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THE IMPACT OF POLLEN ON THE HEALTH STATUS OF ANIMALS AND HUMANS

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Abstract

Pollen is collected by worker bees from different plant species. This is a special product, rich in biologically active substances, that comprises food for larvae in bee colony. Chemically, it consists of approx. 250 chemical compounds, especially A, B, C, D and E vitamins, proteins, amino acids, fatty acids, as well as flavonoids, sterols, simple and complex sugars, and micro- and macronutrients such as potassium, calcium, magnesium, phosphorus, iron. Due to very diverse chemical composition and properties, pollen has been used for many years in natural medicine, but may be an allergen due to its composition. Standardized extracts of pollen positively affect the immune system, which consists of the activity of white blood cells and other forms of transmitters functioning in systemic blood and lymph. However the cross intake with drugs for these diseases shoud be carried out with precautions to avoid serious interactions.

Introduction and objective

In botanical terms, pollen is composed of male reproductive cells, produced by the plant in anthers (part of the stamens). This product, in the form of pollen load (bee pollen) or bee bread (stored inside the hive and add with honey) is the base food for the offspring (larvae) in the bee colony. To use bee pollen as food or as a complement in the diet is most valuable when it comes from as many various plant species as possible given, like this, the major variety of compounds. Its chemical composition depends on the season, in connection with the flowering phases specific to various plant species as well as on the soil fertility, the soil moisture and the

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weather conditions during anthers' formation and maturation. While collecting pollen, worker bees mix it with a small amount of the secretion from their salivary glands. Then, pollen is put in baskets, located on the third pair of legs, and transported to the hive. Finally, pollen is placed in the comb cells (MALERBO-SOUZA 2011).

Pollen (bee pollen) in the form of the so-called pollen load or bee bread is successfully used in apitherapy and alternative medicine because of its diverse composition and chemical properties. As a dietary supplement it is an important alternative solution to standard treatments for many diseases (YILDIZ et al. 2013, DENISOW et al. 2016, KUMARI et al. 2016, BAZMANDEGAN et al 2017). However, until now the scientific data available is not enough to consider this product as safe to be considering a drug. Increasingly, doctors, patients, as well as animal owners, acknowledge diets based on natural products and bee pollen follows that tendency because the traditional use in certain countries. A number of positive effects of pollen have been proven so far, effective against fungal, bacterial and viral diseases, as well as anti–inflammatory and immune-stimulating properties (KROYER et al. 2001, ALMARAZ-ABARCA et al. 2004, KOCOT et al. 2018). Nevertheless, in the majority of the cases, the activity is dependent of the floral source and the quality control of the product wil be the only way to assure that correspondance with the potential bioactivity. Propolis and bee pollen extracts are used instead of the raw substance due to the fact that they contain higher amounts of bioactive components. The properties of the extract depend strongly not only on the solvent used but also on extraction conditions, that is, time and temperature as well (KIM et al. 2015, DENISOW et al. 2016, PASUULETTI et al. 2017) . Pollen is collected from various species of plants by worker bees and the selection of the species involved in the mixtures collected are difficult. This natural product is rich in biologically active substances and more research should be carried out to assure the efficacy, safety and quality control required for the World Health Organization. In apitherapy, a way of treat peoplewith bee products such as honey, propolis, pollen or bee venom are used in the treatment and prevention of various diseases. These products should be safe as all the others used in medicine. (KOMOSIŃSKA-VASSEV et al. 2015, OLCZYK et al. 2016). Many scientific studies have confirmed a wide spectrum of pollen properties, for example: nourishing, detoxification, antisclerotic, antibiotic and anti–inflammatory (MOREIRA et al. 2013, WEI et al. 2017, KOCOT et al. 2018). However, to complete a full analysis a scarce essays were carried out to assure the safety of the product despite various countries have already legislation to assure the quality control of the product in the market

