant sources of their resins, which characterizes their geographical origin.

95. B. Tylkowski, B. Trusheva, V. Bankova, M. Giamberini, G. Peev, A. Nikolova. Extraction of biologically active compounds from propolis and concentration of extract by nanofiltration. *Journal of Membrane Science* **348**, 124–130, 2010.

<https://doi.org/10.1016/j.memsci.2009.10.049>

Abstract: This investigation reports results on the extraction of these compounds with an ethanol–water mixture and sequential concentration of the extract by nanofiltration, using two membranes: Starmem™ 122 (polyimide) and Duramem™ 200 (modified polyimide). The content of flavones, flavonols, flavanones, dihydroflavonols as well as total phenolic substances was determined spectrophotometrically in the feeds and permeates of dead-end nanofiltration experiments. Rejections of over 95% were observed and extract concentration of over three times was achieved with the Duramem™ 200 membrane. The membranes behaviour during the process was characterized by environmental scanning electron microscopy, revealing better stability and higher permeability of the modified polyimide membrane. The established conditions for extract concentration could facilitate cross-flow nanofiltration experiments regarding a technology for medicine or nutritional additive production. They could also be utilized in propolis chemistry investigations for a more reliable identification of some individual compounds.

96. M. Popova, K. Graikou, I. Chinou, V. Bankova. GC-MS profiling of diterpene compounds in Mediterranean propolis from Greece. *J. Agric. Food Chem.* **58**, 3167 – 3176, 2010.

<http://pubs.acs.org/doi/abs/10.1021/jf903841k>

Abstract: The objective of this work is to analyze and identify the diterpene compounds in Mediterranean propolis samples from different Greek regions by GC-MS. The chemical composition of six propolis samples was established using previously isolated diterpenes from Cretan propolis as authentic standards for identification, based on mass spectral fragmentation of the TMS derivatives and retention index. More than 30 diterpenes, among which were new propolis constituents, were identified and characterized by means of authentic standards and interpretation of MS fragmentation as well. This is the first detailed profiling of a new type of propolis, rich in diterpenes. The chromatographic and mass- spectral characteristics of the diterpenes identified could be very useful for rapid GC-MS profiling of this propolis type and for revealing its plant sources.

97. M. Popova, Chia-Nan Chen, Pen-Yuan Chen, Chung-Yang Huang, V. Bankova. A Validated Spectrophotometric Method for Quantification of Prenylated Flavanones in Pacific Propolis from Taiwan. *Phytochem. Anal.* **21**, 186–191, 2010.

<https://doi.org/10.1016/j.memsci.2009.10.049>

Abstract: Propolis is a natural product, rich in biologically active compounds. In the recent years the product is extensively used in food and medical applications. This investigation reports results on the extraction of these compounds with an ethanol–water mixture and sequential concentration of the extract by nanofiltration, using two membranes: Starmem™ 122 (polyimide) and Duramem™ 200 (modified polyimide). The content of flavones, flavonols, flavanones, dihydroflavonols as well as total phenolic substances was determined spectrophotometrically in the feeds and permeates of dead-end nanofiltration experiments. Rejections of over 95% were observed and extract concentration of over three times was achieved with the Duramem™ 200 membrane. The membranes behaviour during the process was characterized by environmental scanning electron microscopy, revealing better stability and higher permeability of the modified polyimide membrane. The established conditions for extract concentration could facilitate cross-flow nanofiltration experiments regarding a technology for medicine or nutritional additive production. They could also be utilized in propolis chemistry investigations for a more reliable identification of some individual compounds.

98. B. Trusheva, I. Todorov, M. Ninova, H. Najdenski, A. Daneshmand, V. Bankova. Antibacterial mono- and sesquiterpene esters of benzoic acids from Iranian propolis. *Chemistry Central Journal* **4:8** (2010)

<https://rd.springer.com/content/pdf/10.1186%2F1752-153X-4-8.pdf>

Abstract: Background: Propolis (bee glue) has been used as a remedy since ancient times. Propolis from unexplored regions attracts the attention of scientists in the search for new bioactive molecules. **Results:** From Iranian propolis from the Isfahan province, five individual components were isolated: the prenylated coumarin suberosin **1**, and four terpene esters: tschimgin (bornyl *p*-hydroxybenzoate) **2**, tschimganin (bornyl vanillate) **3**, ferutinin (ferutinol *p*-hydroxybenzoate) **4**, and tefermin (ferutinol vanillate) **5**. All of them were found for the first time in propolis. Compounds **2** - **5** demonstrated activity against *Staphylococcus aureus*. **Conclusions:** The results of the present study are consistent with the idea that propolis from unexplored regions is a promising source of biologically active compounds.

99. A. Petrova, M. Popova, C. Kuzmanova, I. Tsvetkova, H. Naydenski, E. Muli, V. Bankova. New biologically active compounds from Kenyan propolis. *Fitoterapia* **81**(6), 509–514 (2010)

<https://doi.org/10.1016/j.fitote.2010.01.007>

Abstract: From propolis samples from Kenya, two new arylnaphtalene lignans were isolated, tetrahydrojusticidin B **1** and 6-methoxydiphyllin **2**, along with four known phenolic compounds **5–8**, found for the first time in propolis. The structures of the compounds were elucidated based on their spectral properties. The geranylstilbenes **7** and **8** demonstrated antibacterial activity against *S. aureus*, and the geranylflavon macarangin **6** possessed antiradical activity against DPPH radicals.

100. J.M. Sforcin, V. Bankova. Propolis: Is there a potential for the development of new drugs? *Journal of Ethnopharmacology* **133**(2), 253–260 (2011).

<https://doi.org/10.1016/j.jep.2010.10.032>

Abstract: Propolis has plenty of biological and pharmacological properties and its mechanisms of action have been widely investigated in the last years, using different experimental models in vitro and in vivo. Researchers have been interested in the investigation of isolated compounds responsible for propolis action; however, there is lack of clinical research on the effects of propolis.

101. M. Popova, B. Trusheva, D. Antonova, S. Cutajar, D. Mifsud, C. Farrugia, I. Tsvetkova, H. Najdenski, V. Bankova. The specific chemical profile of Mediterranean propolis from Malta. *Food Chemistry* **126**(3), 1431–1435 (2011).

