# **NEW POSSIBILITIES OF USING MULBERRY SILKWORM FEED**

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In the countries where the silk industry is developed, white mulberry is one of the most important plants. Its leaves are the best food for the caterpillars of mulberry silkworm (Bombyx Mori L.). It is still not fully investigated why the silkworms prefer mulberry. However, it is certain that the protein substances produced in silkworm bodies in 70% come from mulberry leaves. These substances constitute one of the finest fibers - natural silk. Currently, mulberry is cultivated mainly in Asia, where sericulture maintains its high position. The fact that mulberry contributes not only to the formation on the silk but also can be used for other purposes has drawn attention of scientists to this plant. The exploitation of mulberry tree in Poland is limited to the life of silk in summers therefore new potential applications of this raw material are sought after. Literature reports confirm very rich composition of mulberry leaves in terms of bioactive substances. These compounds can be used in prevention of lifestyle diseases,. The paper presents the studies on the content of bioactive substances in the leaves of white mulberry and the potential of using the silkworm feedstock for humans.

**Keywords:** silk industry, silk fiber, mulberry silkworm (Bombyx Mori L.), white mulberry (Morus alba L.), bioactive compounds, lifestyle diseases

#### Introduction

Silk is one of the most ancient and valuable fibers in the world. Back in the antiquity it

was a valued commodity imported to Europe along the Silk Route from China and the process of its manufacturing was kept secret. This thin glossy and very strong natural fiber is apart from wool one of the natural fibers of animal origin. It is derived from cocoons of mulberry silkworms. The fisrt stage of development of mulberry silkworm is a caterpillar, which eats enormous amounts of white mulberry leaves and in its 4<sup>th</sup> week starts producing cocoons made of silk fiber. There are numerous races of silkworms and several other animals capable of producing fibers with similar properties, yet not as good as silk produced specifically by mulberry silkworm (Bombyx Mori L.). Other silkworm varieties have been labeled as wild silkworms. Nowadays, the mulberry silkworm is a completely domesticated monophagy and feeds only on the mulberry. The quality of mulberry leaves has direct effect on the quality of produced silk. The formation of silk in the silkworm organism is a result of absorption of the nutritional substances found in mulberry leaves. Thus the caterpillar is the link between the leaf and the fibber. Silk fiber is composed from two main ingredients i.e. fibroin and sericin, with small amounts of pigments, waxes and minerals. Fibroin is the actual fiber and sericin is a gluing substance that covers the fiber, which is dissolved during boiling of silk. One may say that mulberry leave is the raw material for production of silk and silkworm caterpillar is the element that processes it into the fiber. The exquisite properties of silk result directly from the leaves of white mulberry [Frentzel 1986, Kopański 1955, Krasnodębski 1953, Pieprzyk-Kokocha 2005].

What makes mulberry leaves so special that the fiber derived from them is of highest quality among all natural fibers?

White mulberry (Morus alba L.) is a large long-living tree with light grey bark, which gets darker with age. The leaves are light green soft and shiny in various shapes. The tree does not have special soil requirements and grows in almost whole Europe. However the area of growing does not overlap with the area of mulberry cultivation for silkworm breeding. Despite the fact that mulberry is tolerant to frost the length of vegetation period plays an important role, thus north European countries do not have suitable climate for mulberry. In order to obtain as much good quality leaves as possible, mulberry requires special treatments [Butt 2008, Litwińczuk 1993, Pieprzyk-Kokocha 2005]. Fresh and well insolated leaves contain about: 73% water, 7,5 % protein, 1,1 % lipids, 10,6 % carbohydrates, 2,6 % mineral substances, 5,2 % ciał bezazotowych, and small amounts of vitamins (B, C and D) and enzymes [Kopański 1955, Krasnodębski 1953].

