

### **Research project objectives**

The purpose of the project studies is to evaluate possibility of designing highly biologically active legume seeds in biotechnological processes of germination and lactic acid fermentation on the basis of analysis of bioactive properties of isoflavones from legumes. To achieve this goal three research models will be used, i.e. *in silico* technique of molecular docking, as well as modeling using calorimetric effects of interactions between legume seeds isoflavones and systemic proteins, and finally test with cell cultures, which will include interactions that occur between individual isoflavones from legume seeds and their mixtures with human proteins involved in health-promoting mechanisms. Modifications of methods of biotechnological processes will be aimed at achieving a high concentration of those isoflavones, which will be first assessed as exhibiting high potential of health-promoting activity in model studies. Potential health promoting activity will be indicated for antiproliferative activity of cancer cells, prevention of tumor cell migration and osteoporosis. Comparison of the results of three carried out analytical models should allow to estimate the correlation between the different techniques and the strengths and weaknesses of these techniques.

### **Basic research to be carried out in the project**

Realization of the research that was described in the project will provide new knowledge on interactions of isoflavones from legume plants with proteins, including enzymes and receptors presented in the human body and their effects on cell physiology. In the following interactions will be characterized in terms of energy and stoichiometry and the fragments of proteins that are responsible for interaction with isoflavones will be indicated. The research will validate, based on the results of applied research models, research hypothesis concerning increase of bioactive activity selected legume seeds, due to the biotechnological modification involving the germination and lactic acid fermentation. A new issue is the usage of three different levels of modeling based on a coherent test material to determine the possibility of using selective techniques such as for example molecular modeling. to predict the real body response. Innovative and interdisciplinary nature of the research results of close cooperation of bioinformaticians, biochemists, food technologists and analysts, and microbiologists. This way it is possible to correlate molecular mechanisms with bioactive compounds chemical structure, due to the selected modifications and modulation of cellular signals.

### **Present reasons for choosing the research topic**

Due to their high nutritional value, leguminous plants are one of the world's most common food crops. They are a good source of polyphenols like phenolic acids, flavones and flavonols, and isoflavones not common for other plants. The concentration of isoflavones in legumes seeds is up to 0.1%. Isoflavones structure is similar to female hormone estradiol, which makes them capable of interacting with estrogen receptors. Thus this group of compounds are often called phytoestrogens. Isoflavones are often used to replace or supplement conventional postmenopausal hormone therapy but in contrast to them they do not cause increased risk of breast cancer. Consumption of isoflavones significantly inhibits bone resorption and stimulates their formation. In germinated seeds the content of isoflavones increases by 50%. The effect of growth of microorganisms leading lactic acid fermentation is inhibition of growth of pathogenic microflora, including *Salmonella enterica* and causes an increase of bioactive substances concentration in sprouts due to the probiotic bacteria metabolism.

In Asian countries a significant part of legume seeds is traditionally consumed in the form of fermented food, as natto or tempeh, where the main form of isoflavones are aglycones. In non-fermented products such as soy milk, tofu, soy protein concentrates and food supplements, isoflavones are in the form of glucosides. The tradition of legumes consumption is very popular all over the world, but especially in Asian countries of those rich in isoflavones. Japanese, Chinese and Indonesians consume from 25 to 100 mg of isoflavones per day. Western people eats only about 1 mg per day, mainly in the form of glucosides. In countries using an eastern diet rich in isoflavones it is observed fewer cases of postmenopausal women, who outstand common symptoms of low estrogen levels. After designing conditions of germination and fermentation particularly favorable synthesis of biologically active isoflavones it will be possible to achieve in further development research health promoting foods originating from sprouts of legumes seeds that can help in the prevention of osteoporosis, while not causing the risk of developing breast cancer. This will improve the quality of postmenopausal women life and reduce spending on health care. Currently for this purpose mainly dietary supplements containing soy isoflavones glucosides are used. Despite many studies conducted so far, the problem of confirming the pro-health action of isoflavones from legumes is still very actual. This way confirmed and legally presented on the products packages health-promoting opinion on isoflavones pro-health actions will have a better chance to influence the conscience and health of consumers.