<https://doi.org/10.1016/j.foodchem.2010.11.130>

Abstract: Seventeen Maltese propolis samples were studied by GC–MS after silylation. They exhibited the typical Mediterranean chemical profile, rich in diterpene compounds (18–92% of TIC, GC–MS): 32 individual diterpenes were identified; 22 of them were present in each specimen. The other abundant compound group was that of sugars and sugar derivatives. In some samples, however, another compound group was observed (0–12% of TIC, GC–MS); the corresponding mass spectra were consistent with mono- and sesquiterpenyl esters of substituted benzoic acids. Two new propolis constituents of this group, daucane diterpene esters of hydroxybenzoic acids, were isolated. Their origin is suggested to be *Ferula communis*, as they are taxonomic markers for this species. All propolis samples were active against *Staphylococcus aureus* but only those with high concentrations of terpenyl esters showed antifungal activity against *Candida albicans*. The present results confirm that Mediterranean propolis is a valuable natural product with potential to improve human health.

102. B. Trusheva, M. Popova, E. Budi Koendhori, I. Tsvetkova, C. Naydenski, V. Bankova. Indonesian propolis: chemical composition, biological activity and botanical origin. *Natural Product Research*, **25**(6), 606–613 (2011)

<http://www.tandfonline.com/doi/abs/10.1080/14786419.2010.488235>

Abstract: From a biologically active extract of Indonesian propolis from East Java, 11 compounds were isolated and identified: four alk(en)ylresorcinols (obtained as an inseparable mixture) (**1–4**) were isolated for the first time from propolis, along with four prenylflavanones (**6–9**) and three cycloartane-type triterpenes (**5**, **10** and **11**). The structures of the components were elucidated based on their spectral properties. All prenylflavanones demonstrated significant radical scavenging activity against diphenylpicrylhydrazyl radicals, and compound **6** showed significant antibacterial activity against *Staphylococcus aureus*. For the first time *Macaranga tanarius* L. and *Mangifera indica* L. are shown as plant sources of Indonesian propolis.

103. R. O. Orsi; A. Fernandes Jr; V. Bankova; J. M. Sforcin. Antibacterial effects of Brazilian and Bulgarian propolis and synergistic effects with antibiotics acting on the bacterial DNA and folic acid. *Natural Product Research* **26**(4), 344-9. (2012)

<http://www.tandfonline.com/doi/abs/10.1080/14786411003754355>

Abstract: Propolis is a honeybee product that has been used since ancient times because of its therapeutic effects. It can be used in the development of alternative therapies for the treatment of many diseases, and because propolis shows antibacterial action, this work was carried out in order to investigate a possible synergism between propolis and antibiotics acting on DNA (ciprofloxacin and norfloxacin) and on the metabolism (cotrimoxazole) against *Salmonella* Typhi. Propolis samples collected in Brazil and Bulgaria were compared in these assays, and the synergism was investigated by using $\frac{1}{2}$ and $\frac{1}{4}$ of the minimal inhibitory concentration for

propolis and antibiotics, evaluating the number of viable cells according to the incubation time. Brazilian and Bulgarian propolis showed antibacterial activity, but no synergistic effects with the three tested antibiotics were seen. Previous works by our laboratory have revealed that propolis has synergistic effects with antibiotics, acting on the bacterial wall and ribosome, but it does not seem to interact with antibiotics acting on DNA or folic acid, and only a bacteriostatic action was seen in these assay conditions.

104. R. O. Orsi; A. Fernandes; V. Bankova; J. M. Sforcin. The effects of Brazilian and Bulgarian propolis *in vitro* against *Salmonella Typhi* and their synergism with antibiotics acting on the ribosome. *Natural Product Research* **26**(5), 430–437 (2012)

<http://www.tandfonline.com/doi/abs/10.1080/14786419.2010.498776>

Abstract: *Salmonella enterica* serovar Typhi is the causative agent of typhoid fever in humans, and the use of antibiotics is essential for controlling this infection; however, the excessive use of antibiotics may select resistant strains. Propolis is a honeybee product and its antimicrobial activity has been intensively investigated. Thus, the objective of this study was to investigate a possible synergism between propolis (collected in Brazil and Bulgaria) and antibiotics acting on the ribosome (chloramphenicol, tetracycline and neomycin) against *Salmonella Typhi in vitro*. The synergism was investigated by using $\frac{1}{2}$ and $\frac{1}{4}$ of the minimum inhibitory concentration for propolis and these antimicrobial agents, evaluating the number of viable cells according to the incubation time. Brazilian propolis showed a bacteriostatic action against *S. Typhi*, while Bulgarian propolis showed a bactericidal activity and a synergistic effect with the three antibiotics. Variations in the biological assays might be due to the differences in their chemical compositions. Based on the results, one may conclude that Bulgarian propolis showed an important antibacterial action, as well as a synergistic effect with antibiotics acting on the ribosome, which points out a possible therapeutic strategy evaluating the use of propolis preparations for the treatment of *Salmonella Typhi* infection.

105. M. Popova, B. Trusheva, S. Cutajar, D. Antonova , D. Mifsud, C. Farrugia, V. Bankova. Identification of the plant origin of the botanical biomarkers of Mediterranean type propolis. *Natural Product Communications* **7**(5), 569-570 (2012).

<http://europepmc.org/abstract/med/22799077>

Abstract: *Salmonella enterica* serovar Typhi is the causative agent of typhoid fever in humans, and the use of antibiotics is essential for controlling this infection; however, the excessive use of antibiotics may select resistant strains. Propolis is a honeybee product and its antimicrobial activity has been intensively investigated. Thus, the objective of this study was to investigate a possible synergism between propolis (collected in Brazil and Bulgaria) and

antibiotics acting on the ribosome (chloramphenicol, tetracycline and neomycin) against *Salmonella Typhi* *in vitro*. The synergism was investigated by using ½ and ¼ of the minimum inhibitory concentration for propolis and these antimicrobial agents, evaluating the number of viable cells according to the incubation time. Brazilian propolis showed a bacteriostatic action against *S. Typhi*, while Bulgarian propolis showed a bactericidal activity and a synergistic effect with the three antibiotics. Variations in the biological assays might be due to the differences in their chemical compositions. Based on the results, one may conclude that Bulgarian propolis showed an important antibacterial action, as well as a synergistic effect with antibiotics acting on the ribosome, which points out a possible therapeutic strategy evaluating the use of propolis preparations for the treatment of *Salmonella Typhi* infection.

106. V. Bankova, A. Atanassov, R., Denev, M. Shishinjoiva. Bulgarian bee products and their health-promoting potential. *Biotechnol. & Biotechnol. Eq.* **26**(4), 3086-3088 (2012).

<http://www.tandfonline.com/doi/pdf/10.5504/BBEQ.2012.0001?needAccess=true>

Abstract: This mini-review summarizes the most important results concerning the health-promoting potential of Bulgarian honeys, royal jelly and propolis. Bulgarian honey, propolis and royal jelly have demonstrated remarkable biological activities, such as antibacterial, antiviral, antioxidant, immunostimulating, etc., and deserve further detailed studies. The most promising perspectives for future research and utilization of these valuable Bulgarian bee products are outlined.