Description of the state of knowledge

In chemical terms, pollen contains approx. 250 substances and chemical compounds, especially B vitamins (thiamine, riboflavin, niacin, pantothenic acid, pyridoxine, inositol, cyanocobalamin) and pro-vitamin A in the form of β-carotene, ascorbic acid (vit. C), calciferol (vit. D) and tocopherol (vit. E), 22.7% protein (albumin, globulin, glutelin, prolamin) and protein enzymes, 10.4% amino acids (aspartic acid, threonine, serine, glutamic acid, proline, glycine, alanine, valine, methionine, isoleucine, leucine, tyrosine, phenylalanine, histidine, lysine, arginine, cystine and tryptophan), fatty acids (saturated acids: myristic, palmitic, stearic, arachidic and unsaturated fatty acids: oleic acid, linoleic and α-linolenic acid) and flavonoids 1.4% (mainly kaempferol, quercetin and isorhamnetin), sterols, simple and complex sugars and micro- elements such as potassium, calcium, magnesium, phosphorus, iron, sodium and zinc (Rzepecka-Stojko et al. 2015, Panche et al. 2016). Phospholipids represent 1.5%, phytosterols 1.1% and phenolic compounds 1.6%. The detailed chemical composition of pollen depends on the plant type, geographical origin, climate conditions, soil type and even the breed of bees (Nougeira et al. 2012, Silva et al. 2014). Furthermore, the color, shape (round, cylindrical, triangular) and weight of grains of pollen depend on the plant species from which it is collected (Almeida-Muradian et al. 2005, Arraez et al. 2007, Campos et al. 2003, 2008, Kędzia et al. 2005, Khalil et al. 2010, Leja et al. 2012, Saric et al. 2009, Shubharani et al. 2013, Xu et al. 2009). Some studies have shown that the content and properties of bee pollen are dependent on the kind of its plant source and the conditions of the plants growing for example like soil and climate (Feas et al. 2012, Pascoal et al. 2014).

Research has confirmed a broad spectrum of pollen properties. For example, pollen as a potential as analgesic agent thanks to the content of ethanol extracts obtained from a pine species growing in Korea (*Pinus densiflora*), Choi et al. (2007), have demonstrated its antinociceptive effects. They administered intragastrically to mice, these extracts in a dose of 100 and 200 mg/kg b.w. an hour before testing for analgesic activity. They have proved that these extracts were characterized by potent antinociceptive properties comparable with aminopyrine (a standard painkiller).

Moreover, anti-angiogenic properties of pollen were study. The angiogenesis is a process of formation of blood vessels from a primary endothelialand increases the supply of nutrients, growth factors and molecular oxygen to sites damaged or renewed during such processes as pregnancy, menses, wound healing and revascularization of ischemic tissues. How-

ever, an excessive angiogenesis (neovascularization) is characteristic of rheumatoid arthritis, atherosclerosis or retinal vascularization (IZUTA et al. 2009). IZUTA et al. (2009) considered the effect of the pollen of two plants: *Cistus ladanifer* and *C. albidus* (grey-leaved cistus) on angiogenesis of endothelial cells of human umbilical vein (HUVEC). They found that the ethanolic extract is characterized by potent anti-angiogenic properties, i.e. inhibiting angiogenesis. They also found that humans receiving the pollen showed a 30% reduction of the ability of blood platelets to aggregate and a reduction of the level of lipids and cholesterol from 30 to 20%. This therapeutic potential of *C, ladanifer* and *C. albidus* bee pollen should be study in further research, however the hemorrhagic side effect due to the reducing of platelets aggregation need to be controlled.

SIAFAKA-KAPADAI et al. (1986) also demonstrated that a glyceryl ether fraction obtained from the *Pinus halepensis* pollen by chromatography showed anti-platelet aggregation properties. The fraction at a concentration of 4.5 µmol l^{-1} inhibited in vitro aggregation of a rabbit's blood platelets induced with an activating factor (PAF) at a concentration of 1.5 nmol l^{-1}. This bioactivity as a potential for prevent atherosclerotic events but, as mentioned above, the precaution with the hemorrhagic side effect is crucial for the success of further investigatios.