More and more information is available on the mulberry leaves as a good source of bioactive substances i.e. the substances that stimulate desired course of metabolic processes in humans. Functional food, which shows beneficial and well documented effect on human health, is a potential source of these substances. The bioactive ingredients that have already been tested for their beneficial effect are for example dietary fiber, poly-unsaturated fatty acids, vitamins, mineral elements and phyto-chemical substances such as flavonoids and phenolic acids [Charunuch 2008, Mazza 1998, Srivastava 2003, Świderski 2003]. The bioactive substances play an important role in prevention of the so called lifestyle diseases, which prevalence depends on the civilization development of a society. These diseases include mainly diabetes, hypertension, coronary disease and obesity. Health burden linked to lifestyle, air pollution, and UV radiation is additionally increased as a result of high amounts of free radicals in human organisms. The radicals may cause damage to the lipids in cell walls, proteins, enzymes and to DNA, what results in changes of functioning of body cells. Human organism is equipped with a

defense mechanism in the form of endogenic anti-oxidants. However, anti-oxidants from food (e.g. phyto-chemical poly phenolic compounds) can have an important role in decreasing the oxidative damage [Dudek-Makuch 2007, Betlejewski 2007].

#### Materials and methodes

The studies were conducted on water and water-ethanol extracts of white mulberry leaves that were analyzed with HPLC.

The materials for the study were the leaves of white mulberry (Morus alba L.) of Wielkolistna Żółwińska variety, collected at the experimental Farm of the Institute of Natural Fibers and Medicinal Plants in Petkowo (Poland). This variety has been bred specially for use in sericulture.

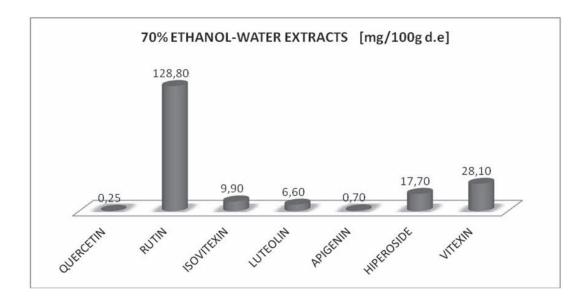
Water and 70% ethanol (V/V) were used for the extraction of biologically active substances, and then the extract was filtered. In case of alcohol, the extraction was complemented with distillation under low pressure. The extracts were frozen and lyophilized.

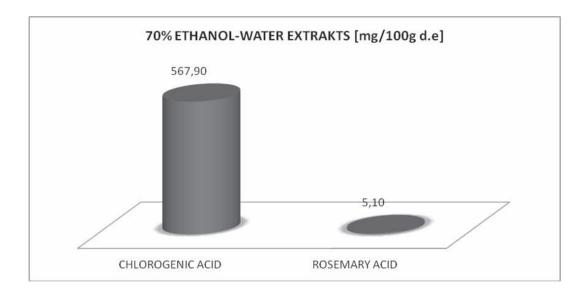
he poly phenolic profile was determined with the use of HPLC with DAD detection. Methanol was added to the dry extract. The solution was heated with reverse cooler, and then it was cooled and infiltrated. The methanol extraction was run twice, then the infiltrate was evaporated until dry under low pressure and finally water was added. The extract was centrifuged three times with methylene chloride. The water layer was evaporated until dry under low pressure. The remaining matter was dissolved in methanol and determined with the use of HPLC.