107. K. Bilikova, M. Popova, B. Trusheva, V. Bankova. New anti-*Paenibacillus larvae* substances purified from propolis. *Apidologie* **44**, 278 – 285 (2013)

<https://rd.springer.com/content/pdf/10.1007%2Fs13592-012-0178-1.pdf>

Abstract: Propolis plays an important role in the exogenous defense of honeybee colony against pathogens. However, the studies dealing with the activity of propolis against bee pathogens are scarce. Poplar propolis extracts demonstrated promising activity against *Paenibacillus larvae*, the causative agent of American foulbrood. From the same propolis, five individual components and a mixture of caffeates were isolated, and their structures confirmed by spectroscopic data. Among the isolated propolis constituents are flavonoids, ferulic acid esters, and the oxylipin 9-oxo-10(*E*)-12(*Z*)-octadecadienoic acid, newly identified as propolis component. These substances were tested for their activity against *P. larvae* strains. The most active constituents were pinocembrin, 3-*O*-acetyl pinobanksin, and the caffeate mixture. This is the first communication of antimicrobial activity of individual propolis constituents against *P. larvae*; their important advantage is the fact that they are naturally present in the hive.

108. V. Bankova, B. Trusheva. Chemical profiles of different propolis types in relation to their biological activity. In: T. Farooki and A. Farooki, Eds., *Beneficial effects of propolis on human health and chronic diseases*. NOVA Science Publishers, New York. 2013. ISBN: 978-1-62081-440-6
https://www.novapublishers.com/catalog/product_info.php?products_id=33046

Abstract: In the last decade, the paradigm shift in propolis research was completed: the awareness that propolis is a product of variable chemical composition and that there are many propolis types became common knowledge. For this reason and unlike previous review articles, the present review on propolis is organized in agreement with this concept. Recent findings in propolis research are summarized and discussed, concerning chemical profiles, new constituents, biological activity and active principles of different propolis types, as determined by their plant source(s). European poplar propolis, Brazilian green (*Baccharis*) propolis, Brazilian and Cuban red propolis (*Dalbergia*) and other recently discovered propolis types are considered.

109. M. Popova, B. Trusheva, R. Khismatullin, N. Gavrilova, G. Legotkina, J. Lyapunov, V. Bankova. The Triple Botanical Origin of Russian Propolis from the Perm Region, Its Phenolic Content and Antimicrobial Activity. *Natural Product Communications* **8**(5), 617-620 (2013)
https://www.researchgate.net/publication/267030781_The_Triple_Botanical_Origin_of_Russian_Propolis_from_the_Perm_Region_Its_Phenolic_Content_and_Antimicrobial_Activity

Abstract: Russian propolis from the Perm Region was studied by GC-MS, its antibacterial activity was determined, and the bioactive compounds (total phenolics and flavonoids) were quantified by spectrophotometric methods. It was found that Permian propolis belonged to a bee-glue type of triple plant origin: aspen-birch-poplar. Such propolis type has not yet been described. It contains many constituents which could be responsible for diverse biological activities: antibacterial, antifungal, antiviral, antioxidant, anti-inflammatory, and anti-tumor. The combination of the protective excretions of three plant species, resulting in the presence of secondary plant metabolites of many different structural classes gives reason to believe that the pharmacological actions are implemented in different, complementary mechanisms. Together with the detected antibacterial activity, this observation leads to the expectation that such combination could be especially effective in treatment of infections of different origin.

110. B.J.Conti, M.C. Búfalo, M. de Assis Golim, V. Bankova, J.M. Sforcin.
Cinnamic Acid Is Partially Involved in Propolis Immunomodulatory Action

on Human Monocytes. *Evidence-Based Complementary and Alternative Medicine*, Volume 2013, Article ID 109864, 7 pages.

<http://downloads.hindawi.com/journals/ecam/2013/109864.pdf>

Abstract: Propolis is a beehive product used in traditional medicine due to its biological properties. It shows a complex chemical composition including phenolics, such as cinnamic acid (Ci). The mechanisms of action of propolis have been the subject of research recently; however, the involvement of Ci on propolis activity was not investigated on immune cells. Ci effects were evaluated on human monocytes, assessing the expression of Toll-like receptors (TLRs), HLA-DR, and CD80. Cytokine production (TNF- α and IL-10) and the fungicidal activity of monocytes were evaluated as well. Data showed that Ci downregulated TLR-2, HLA-DR, and CD80 and upregulated TLR-4 expression by human monocytes. High concentrations of Ci inhibited both TNF- α and IL-10 production, whereas the same concentrations induced a higher fungicidal activity against *Candida albicans*. TNF- α and IL-10 production was decreased by blocking TLR-4, while the fungicidal activity of monocytes was not affected by blocking TLRs. These results suggest that Ci modulated antigen receptors, cytokine production, and the fungicidal activity of human monocytes depending on concentration, and TLR-4 may be involved in its mechanism of action. Ci seemed to be partially involved in propolis activities.

111. M. Popova, R. Dimitrova, H.T. Al-Lawati, I. Tsvetkova, H. Najdenski, V. Bankova. Omani propolis: chemical profiling, antibacterial activity and new propolis plant sources. *Chemistry Central Journal*, 7: 158 (2013).

<https://ccj.springeropen.com/track/pdf/10.1186/1752-153X-7-158?site=ccj.springeropen.com>

Abstract: Background: Propolis (bee glue) is a resinous honeybee product collected by bees from plants and used as a building material and as a defensive antimicrobial substance in their hives. It has a long history of application in many countries as a traditional remedy for treating wounds, burns, soar throat, stomach disorders, etc. It has been proved to possess beneficial biological effects, including antimicrobial, antioxidant, anti-inflammatory, cytotoxic, antiulcer, and many others. Bees gather propolis from diverse resinous plant parts and in different phytogeographic regions its chemical composition might vary significantly. In this article we report the results of the first study on the chemical profiles of propolis from Oman, its plant origin and antibacterial activity. **Results:** The chemical profiles of Omani propolis extracts were obtained by GC-MS analysis after silylation. Over 50 individual compounds were identified in the samples, belonging to different compound types: sugars, polyols, hydroxy acids, fatty acids, cardanols and cardols, anacardic acids, flavan derivatives, triterpenes, prenylated flavanones and chalcones. The profiles were dissimilar from other known propolis types. They demonstrate that although Oman is not a large country, the plant sources of propolis vary significantly, even in the same apiary and the same season. Based on chemical profiles, and isolation and identification of

major marker compounds (new propolis constituents), new plant sources of propolis were found: *Azadirachta indica* (neem tree) and *Acacia* spp. (most probably *A. nilotica*). The ethanol extracts of the studied propolis samples demonstrated activity against *S. aureus* (MIC < 100 µg. mL⁻¹) and *E. coli* (MIC < 380 µg. mL⁻¹). **Conclusion:** Omani propolis is different from the known propolis types and demonstrates significant chemical diversity. Its most important plant source is the resin of *Azadirachta indica*, and as a result its typical components are C₅-prenyl flavanones. Other plant sources have been identified, too, playing some role in resin collection by bees in Oman: *Acacia* spp. (most probably *A. nilotica*) and *Mangifera indica*. The results demonstrate also the potential of Omani propolis as antimicrobial.