Antiatherosclerotic properties preventing ischemic myocardial disease and strokes in humans receiving the pollen were carried out by KĘDZIA et al.(1994) and SZCZĘSNA et al. (1999) among others. Patients with multiple atherosclerotic arteries and advanced myopia and partial optic nerve atrophy showed a reduction of the levels of serum cholesterol as well as an increased field of view and stabilization of visual acuity after administration of the pollen (MACHOY-MOKRZYŃSKA et al. 1992). In several studies on animals, pollen bioactive substances improved liver function (UZBEKOVA et al. 2003). KLARIC et al (2018) determined the influence of dietary supplementation with propolis and bee pollen on liver pathology in broiler chickens. The control group of chickens received a basal diet, the experimental groups of chickens were fed with the same diet further supplemented with bee pollen and propolis. Researchers showed that the clusters of lymphocytes in the hepatocytes, the vacuolar degeneration and necrosis of the liver parenchyma, the bile ductile hyperplasia, and the various forms of pathological changes in the liver arteries and veins were more frequent in liver tissue samples of the control group compared to liver tissue samples of all the experimental groups. KLARIC et al. (2018) suggested that the supplementation of broiler chickens with bee pollen and propolis has a strong protective effect on liver pathology in broiler chickens.

The protective properties of pollen against ionizing radiation and its anti-platelet activity with regard to blood platelets as well as inhibiting the production of lipofuscin pigment in experimental animals have also been described. WANG et al. (1987) γ-irradiated with a dose of 7-8 Gy (grays) a group of mice fed with standard diet, and another group whose feed with a diet contained pollen. The experiment showed that in the group of mice whose feed contained pollen, the mortality rate reached 23.3%, while in the other group, with no pollen added to the diet, the mortality rate was in a range of 83.3% .

The consumption of pollen also has an effect on the production of lipofuscin. As an organism ages, a brown pigment (lipofuscin) accumulates in the cells. This accumulation is directly proportional to age. LIU and LI (1990) have demonstrated a significant reduction in concentration of lipofuscin in the myocardium, liver, brain and adrenal glands of mice fed with pollen as compared to the levels found in animals which were not receiving pollen. They have proved that the pollen slows down aging processes of animals (LIU et al. 1990).

Through the presence of phospholipids, pollen can protect the body against hepatosis (functional disorder of the liver), thus against the development of atherosclerosis. Phospholipids found in pollen, as lipotropic factors, inhibit the accumulation of lipids in hepatocytes, and protect the body against hepatosis. These compounds are part of cell membranes and they regulate selectively the penetration of substances into cells, thus they play a very important role in metabolism (PAUPIERE et al. 2014). Pollen supplementation also regulates the lipid-protein metabolism of the organism. Matuszewski and Drake's research proves that pollen normalizes the lipid-protein metabolism, as the level of liver enzymes in the blood serum has been significantly reduced. Served with toxic substances, pollen protected the liver cells from their harmful influence. The phenolic compounds contained in pollen exhibit a broad spectrum of biological activity in the body, i.e. they have an anti-inflammatory, antioxidant and anti-arteriosclerosis function, they strengthen capillaries and protect against ionizing radiation. Tikhonov et al. investigated the effect of pollen in the case of hypoxia in experimental animals – rats. The first group of animals had been receiving an intragastric preparation based on pollen preventively for 14 days, while the other group within the same period had been treated with Piracetam (a drug acting on cells of the central nervous system). Piracetam is a product which increases the use of oxygen and glucose, and improves blood circulation in the brain by reducing blood viscosity. Moreover, it improves microcirculation without broadening blood vessels or changes in blood pressure. Brain hypoxia was invoked in rats by ligature

of both artery blood vessels. The analysis of the results showed that after 7 days in the control group receiving no preparation or drug – 27% of animals survived, in the group receiving Piracetam it was 38% and in the group receiving the pollen preparation – 55% of animals (TRICHONOW et al. 2008). Thus, flavonoids and phenolic acids play an important role in the process of detoxification of liver tissue (ADRITOIU et al. 2014, FLOREK et al. 1995, JUŹWIAK et al 1992, KHALIL et al. 2010, PUT et al. 1994, WÓJCICKI et al. 1985, YILDIZ et al. 2013).

