

# Summary of professional accomplishments

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## I. RESEARCH ACTIVITIES

### 1. DIPLOMAS AND ACADEMIC DEGREES

**2004: Ph.D. in Agriculture Sciences;** branch: food and nutrition technology; Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences. The thesis title: *Assessment of the nutritional quality in Poland in years 1980-2000* (thesis supervisor: prof. dr hab. Anna Gronowska-Senger).

**1997: Master of Science;** specialization: food and nutrition technology; Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences. The thesis title: *Determination of the content of resistant starch in selected food products* (thesis supervisor: prof. dr hab. Anna Gronowska-Senger).

### 2. WORK EXPERIENCE

**Science 2005:** Assistant Professor, Department of Human Nutrition, Chair of Nutritional Assessment, Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences.

**2004 – 2005:** Assistant, Department of Human Nutrition, Chair of Nutritional Assessment, Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences.

**2000 – 2004:** Ph.D. student on Faculty of Human Nutrition and Consumer Sciences, Warsaw University of Life Sciences.

**1997 – 2004:** technologist in Food Technology Laboratory of the National Food and Nutrition Institute in Warsaw.

### 3. SCIENTIFIC ACHIEVEMENT JUSTIFYING THE APPLICATION ACCORDING TO ART. 16 PARAGRAPH 2 OF ACT FROM MARCH 14, 2003 ON SCIENTIFIC DEGREES AND PROFESSORSHIP (DZ. U. NO. 65, IT. 595 WITH NEXT AMENDMENTS): PUBLISHED WORK

#### 3.1. INDICATION OF SCIENTIFIC ACHIEVEMENT:

**MAGDALENA GÓRNICKA:** Study on the effect of  $\alpha$ -tocopherol on redox homeostasis of organism subjected to the physical effort. SGGW Press, Warsaw, 2013, p. 1-110

## INTRODUCTION

Systematic, moderate physical effort along with a proper diet is an important component of the lifestyle with a positive influence on health, contrary to hypokinesia, which is a risk factor of chronic diseases. Intense workout results in an increased production of free radicals as a consequence of an intensified oxidative metabolism. Free radicals emerge not only as a result of physical exercise but also constitute an inherent product of processes taking place in the cells. Their increased production entails the disturbance of the oxidation-reduction balance and intensifies the oxidation reaction in the cells. High concentrations of oxidizing compounds lowers the level of natural antioxidants that, as a consequence, leads to oxidation modifications of cell components such as proteins, fats, carbohydrates and DNA. Oxygen organisms developed defence mechanisms to maintain the redox homeostasis, both enzymatic (antioxidant enzymes) and non-enzymatic based on exo- and endogenous antioxidants. Among others, they include tocopherol as well as carotenoids, ascorbic acid, glutathione and flavonoids.

$\alpha$ -Tocopherol is an integral part of the cellular anti-oxidation system protecting cell components from oxidation. It plays a very important role in the human body and is indispensable for the maintenance of homeostasis. The main function of tocopherol is to prevent the peroxidation of lipids in cellular membranes as well as to protect them from free radicals.

Its antioxidant role during physical exercise has not been explained completely and the evidence of the supplementation during exercise is still limited. The optimisation of doses of individual antioxidants, including  $\alpha$ -tocopherol, recommended to individuals exposed to oxidative stress is of interest to scientists and subject to intense research.

Results of the research done so far on the influence of tocopherol on oxidative changes caused by physical exercise are ambiguous, with differences resulting from the type and dose of applied supplements, type and duration of physical exercise as well as the methodology of biological experiments. It is indicative of the need for further research in order to determine the role of  $\alpha$ -tocopherol in the maintenance of homeostasis in the body subjected to physical exercise. In addition, a lack of data on the effects of physical exercise on previously untrained young organisms, that seem to be more sensitive and thus more

strongly exposed to the effects of oxidative stress. Therefore, the aim of this study was to investigate the role of  $\alpha$ -tocopherol in the maintenance of redox homeostasis in young subjects treated differentiated effort. The study was conducted on animal models - male Wistar rats.