112. M. Popova, M. Reyes. Y. Le Conte, V. Bankova. Propolis chemical composition and honey bee resistance against *Varroa destructor*. *Natural Product Research* **28**(11), 788 – 792, DOI: 10.1080/14786419.2014.881366 (2014).

<http://www.tandfonline.com/doi/pdf/10.1080/14786419.2014.881366?needAccess=true>

Abstract: Propolis is known as honeybee chemical defence against infections and parasites. Its chemical composition is variable and depends on the specificity of the local flora. However, there are no data concerning the relationship between propolis chemical composition and honeybee colony health. We tried to answer this question, studying the chemical composition of propolis of bee colonies from an apiary near Avignon, which are tolerant to *Varroa destructor*, comparing it with colonies from the same apiary which are non-tolerant to the mites. The results indicated that non-tolerant colonies collected more resin than the tolerant ones. The percentage of four biologically active compounds – caffeic acid and pentenyl caffeates – was higher in propolis from tolerant colonies. The results of this study pave the way to understanding the effect of propolis in individual and social immunity of the honeybees. Further studies are needed to clarify the relationship between propolis chemical composition and honeybee colony health

113. V. Bankova, M. Popova, B. Trusheva. Propolis volatile compounds: chemical diversity and biological activity: a review. *Chemistry Central Journal*, **8**: 28 (2014)

<https://ccj.springeropen.com/track/pdf/10.1186/1752-153X-8-28?site=ccj.springeropen.com>

Abstract: Propolis is a sticky material collected by bees from plants, and used in the hive as building material and defensive substance. It has been popular as a remedy in Europe since ancient times. Nowadays, propolis use in over-the-counter preparations, “bio”-cosmetics and functional foods, etc., increases. Volatile compounds are found in low concentrations in propolis,

but their aroma and significant biological activity make them important for propolis characterisation. Propolis is a plant-derived product: its chemical composition depends on the local flora at the site of collection, thus it offers a significant chemical diversity. The role of propolis volatiles in identification of its plant origin is discussed. The available data about chemical composition of propolis volatiles from different geographic regions are reviewed, demonstrating significant chemical variability. The contribution of volatiles and their constituents to the biological activities of propolis is considered. Future perspectives in research on propolis volatiles are outlined, especially in studying activities other than antimicrobial.

114. V. Bankova, A.S. Galabov, D. Antonova, N. Vilhelmova, B. Di Perri. Chemical composition of propolis extract ACF[®] and activity against herpes simplex virus. *Phytomedicine*, 21(11):1432-8 (2014)

<https://doi.org/10.1016/j.phymed.2014.04.026>

Abstract: Propolis Extract ACF[®] (PPE) is a purified extract manufactured from propolis collected in a Canadian region rich in poplar trees, and it is the active substance of a topical ointment used against herpes *labialis* (cold sores or fever blisters). Aim of this study was to analyze the chemical composition of PPE in order to understand the plant origin and possible relations between compounds and antiviral activity, and to characterize the antiviral activity of the extract against herpes simplex virus *in vitro*. **Materials and Methods:** The analysis of the propolis extract samples was conducted by Gas Chromatography–Mass Spectrometry (GC–MS). The antiviral activity was tested against herpes simplex viruses type 1 and type 2 in MDBK cell cultures by treating the cells with PPE at the time of virus adsorption, and by incubating the virus with the extract before infection (virucidal assay). **Results:** Results from the GC–MS analyses revealed a dual plant origin of PPE, with components derived from resins of two different species of poplar. The chemical composition appeared standardized between extract samples and was also reproduced in the sample of topical ointment. The antiviral studies showed that PPE had a pronounced virucidal effect against herpes simplex viruses type 1 and type 2, and also interfered with virus adsorption.

115. M.J.A. Mendes Araújo, M.C. Búfalo, B.J. Conti, A. Fernandes Jr, B. Trusheva, V. Bankova, J.M. Sforcin. The chemical composition and pharmacological activities of geopropolis produced by *Melipona fasciculata* Smith in northeast Brazil. *J Mol Pathophysiol* 4(1), 12-20. (2015)

<https://www.ejmanager.com/fulltextpdf.php?mno=177399>

Abstract: Aim: Geopropolis is produced by stingless bees from resinous materials of plants, adding salivary secretions, wax, mud or clay, and has been used in folk medicine for the treatment of respiratory diseases and dermatoses. Therefore, it is important to study its

antibacterial, antitumor and immunomodulatory properties are important to confirm the ethnopharmacological applications. This work aimed to evaluate its chemical composition, and its antimicrobial, anti-tumoral and immunomodulatory activities. **Materials and Methods:** Geopropolis composition was investigated using GC-MS analysis. Antibacterial tests were performed to determine the minimum inhibitory concentration. HEp-2 cells viability was determined by the reduction of MTT, and cytokine production by human monocytes was determined by ELISA. **Results:** The major constituents of geopropolis compounds were carbohydrates and their derivatives, triterpenes, anacardic acid, alkylresorcinols, and sugar alcohols. Geopropolis alone showed no antibacterial activity against *Staphylococcus aureus* and *Escherichia coli*, but its combination with chloramphenicol exerted a greater action against *S. aureus* than chloramphenicol alone. Geopropolis exhibited a cytostatic action toward human laryngeal epidermoid carcinoma cells and stimulated tumor necrosis factor alpha and interleukin-10 production by human monocytes, showing an activator profile for human monocytes. **Conclusion:** The synergistic effect of geopropolis and chloramphenicol deserves further investigation due to bacterial resistance to antibiotics. Geopropolis also displayed antitumoral and immunomodulatory activity, and its biological properties may be due to triterpenes - one of its major chemical constituents.