Pollen supplementation in the diet intends to increase the mental and physical efficiency of people. It has been demonstrated that pollen has an adaptogenic (adaptive) effect by increasing the resistance against harmful factors: physical, chemical and biological. The increase encompasses both physical efficiency of the organism in situations of increased physical effort and the improvement of brain functions such as memory, learning, thinking and the ability to concentrate, as well as an increase of the body's resistance to infection. Clinical studies were conducted with the participation of people suffering from mental illnesses and the elderly. Due to its nourishing and toning properties, pollen improves mental abilities and strengthens the nervous system weakened by stress or overwork. Particularly positive results were obtained in the elderly. The long-term use of pollen, even in small doses, results in a gradual improvement in the mood, restores interest in life and strengthens the body (WÓJCICIKI 1991). Both pollen and its extracts can be used in post-infarction states, as well as in peripheral circulatory disturbances and hypertension. In addition, in the case of the elderly the administration of small doses of this product helps inhibit cerebral atherosclerosis and improves cerebral circulation (HOWE et al. 1985).

TELESZUN et al. (1993) compared the effects of pollen in the form of bee pollen loads, and a lyophilised extract of pollen in the rehabilitation of workers eliminating the breakdown in the Chernobyl nuclear power plant, and suffering from complex neurological diseases. Bee pollen basket and the lyophilized extract of pollen were administered to patients with vegetative-vascular dystonia and organic damage of the central nervous system caused by ionizing radiation. It was proved that the neurological status of both groups of patients improved significantly after the treatment with pollen. They obtained better results using pollen loads compared to treatment with the lyophilised extract. The symptoms like headaches, pain of vegetative points, irritability, vascular lability and sleeping disorder disappeared after a few days. Also, it was observed that the administration of pollen results in a significant increase in phagocytic activity of peripheral blood. In the case of treatment using pollen loads, the number

of phagocytes capable of absorbing and disposing of pathogenic microorganisms has increased x 2.26, while using the lyophilized extract of pollen x 1.9 (TELESZUN et al. 1993).

GULHAN (2018) investigated therapeutic and protective effects propolis and pollen on reproductive functions of L-NAME-induced hypertensive male rats. Animals were indiscriminately separated into four groups: control, L-NAME, L-NAME+ propolis, L-NAME+ pollen. He showed that the levels of parameters (TOS, NF-kB, MDA) in L-NAME+ propolis and L-NAME+ pollen groups compared to the L-NAME group have decreased. TAS levels, PON1 and CAT activities were significantly decreased in testis tissue samples in the L-NAME-induced group. He thought that propolis or pollen is thought to help regulate reproductive function by inhibiting the functioning of inflammatory pathways leading to hypertension.

The impact of pollen on the physical condition of climbers was also evaluated. It was noticed that after pollen supplementation the number of various forms of white blood cells, including lymphocytes, monocytes and eosinophils, significantly increased. The level of eosinophils in climbers' blood serum at the end of the experiment was 2x higher, monocytes 2.5x higher than in serum prior to the experiment (DROŻDŹ et al. 1986). MALMSTRÖM and CEDERLÖF (1983) used the properties of pollen in the treatment of the upper respiratory tract. For 14 days soldier trainees received pollen preparation to prevent a cold. It was demonstrated that only 35% of them developed various kinds of infections of the upper respiratory tract. Those who did not receive the pollen, had a cold of various intensity. There are many reports on the antibacterial and antifungal effect of pollen. It affects both human pathogenic bacteria (*Staphylococcus aureus* and Gram-negative bacteria including *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeurgionsa*) and the yeast-like fungi (*Candida albicans*). Mainly flavonoids and phenolic acids are responsible for this effect (BALTRUSYAT et al. 2007, ERKMEN 2008, LIEBELT et al. 1994, MATEESCU et al. 1997).