### **Results and discussion**

The content of bioactive substances in ethanol-water and water extracts of white mulberry leaves is presented in Figures 1 and 2. The following poly phenolic compounds have been identified in the studied extracts: guercetin, rutin, hyperozide, vitexin, apigenin, isovitexin, and phenolic acids i.e. chlorogenic and rosemary acids. Quercetin is a flavonol that reduces the risks of stomach ulcers; it is an anti-alergic, anti-inflammatory and immune modulating compounds. It is aslo found to be one of the factors reducing the occurrence of pathogenic lesions in diabetes. Rutin is a flavonol with anti-cancer, anti-inflammatory and anti-thrombotic properties. It has chelating capabilities towards several metals e.g. iron and copper. Another flavonol found in mulberry leaves, hyperozide, has vasoprotective, hypotensive (lowers blood pressure), diuretic, anti-inflammatory, anti-sclerotic activity. It also improves blood circulation and prevents clotting of veins. Luteolin is also a flavonoid compound with diuretic, anti-inflammatory and anti-alergic properties. Anther flavonoid determined in the mulberry leaves is vitexin which is a compound of high medical importance as it improves coronary circulation and is regarded as mild cardiac medicine. Due to its low toxicity it can be used over long periods. Apigenin shows strong anti-oxidant and anti-inflammatory properties – it has radical scavenging activity, inhibits the lipid peroxidase activity what prevents oxidation of LDL fractions to their oxy-LDL fraction that causes sclerosis. Apigenin also has certain anti-cancer properties. Isovitexin is found to be a strong anti-oxidant. Within phenolic acids detected in mulberry leaf extracts was rosemary acid, a substance used in treatment and prevention of numerous diseases e.g. in stabilizing biological tissues, protection against UV radiation and reactive oxygen species including free radicals. Rosemary acid is characterized with antioxidant, anti-inflammatory, anti-viral anti-hormonal activity. The other phenolic acid found in mulberry was chlorogenic acid, wheih lowers carbohydrate absorption in alimentary tract, what forces the organism to use its reserves of sugars. This acid improves the sensitivity of cells to insulin and acts as a strong anti-oxidant [Charunuch 2008, Dudek-Makuch 2007, Dugo 2009, Gawlik 2004, Grajek 2011, Jeszka 2009, Kopacz 2008, Naowaboot 2009].

FIGURE 1. The content of bioactive compounds in ethanol-water extracts from the leaves of the white mulberry [mg/100 g s.e.]





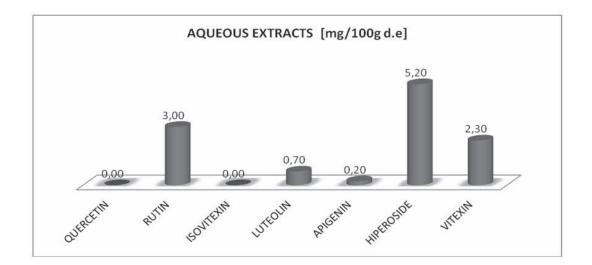
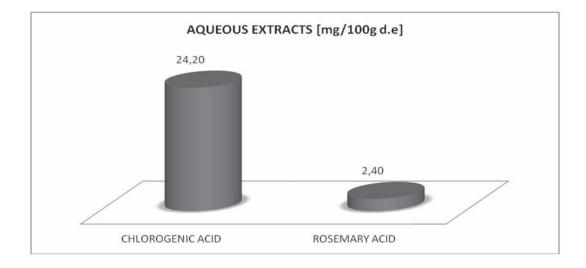


FIGURE 2. The content of bioactive compounds in aqueous extracts from the leaves of the white mulberry [mg/100 g s.e.]



The results obtained in the study allow to state that both types f extracts contained phenolic compounds, whereas the content of these substances in ethanol-water extracts was much higher than in aqueous extracts. In both types of extracts (ethanol-water and water) the highest amounts were observed for chlorogenic acid (567,90 mg/100g d.e and 24,20 mg/100g s.e respectively). In ethanol-water extracts significant amounts of rutin were determined (128,80 mg/100g s.e), and then vitexin, hiperozide, isovitexin, lueolin, rosemary acid, apigenin and quercetin (see Figure 1). However, water extracts were characterized with high content of hyperozide (5,20 mg/100g s.e), and smaller amounts of rutin, rosemary acid, vitexin, lueolin

and apigenin. In case of water extracts two compounds were not detected i.e. quercetin and isovitexin (Figure 2).

Phenolic compounds that are natural anti-oxidants, acting as oxygen scavengers, chelating metals and protecting cells and reducing the oxidative damage are of high importance to human organism. Therefore, the slogan "mulberry not only for silkworms" seems more and more justified by scientific data [Cieślik 2005, Grajek 2011, Kolanowski 1999].

#### Conclusion

The leaves of white mulberry, apart from being an important element of sericulure, are also a source of bioactive substances beneficial for human health. Thus it is worthwhile to introduce them as a permanent element of human diet for prevention of lifestyle diseases. Although there are several products with the addition of mulberry leaves such as tea made of dried and powdered leaves and rice snacks with the leaf extracts, the studies on new food applications of mulberry and its nutritional components should e continued.

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