116. M.A. Savka, L. Dailey, M. Popova, R. Mihaylova, B. Merritt, M. Masek, P. Le, S.R. Mat Nor, M. Ahmad, A.O. Hudson, V. Bankova. Chemical composition and disruption of quorum sensing signaling in geographically diverse United States propolis. *Evidence-Based Complementary and Alternative Medicine*, vol. 2015, Article ID 472593, 10 pages, 2015. doi:10.1155/2015/472593

<http://downloads.hindawi.com/journals/ecam/2015/472593.pdf>

Abstract: Propolis or bee glue has been used for centuries for various purposes and is especially important in human health due to many of its biological and pharmacological properties. In this work we showed quorum sensing inhibitory (QSI) activity of ten geographically distinct propolis samples from the United States using the acyl-homoserine lactone- (AHL-) dependent *Chromobacterium violaceum* strain CV026. Based on GC-MS chemical profiling the propolis samples can be classified into several groups that are as follows: (1) rich in cinnamic acid derivatives, (2) rich in flavonoids, and (3) rich in triterpenes. An in-depth analysis of the propolis from North Carolina led to the isolation and identification of a triterpenic acid that was recently isolated from Hondurian propolis (Central America) and ethyl ether of p-coumaric alcohol not previously identified in bee propolis. QSI activity was also observed in the second group US propolis samples which contained the flavonoid pinocembrin in addition to other flavonoid compounds. The discovery of compounds that are involved in QSI activity has the potential to facilitate studies that may lead to the development of antivirulence

therapies that can be complementary and/or alternative treatments against antibiotic resistant bacterial pathogens and/or emerging pathogens that have yet to be identified.

117. A.C. de Groot, M. Popova, V. Bankova. An update on the constituents of poplar-type propolis. Wapserrveen, the Netherlands: acdegroot publishing, 2014. ISBN/EAN: 978-90-813233-0-7.
https://www.researchgate.net/profile/Anton_De_Groot2/publication/262851225_AN_UPDATE_ON_THE_CONSTITUENTS_OF_POPLAR-TYPE_PROPOLIS/links/0deec5390423cf10a6000000.pdf

118. S. Sanpa, M. Popova, V. Bankova, T. Tunkasiri, S. Eitssayeam, P. Chantawannakul. Antibacterial compounds from propolis of *Tetragonula laeviceps* and *Tetrigona melanoleuca* (Hymenoptera: Apidae) from Thailand. *PLOS ONE* 10(5): e0126886. DOI:10.1371/journal.pone.0126886; May 18, 2015
<http://journals.plos.org/plosone/article/file?id=10.1371/journal.pone.0126886&type=printable>

Abstract: This study investigated the chemical composition and antimicrobial activity of propolis collected from two stingless bee species *Tetragonula laeviceps* and *Tetrigona melanoleuca* (Hymenoptera: Apidae). Six xanthonenes, one triterpene and one lignane were isolated from *Tetragonula laeviceps* propolis. Triterpenes were the main constituents in *T. melanoleuca* propolis. The ethanol extract and isolated compounds from *T. laeviceps* propolis showed a higher antibacterial activity than those of *T. melanoleuca* propolis as the constituent α -mangostin exhibited the strongest activity. Xanthonenes were found in propolis for the first time; *Garcinia mangostana* (Mangosteen) was the most probable plant source. In addition, this is the first report on the chemical composition and bioactivity of propolis from *T. melanoleuca*.

119. B.J. Conti, V. Bankova, J.M. Sforcin. Chemical composition of the same Brazilian propolis sample analyzed in 1997 and in 2012: No freezing effect. *Natural Product Communications* 10(7), 1279 – 1280 (2015).
<http://europepmc.org/abstract/med/26411030>

Abstract: The same propolis sample that was analyzed by GC-MS in 1997 and 2012 was examined again after being frozen for 15 years. No qualitative changes were seen in its composition; artepilin C, prenyl-p-coumaric acid, dihydrocinnamic acid, caffeic acid were the major compounds.

120. K. Graikou, M. Popova, O. Gortzi, V. Bankova, I. Chinou.
Characterization and biological evaluation of selected Mediterranean

propolis samples. Is it a new type? *LWT - Food Science and Technology* **65**, 261 - 267 (2016)

<https://doi.org/10.1016/j.lwt.2015.08.025>

Abstract: Chemical composition, antioxidant and antimicrobial activity of thirty two (32) selected Mediterranean propolis extracts from mainland Greece, Greek islands, Cyprus, Croatia, and Algeria were determined. Chemo-geographical patterns within Mediterranean propolis were further analyzed by chemometrics. Knowledge of propolis composition, which depends on the geographical and climatic origin and its biological properties from different geographic regions samples, is extremely valuable with respect to the problem of propolis standardization. This work concluded that the Greek propolis samples share characteristics that differentiate them from typical European propolis, like the presence of diterpenes in significant amounts and the relatively low quantity of phenolic acid esters and besides their potential pharmaceutical and nutraceutical value, they are also attractive candidates for use as natural antioxidant and microbicidal additives in food systems.

121. M. Popova, B. Lioussi, S. Aazza, D. Antunes, V. Bankova, G. Miguel. Antioxidant and α -glucosidase inhibitory properties and chemical profiles of Moroccan propolis. *Natural Product Communications*, **10**(11), 1961 - 1964 (2015).

<http://europepmc.org/abstract/med/26749837>

Abstract: The chemical profiles of propolis hydroalcoholic extracts from different regions of Morocco were studied by gas chromatography coupled to mass spectrometry after silylation. Samples from Khamissat and Imouzzar belong to the poplar type, as typical poplar flavonoids predominated. Propolis from Rabat also contained high percentage of flavonoids, but had significantly lower amount of phenolic acid esters and contained no pinobanksin-3-O-acetate. Propolis from Bhalil demonstrated a profile rich in diterpenes (74.3%), and is a typical Mediterranean propolis sample. All samples had the capacity for inhibiting glucosidase and amylase enzymes, as well as the capacity for scavenging free radicals and preventing lipid peroxidation. Both activities were significantly higher in the flavonoids-rich samples.

122. B. Trusheva, K. Stancheva, N. Gajbhiye, R. Dimitrova, M. Popova, R. Saraf, V. Bankova. Two new prenylated stilbenes with irregular sesquiterpenyl side chain from propolis from Fiji Islands. *Records of Natural Products* **10**(4), 465 – 471 (2016).

<http://www.acgpubs.org/RNP/2016/Volume10/Issue%201/56-RNP-1507-128.pdf>

Abstract: Two new prenylated stilbenes with an irregular sesquiterpenyl side chain, solomonin B (1) and solomonin C (2), together with four known compounds, glyasperin A (3), isolated for the first time from propolis, kumatakenin (4), macarangin (5) and mangiferolic acid (6) were isolated from ethanol extract of propolis from Fiji islands. The compounds structures were determined based on their spectral data analysis (1D and 2D NMR, UV and HREIMS) and comparison with literature data. The chemical composition of propolis from Fiji islands was determined for the first time.