Pollen also has a positive impact on curing prostate inflammation. In the research, approximately 87% of patients with nonbacterial inflammation of prostate felt better after pollen treatment. What is more, a positive effect was observed in 50% of patients with the benign prostatic hypertrophy (DOBROVODA 1986, KRIWCZANSKIJ 1987). Pollen can also be used in curing iron deficiency anemia. It was noticed that in people treated with pollen, the number of red blood cells and the level of hemoglobin and Fe increased. Clinicians have proved that pollen is very effective in curing duodenal ulcer disease. The radiological and endoscopic research has proved that after administration of pollen the duodenal ulcers began to cicatrize. Satisfying results were also obtained in the treatment of bleed-

ing duodenal ulcers not qualified for surgery. The hemorrhages disappeared after 3–4 days of pollen treatment, while in the traditional way it takes 10 days to stop duodenal ulcers bleeding (KEDZIA 1994). Pollen showed good effects in treating: gout, kidney inflammation, urinary tract disorders, climacteric and vegetative dystonia. There is an increasing amount of data on supplementation of pollen to animal diets, for example, the research on its influence on the condition of pigs. Pollen is characterized by a high anti-inflammatory effect confirmed in animal studies. Its properties can be compared to anti-inflammatory drugs such as naproxen or indomethacin. The mechanism is based on inhibiting the activity of enzymes responsible for forming inflammatory mediators in tissues. Concentrated extract of the pollen fed to a rat eliminated paw edema associated with the administration of carrageenan. Flavonoids, phenolic acids, fatty acids and phytosterols which are present in pollen are responsible for the antioedematous effect. It has been demonstrated in experimental studies that the administration of pollen in a dose of 50 mg/kg b.w. eliminates 75% of paw edema (CHOI 2007, LOSCHEN et al. 1991).

It has been proved that pollen has a high nutritional value. Mice and rats fed with pollen were characterized by a higher content of vitamin C and Mg in tissues and a higher content of hemoglobin, as well as a bigger number of red blood cells compared to animals given a standard feed. Feeding with pollen caused a rapid increase in the body weight compared to an ordinary diet served to starved animals or animals on a diet without vitamins. OLIVEIRA et al. proved that a crucial role in this process is played by exogenous amino acids, vitamins and bioelements. Moreover, studies in rodents showed that the contents of vitamin C and Mg in the thymus, heart muscle and skeletal muscles increase after pollen supplementation. Both the level of hemoglobin and the number of red blood cells was higher compared to the results obtained in animals fed with standard feed (OLIVEIRA et al. 2009, LIEBELT et al. 1994, MATEESCU et al. 1997).

A hypolipidemic effect of the pollen has also been reported. Scientists conducting research on rats and rabbits have discovered that pollen reduces the content of total lipids, triglycerides, total cholesterol, LDL cholesterol fraction and β-lipoproteins in blood serum (JUŹWIAK et al. 1989). Similar studies were conducted in humans. They confirmed that in the blood serum of people with impaired lipid metabolism, there was a reduction in the content of lipids after the administration of pollen (MANNING 2001, KASSYANENKO et al. 2010).

Due to high contents of phytosterols, pollen shows an estrogenic effect by stimulating the process of producing and maturation of ova in animals and humans (TRICHONOW et al. 2006). Pollen contains 0.1–1.6% of phytos-

terols. Phytoestrogens are structurally similar to endogenous estrogens and therefore they connect with estrogen receptors. Also, a relationship has been detected between the activity of estrogen receptors and cardiac physiology, which influences the regulatory systems of oxytocin. The estrogenic activity of these compounds has a beneficial effect on the maturation of oocytes in animals. Researchers conducted tests on rabbits fed with pollen which checked:

a) the functioning of ovaries;
b) therapeutic effects on blood profiles and parameters;
c) the number of offspring.