### **3.2. PURPOSE, RESEARCH HYPOTHESES AND CHOSEN ASPECTS OF RESEARCH METHODOLOGY**

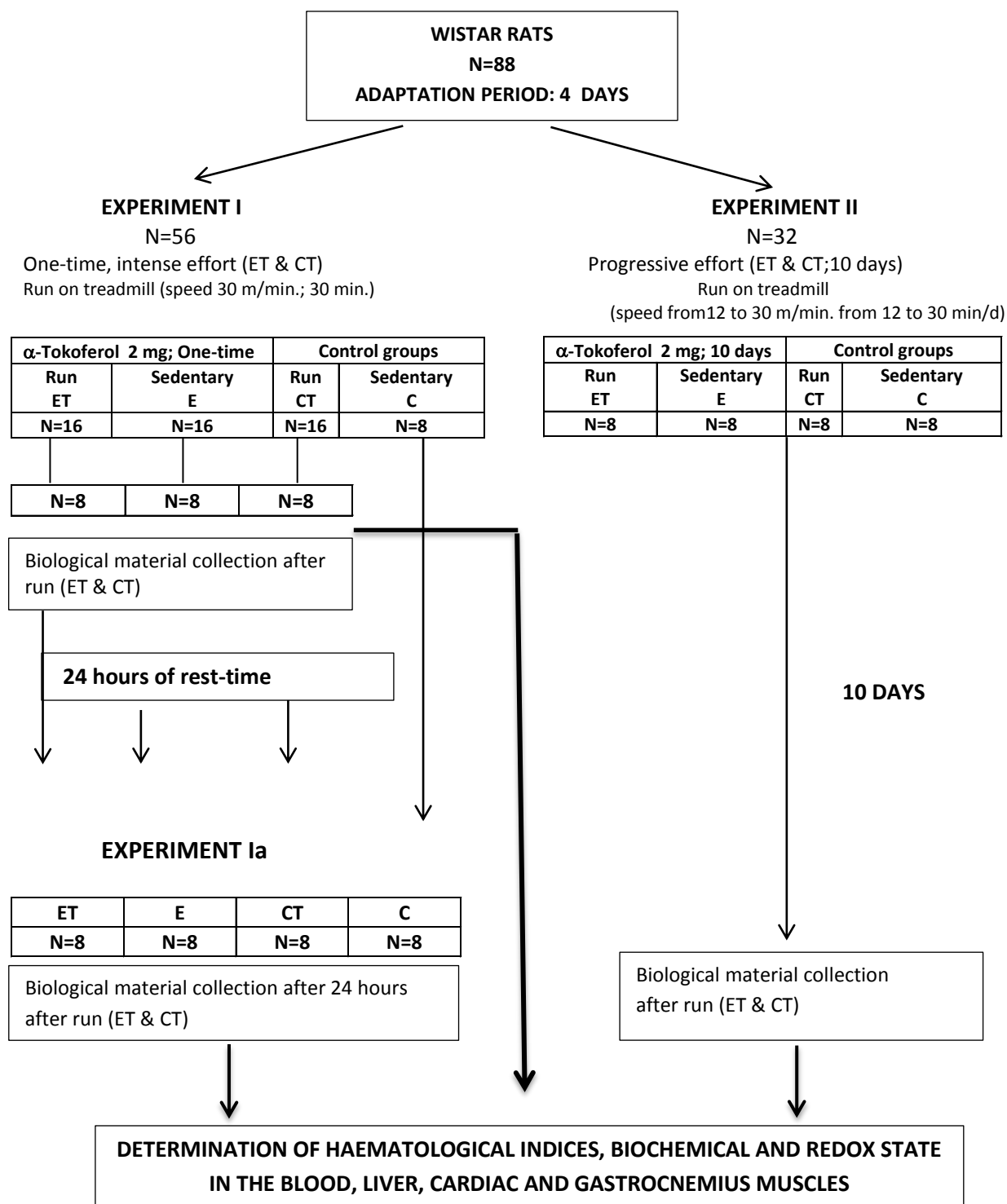
The aim of the presented research was to examine the influence of  $\alpha$ -tocopherol acetate on the redox homeostasis (measured by values of selected biochemical and oxidative stress markers) disturbed due to physical exercise of varied intensity and duration in previously untrained young subjects.

Based on the main aim the following hypotheses were formulated:

1. One-time intense physical exercise disturbs redox homeostasis in previously untrained subject and a single application of  $\alpha$ -tocopherol 12 hours before the exercise, causes the balancing of values of biochemical and oxidative stress markers within one day after the exercise.
2. Subjecting a body previously untrained to progressive physical exercise (with increasing intensity and duration) activates enzymatic antioxidation defence systems and does not require the application of  $\alpha$ -tocopherol.