123. P. Petrov, C. B. Tsvetanov, P. Mokreva, K. Yoncheva, S. Konstantinov, B. Trusheva, M. Popova, V. S. Bankova. Novel micellar form of poplar propolis with high cytotoxic activity. *RSC Adv.* **6**, 30728-30731 (2016). DOI: 10.1039/C6RA03577A

[http://pubs.rsc.org/-
/content/articlelanding/2016/ra/c6ra03577a/unauth#!divAbstract](http://pubs.rsc.org/-/content/articlelanding/2016/ra/c6ra03577a/unauth#!divAbstract)

Abstract: A novel micellar form of propolis based on a biocompatible poly(ethylene oxide)-*block*-poly(propylene oxide)-*block*-poly(ethylene oxide) *block* copolymer (PEO₂₆PPO₄₀PEO₂₆) is developed. Beneficially, all biologically active lipophilic constituents of propolis are solubilized with the aid of polymeric micelles in aqueous media. The micellar form of the propolis exhibits high *in vitro* cytotoxicity at relatively low concentrations.

124. V. Bankova, M. Popova, B. Trusheva. New emerging fields of application of propolis. *Macedonian Journal of Chemistry and Chemical Engineering* **35**(1), 1 -11, DOI: 10.20450/mjcce.2016.864 (2016)

[http://www.mjcce.org.mk/index.php/MJCCE/article/view/mjcce.2016.864/
489](http://www.mjcce.org.mk/index.php/MJCCE/article/view/mjcce.2016.864/489)

Abstract: Propolis (bee glue) is a sticky resinous material applied by honey bees *Apis mellifera* L. as a building material in their hives and as a defensive substance against infections. Propolis has been used as a remedy in traditional medicine systems all over the world, mainly to treat wounds and burns, sore throat, stomach ulcer, etc. Modern science has confirmed the antimicrobial and antiviral action of propolis, and has discovered numerous other beneficial pharmacological properties of bee glue: immunomodulating, anti-inflammatory, antiobesity, antitumor, and many others. For this reason, significant number of products containing propolis have been developed and commercialised: medical devices, over-the-counter preparations, health foods and beverages, cosmetics. This review is not dealing with propolis applications in improvement and protection of human health. Instead, it is focused on some new and promising areas of propolis use and innovative propolis containing products, emerging in the last few years: improving growth performance of livestock, food preservation, food packaging, textile materials for biomedical application, etc.

125. V. Bankova, M. Popova, B. Trusheva. Latest Developments in Propolis Research: Chemistry and Biology. In: S.M. Cardoso, A.M.S. Silva, (Eds.) *Chemistry, Biology and Potential Applications of Honeybee Plant-Derived Products*. Bentham E-books, pp. 45 – 66, 2016. ISBN: 978-1-68108-238-7, eISBN: 978-1-68108-237-0, 2016; DOI: 10.2174/97816810823701160101
<https://ebooks.benthamscience.com/book/9781681082370/chapter/142959/>

Abstract: Propolis is a plant derived bee product which serves dual purposes in the honeybee colony: building material and protective substance. Propolis has been used as remedy in the traditional medicine of numerous nations, because of its antimicrobial, antioxidant, and many other beneficial pharmacological actions. In this chapter, the results of the newest (in the last 5 years) chemical studies of propolis from different geographic and plant origin are reviewed, together with the new identified source plants: 152 new constituents of propolis, being 57 new chemical entities, and 12 new chemical types of propolis are listed. The importance of propolis for the bee colony is discussed, with special attention to the activity of propolis and its constituents against bee pathogens and parasites. The review of recent propolis literature demonstrates its potential to serve as a source of new chemical structures and new bioactive compounds due to its chemical diversity. It also reveals the potential of propolis to be used for development of innovative products, mainly in the field of food industries, animal husbandry, and beekeeping. For this to happen, the combined efforts of researchers and technologists from different areas are necessary, in order to make better use of bee glue.

126. A.N. Tamfu, M.E.C. Domgnim, E. Talla, P.V. Tan, T.J. Mbafor, M. Popova, V. Bankova. Chemical constituents and anti-ulcer activity of propolis from the North-West of Cameroon. *Research Journal of Phytochemistry* DOI: 10.3923/rjphyto.2016 (2016)
<http://scialert.net/qredirect.php?doi=rjphyto.2016.45.57&linkid=pdf>

Abstract: Three extracts of propolis harvested from Nkambe, North-West region of Cameroon were characterized by GC-MS analysis and their gastric cytoprotective, antisecretory and antioxidant properties evaluated using experimentally-induced gastric ulcers in rats. The propolis extracts were rich in phytoconstituents occurring as sugars, triterpenes and a mixture (**fatty acids**+triterpenes+alkenyl resorcinols) in the methanol, hexane and acetone extracts, respectively with *Mangifera indica* as major plant source. Three triterpenes, lupeol, lupenone, 27-hydroxymangiferonic acid and an ester of **fatty acid** heptadecyl butanoate were isolated and characterized. The methanol, acetone and hexane extracts (200-600 mg kg⁻¹) dose-dependently prevented the formation of ethanol-induced gastric lesions (percentage of inhibition, 61, 54 and

55%, respectively, at the dose of 600 mg kg⁻¹). Increasing doses of the extracts inhibited pylorus ligation-induced lesions by 64.5, 73.1 and 16.2%, respectively for the highest dose but none of them showed antisecretory activity compared with controls. The most further significantly ($p < 0.01$) reduced HCl/ethanol-induced ulcer indices from 4.33±0.32 in cytoprotective (acetone) extract (56.6-73.1% inhibition under highly acidic gastric environments), the controls to 1.25±0.53 and 0.6±0.04 at the dose of 400 and 600 mg kg⁻¹, respectively (percentage of inhibition: 71-86%). Furthermore, upon pretreatment of the rats with indomethacin prior to HCl/ethanol, the acetone extract significantly ($p < 0.001$) decreased ulcer index from 5.55±0.73 in the controls to 1.89±0.15 at the dose of 600 mg kg⁻¹. Although pretreatment with indomethacin reduced the protective effect of the acetone extract by 23-27% and cytoprotection remained high (62-66% inhibition). The cytoprotective action of the most active (acetone) extract may involve the mediation of endogenous prostaglandin

127. S. El-Guendouz, S. Aazza, B. Lyoussi, V. Bankova, J.P. Lourenço, A.M. Rosa Costa, J.F. Mariano, M.G. Miguel, M.L. Faleiro. Impact of biohybrid magnetite nanoparticles and Moroccan propolis on adherence of Methicillin Resistant Strains of *Staphylococcus aureus*. *Molecules*, **21**(9), 1208; doi:10.3390/molecules21091208 (2016)