They discovered that a dose of 200 mg/kg b.w. in a rabbit caused an increase in the quality of sperm, increased fertility, as well as improved blood and biochemical parameters (ATTIA et al. 2011).

Many publications are devoted to the detoxifying properties of pollen (FLOREKET al. 1995, JUŹWIAK et al. 1992, WÓJCICKI et al. 1989). Polyphenols, mainly flavonoids and phenolic acids present in pollen play an important role in the process of body detoxification. Studies were conducted in rats which were administered organic solvents, such as carbon tetrachloride and trichloroethylene. These substances caused deep damages of the rats' liver cells. The animals were also given ethanol and allyl alcohol which caused fatty liver cirrhosis, as well as drugs: acetaminophen and hydrocortisone. After feeding rats with pollen the level of liver enzymes and bilirubin in the blood serum was reduced to physiological values. There was a very high concentration of alanine, aspartate transaminase, acid phosphatase and bilirubin in the blood serum. Hence, pollen is recommended in acute and chronic inflammation, in the early stages of degenerative diseases, congestive liver diseases, as well as toxic and traumatic injuries of this organ.

Metabolizing properties of Fe, Ca and Mg in rats with anemia were examined, as well as the antiallergic properties of myricetin present in pollen given to mice. The impact of the pollen and propolis on the organism was studied. It was found that the addition of these products to the diet resulted in body weight gain. An increased level of hemoglobin in animals with anemia was observed, and a positive effect of these products on the level of Mg in the organism (HARO et al. 2008). The therapeutic effects of the pollen present in Apiter product were observed in dogs and cats infected with rickettsiales. The pollen was administered to animals at the age of 2–8 months with bone and articular diseases and ascertained rickettsiosis. Patient selection was based on the analysis of X-ray images and biochemical tests. Research has shown that in animals supplemented with pollen, biochemical parameters have reached a physiological level: Ca level

increased from 4 mg dl^{-1} to 9.2–11.5 mg dl^{-1}, P from 1.6–3.81 mg dl^{-1} to 4.1–5.7 mg dl^{-1}, Mg level normalized (2.5–3 mg dl^{-1}). The reduction in alkaline phosphatase 116-443 UI l^{-1} to 77–187 Ul l^{-1} has been reported, with a simultaneous normalization of liver enzymes (ALT, GGT, AP) (SAPCALIU et al. 2009). The influence of pollen on the length of the villi of the small intestine in broilers was studied. It was found that the supplementation of pollen in the feed has a beneficial effect on the intestinal villi, increasing their length (in the 1st and 2nd week of age in chickens) by 37.1% and 29.4% in duodenum, 28.1% and 33.7% in jejunum and 18.6% and 16.2% in ileum (Wang et al. 2009).

On the basis of the above mentioned research results it can be concluded that flower pollen (bee pollen) is characterized by a number of favorable biological properties which are often used in medicine to improve health (ALYANE et al. 2008, ARCT et al. 2008, PASUPULETI et al. 2017). For many years we have been observing an increased interest of supplements based on bee products which are often used in alternative medicine or as a supplement to conventional medicine.

The major problem is that bee-collected pollen may constitute risk factor concerning the presence of contaminants like a heavy metals, pesticides, bacteria, antibiotics). Therefore standardized pollen tablets are recommended. Moreover, allergic reactions including anaphylaxis have been recognised after intake of bee-collected pollen. Patients who are prone to allergies or atopic individuals should avoid any type of bee pollen, bee-collected and extracts. The application of bee pollen should be discussed with doctors in order to avoid complications (DENISOW et al. 2016).

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