<http://www.mdpi.com/1420-3049/21/9/1208/pdf>

Abstract: Biofilm bacteria are more resistant to antibiotics than planktonic cells. Propolis possesses antimicrobial activity. Generally, nanoparticles containing heavy metals possess antimicrobial and antibiofilm properties. In this study, the ability of adherence of Methicillin Resistant Strains of *Staphylococcus aureus* (MRSA) to catheters treated with magnetite nanoparticles (MNPs), produced by three methods and functionalized with oleic acid and a hydro-alcoholic extract of propolis from Morocco, was evaluated. The chemical composition of propolis was established by gas chromatography mass spectrometry (GC-MS), and the fabricated nanostructures characterized by X-ray diffraction (XRD), transmission electron microscopy (TEM), Mossbauer spectroscopy and Fourier transform infrared spectroscopy (FTIR). The capacity for impairing biofilm formation was dependent on the strain, as well as on the mode of production of MNPs. The co-precipitation method of MNPs fabrication using Fe³⁺ and Na₂SO₃ solution and functionalized with oleic acid and propolis was the most effective in the impairment of adherence of all MRSA strains to catheters ($p < 0.001$). The adherence of the strain MRSA16 was also significantly lower ($p < 0.001$) when the catheters were treated with the hybrid MNPs with oleic acid produced by a hydrothermal method. The anti-MRSA observed can be attributed to the presence of benzyl caffeate, pinocembrin, galangin, and isocupressic acid in propolis extract, along with MNPs. However, for MRSA16, the impairment of its adherence on catheters may only be attributed to the hybrid MNPs with oleic acid, since very small amount, if any at all of propolis compounds were added to the MNPs.

128. V. Bankova, D. Bertelli, R. Borba, B.J. Conti, I.B. da Silva Cunha, C. Danert, M.N. Eberlin, S.I. Falcão, M.I. Isla, M.I. Nieva Moreno, G. Papotti, M. Popova, K. Basso Santiago, A. Salas, A.C.H. Frankland Sawaya, N.V. Schwab, J.M. Sforcin, M. Simone-Finstrom, M. Spivak, B. Trusheva, M. Vilas-Boas, M. Wilson, C. Zampini. Standard methods for *Apis mellifera* propolis research. *Journal of Apicultural Research*, (2016) DOI: 10.1080/00218839.2016.1222661

<http://www.tandfonline.com/doi/pdf/10.1080/00218839.2016.1222661?nedAccess=true>

Abstract: Propolis is one of the most fascinating honey bee (*Apis mellifera* L.) products. It is a plant derived product that bees produce from resins that they collect from different plant organs and with which they mix beeswax. Propolis is a building material and a protective agent in the bee hive. It also plays an important role in honey bee social immunity, and is widely used by humans as an ingredient of nutraceuticals, over-the-counter preparations and cosmetics. Its chemical composition varies by geographic location, climatic zone and local flora. The understanding of the chemical diversity of propolis is very important in propolis research. In this manuscript, we give an overview of the available methods for studying propolis in different aspects: propolis in the bee colony; chemical composition and plant sources of propolis; biological activity of propolis with respect to bees and humans; and approaches for standardization and quality control for the purposes of industrial application.

129. E. Talla, A.N. Tamfu, I. S. Gade, L. Yanda, J.T. Mbafor, S. Laurent, L. Vander Elst, M. Popova, V. Bankova. New mono-ether of glycerol and triterpenes with DPPH radical scavenging activity from Cameroonian propolis. *Natural Product Research*, 31(12), 1379-1389, (2017)

<http://www.tandfonline.com/doi/pdf/10.1080/14786419.2016.1253077?nedAccess=true>

Abstract: The extracts of some propolis samples were analysed by GC-MS and then purified by column chromatography. The latter led to the isolation of a new mono-ether of glycerol, 1'-O-eicosanyl glycerol and a new triterpene, methyl-3 β ,27-dihydroxycycloart-24-en-26-oate together with known triterpenoids namely betulin, 3 β -hydroxy lanostan-9,24-dien-21-oic acid, mangiferonic acid, a mixture of ambolic acid and β -sitosterol, 3 β -hydroxycycloartan-12,24(25)-diene and 27-hydroxymangiferonic acid. The DPPH radical scavenging potential of some extracts and compounds were measured. The radical scavenging activity varied from Hexane extract of Fouban propolis (IC₅₀ = 5.6 mg/mL) to Methanol extract of Fouban propolis (IC₅₀ = 1.07 mg/mL) for the extracts and from 3 β -hydroxy lanostan-9,24-dien-21-oic acid

(IC₅₀ = 1.22 mg/mL) to 1'-O-eicosanyl glycerol (IC₅₀ = 0.93 mg/mL) for the compounds. Activities of samples were moderate as they remained closer to those of the standard antioxidants Gallic acid (IC₅₀ = 0.30 mg/mL) and vitamin C (IC₅₀ = 0.80 mg/mL), especially 1'-O-eicosanyl glycerol, the most active compound.

130. B. Trusheva, D. Ivanova, M. Popova, V. Bankova. Insights into the essential oil compositions of Brazilian red and Taiwanese green propolis. *Natural Product Communications*, **12** (2), 197 - 200 (2017).

https://www.researchgate.net/publication/318429251_Insights_into_the_Essential_Oil_Compositions_of_Brazilian_Red_and_Taiwanese_Green_Propolis

Abstract: The objective of the present study was to characterize chemically the essential oils of two distinct propolis types: Brazilian red and Taiwanese green. Unlike the non-volatile chemical composition of these types of propolis, which has been extensively studied, the knowledge of the essential oils is scarce or even not investigated. The essential oils were obtained by hydrodistillation of raw propolis samples using a Likens-Nickerson type apparatus and then analyzed by GC/MS. The main volatile components of Brazilian red propolis were the phenylpropanoids: elemicin (26.1-27.5%), methyl eugenol (16.3-23.8%), transmethyl isoeugenol (9.2-11.6%), isoelemicin (6.1-7.1%) and trans-anethole (4.4-7.1%), while the major constituents of Taiwanese green propolis essential oil were: β -eudesmol (13.9%), 6-methyl-3,5-heptadiene-2-one (12.2%), γ -eudesmol (4.4%), geranial (4.1%) and 6-methyl-5-heptene-2-one (3.7%).

131. S. Sanpa, M. Popova, T. Tunkasiri, S. Eitssayeam, V. Bankova, P. Chantawannakul. Chemical profiles and antimicrobial activities of Thai propolis collected from *Apis mellifera*. *Chiang Mai J. Sci.* 44(2), 438-448 (2017)

<http://www.thaiscience.info/journals/Article/CMJS/10985627.pdf>

Abstract: The aim of this study was to examine the antimicrobial properties and chemical composition of Thai *Apis mellifera* propolis from different locations. All propolis samples demonstrated significant activity (Minimal Inhibitory Concentration < 1000 μ g/mL) against Gram-positive bacteria *Bacillus cereus*, *Staphylococcus aureus*, *S. epidermidis* and methicillin-resistant *Staphylococcus aureus* (MRSA) whereas the activities against Gram-negative bacteria and yeasts were lower. There were statistically significant differences ($p < 0.001$) between the Thai propolis extracts due to their DPPH free radical scavenging activity and total phenolic contents. The GC/MS chemical profiles of all the propolis samples demonstrated similar composition but different proportion of sugars and sugar derivatives, triterpenes and phenolic lipids. From the most active sample (Phayao), several triterpenes and three inseparable mixtures of phenolic lipids (cardols, cardanols, and anacardic acids) were isolated by chromatographic

methods and they showed high antibacterial activities. This indicates that propolis from the studied regions belongs to the tropical propolis type, originating mainly from mango (*Mangifera indica*). Our results provide the information that is useful for future standardization of Thai propolis.

132. M. Popova, B. Trusheva, V. Bankova. Content of biologically active compounds in Bulgarian propolis: a basis for its standardization. *Bulgarian Chemical Communications*, **49**(Special Edition B), 115 – 120 (2017).

http://www.bcc.bas.bg/BCC_Volumes/Volume_49_Special_B_2017/BCC_2017-49-SE-B-115-120.pdf

Abstract: In Bulgaria, propolis tincture is among of the most popular home-made remedies. Bulgarian propolis has been studied and it was found to originate from the bud resin of the black poplar *Populus nigra* L.; the chemical constituents responsible for its biological activity are flavonoid aglycones (flavones, flavonols, flavanones, dihydroflavonols), substituted cinnamic acids and their esters. However, the specific quantitative characteristics of Bulgarian poplar propolis have not been studied. Validated spectrophotometric procedures were used to quantify the three main groups of bioactive substances: total phenolics, total flavones/flavonols, total flavanones/dihydroflavonols, in 22 samples of Bulgarian propolis from different regions of the country. Based on the results, we characterized raw poplar propolis in terms of minimum content of its bioactive components (antimicrobial and antioxidant) as follows: 46% resin, 24% total phenolics, 7% total flavones/flavonols; 5.4% total flavanones/dihydroflavonols. These values can be used as a basis for Bulgarian propolis standard. They are somewhat higher than the ones suggested by the International Honey Commission for poplar type propolis. This is a proof that Bulgarian propolis is a valuable bee product of high quality, higher than that of the average poplar propolis samples coming from other regions.

133. M. Popova, E. Giannopoulou, K. Skalicka-Woźniak, K. Graikou, J. Widelski, V. Bankova, H. Kalofonos, G. Sivolapenko, K. Gaweł-Bęben, B. Antosiewicz, I. Chinou. Characterization and biological evaluation of propolis from Poland. *Molecules* **22**, 1159 (2017)

<http://www.mdpi.com/1420-3049/22/7/1159/htm>

Abstract: In this study, we assessed the therapeutic potential of propolis from Poland and performed chemical analysis by GC–MS, as well as determined its botanical origin. Chemical constituents typical for bud exudates of *Populus nigra* (section Aigeiros) were determined, however, glycerol esters of phenolic acids, as well as unusually high amounts of p-coumaric and ferulic acid and their benzyl esters, were also detected. These constituents are characteristic for buds of *Populus tremula* (section Leuce). We also evaluated the antiproliferative effect of

propolis extracts against nine human cancer cell lines. Additionally, promising antibacterial activity of the dichloromethane extract (Minimal Inhibitory Concentration MIC values of 0.95–1.24 mg/mL), as well as a moderate antifungal activity (MIC values of 1.25–1.40 mg/mL), was noticed. Propolis from Poland appeared as a rich source of antibacterial and antiproliferative compounds and this confirmed that it is a valuable natural product with the potential to improve human health.

134. M. Popova, D. Antonova, V. Bankova. Chemical composition of propolis and American foulbrood: Is there any relationship? *Bulgarian Chemical Communications*, **49**(Special Edition D), 171 – 175 (2017).

https://www.google.bg/url?sa=t&rct=j&q=&esrc=s&source=web&cd=1&cad=rja&uact=8&ved=0ahUKEwiJ76m1rsLWAhVmYpoKHUXYAQUQFggrMAA&url=http%3A%2F%2Fwww.bcc.bas.bg%2FBCC_Volumes%2FVolume_49_Special_D_2017%2FBCC2017-49-SE-D-171-175.pdf&usg=AFQjCNGdVw2YgFqhlq-x8RGDo9wBtsNWKw

Abstract: American foulbrood (AFB) is the most destructive brood diseases of honeybees, causing significant losses to beekeepers. Propolis is an important element of the bee colony social immunity and has demonstrated activity against the causative agent of AFB *Paenibacillus larvae*. However, knowledge on the relationship between propolis chemical composition and the health of the bee colony is still scarce. We studied by GC-MS the chemical profiles of propolis samples from healthy bee colonies and colonies with clinical symptoms of AFB. Healthy colonies produced propolis with higher content of balsam. Although the qualitative composition of all samples was the same, there were quantitative differences: propolis from healthy colonies contained much higher levels (statistically significant, $p < 0.01$) of ferulic acid and coniferyl benzoate, than the propolis from colonies with AFB. Our results are only preliminary, further research should be performed to clarify whether these differences are indeed related to the health of the colonies.

135. P.D. Petrov, G. Grancharov, V. Gancheva, B. Trusheva, V. Bankova, C.o B. Tsvetanov, Development of propolis-loaded block copolymer micelles of superior structural stability and high loading capacity, *Polymer*, **125**, 102-109 (2017)

<http://www.sciencedirect.com/science/article/pii/S0032386117307681>

Abstract: We report on the efficient loading of poplar propolis into core-shell micelles based on amphiphilic poly(ethylene oxide)-*block*-poly(*n*-butyl acrylate) (PEO-*b*-PnBA) diblock copolymers. The copolymers comprising a hydrophilic PEO block of constant length (113 monomer units) and a soft hydrophobic PnBA block of three different lengths (53, 99 and 157

monomer units) were synthesized by atom transfer radical polymerization (ATRP) of n-butyl acrylate initiated by PEO bromoisobutyrate. The study focused on fabrication of stable aqueous colloid systems characterized by complete encapsulation of all bioactive lipophilic propolis constituents within the micellar carrier. The effects of copolymer/propolis mass ratio, copolymer composition and concentration on system characteristics were examined by dynamic light scattering (DLS), atomic force microscopy (AFM) and turbidity measurements. The results revealed that stable aqueous colloids and complete encapsulation of the lipophilic bioactive compounds into the micelles can be achieved at twofold excess of copolymer to propolis amount.