To verify the hypotheses proposed above, the research was conducted on 88 rats previously untrained according to the studies' design presented in Fig. 1. Animals were subjected to a four-day adaptation to changed living conditions during which they started to be fed with a vitamin E deficient diet. The research consisted of two animals experiments:

- Experiment I – to determine whether and in what manner a single application of  $\alpha$ -tocopherol influences the redox status disturbed due to a single physical exercise of great intensity, measured in the biological material collected immediately after the



Symbols mean groups: E – groups with tocopherol, C – control groups, T – running groups

Fig. 1. Design of the animals studies

exercise. As a part of experiment I, animals from groups receiving tocopherol, because of the time necessary to absorb it, were once fed *per os* vitamin E (oil solution *all-rac* of  $\alpha$ -tocopherol acetate in soybean oil, dosage: 2 mg of  $\alpha$ -tocopherol/d/rat) 12 hours before the one-time exercise. Some animals previously untrained were subjected to one-time physical exercise on a treadmill with the belt speed 30 m/min. (zero angle of inclination) for 30 minutes. Immediately after the physical exercise, a half of the animals from groups subjected to exercise as well as the untrained group after one-time tocopherol supplementation were anaesthetized and biological material was collected.

- Experiment Ia – the continuation of experiment I, was aimed as the study the influence of  $\alpha$ -tocopherol on the redox condition of the subject disturbed with intense one-time exercise, measured in the research material collected 24 hours after the end of the one-time intense exercise.
- Experiment II – to determine whether and in what manner the application of  $\alpha$ -tocopherol influences on the redox status indicators during progressive physical exercise (increasing intensity and duration in the course of 10 days) measured in the biological material collected immediately after the exercise. During the experiment II, animals were fed with a vitamin E deficient diet for 10 days. A half of the rats were administered an oil solution of  $\alpha$ -tocopherol acetate *per os* in soybean oil for 10 consecutive days; dosage: 2 mg of  $\alpha$ -tocopherol/d/rat after the training while other animals were fed soybean oil without the vitamin. Some animals previously untrained from groups with/without  $\alpha$ -tocopherol have been subjected to daily exercise of increasing intensity for 10 consecutive days (treadmill training: 12-30 min; speed: 12-30 m/min., zero angle). Initially, the training lasted 12 minutes and the belt speed amounted to 12 m/min. On consecutive days, the belt speed was increased by 2m/min. and the duration increased by 2 minutes attaining the speed of 30 m/min. and the duration of 30 minutes on the tenth day of training. Immediately after the physical exercise, the animals were anaesthetized and biological material was collected.

Upon the end of each experiment, the animals were anaesthetized by inhalation with the use of isoflurane in order to collect blood samples and extract selected tissues i.e. liver, heart and the gastrocnemius muscle (*gastrocnemius surae*).

The influence of  $\alpha$ -tocopherol and physical exercise on the body was evaluated comprehensively with the use of measures such as haematological (haemoglobin and haematocrit concentrations), biochemical related to the metabolism of energy substrates (lipid profile, glucose concentration) indicating the intensity of exercise (the activity of creatine kinase – CK) and inflammation (concentrations of the C-reactive protein CRP) marked in the blood. Additionally, biomarkers of the redox status determining the activity of antioxidant enzymes (SOD and GPx) were marked along with the level of lipid peroxidation (concentrations of lipid peroxides – TBARS method) and the concentration of the studied antioxidant, i.e.  $\alpha$ -tocopherol in the plasma and tissues. Selected animal development indicators were determined, i.e.: body weight gain, diet consumption and the efficiency ratio diet.

All procedures were carried out in compliance with ethical requirements and were approved by Third Local Ethical Committee for Animal Experiments at the Warsaw University of Life Sciences (WULS-SGGW, Poland (Resolution No. 33/2008 and extension: Resolution No. 13/2012).

### 3.3. RESULTS

#### ***Influence of $\alpha$ -tocopherol acetate and one-time intense physical exercise on the redox homeostasis***

One-time physical exercise and the application of  $\alpha$ -tocopherol did not significantly influence the concentrations of haemoglobin, haematocrit, glucose and CRP protein measured in the animal blood collected immediately after the exercise. 24 hours after the end of the exercise, higher haematocrit values were demonstrated in the blood of rats from the control group subjected to intense exercise, which is indicative of the significant impact of the duration of rest after exercise ( $p \leq 0,002$ ) and one-time exercise ( $p \leq 0,003$ ) on the increased haematocrit values. Exercise can cause a temporary disturbance of rheological properties of blood due to the relocation of fluids from intra-vascular space to working muscles, inducing hemoconcentration, among other things, accompanied by an increase in the haematocrit value.

One-time exercise significant ( $p < 0.001$ ) increased the activity of creatinine kinase (CK) measured in the plasma of groups subjected to exercise, collected immediately after the exercise. Additionally, CK activity in the plasma of rats belonging to these groups significantly increased after 24 hours, which indicates that that time period was not sufficient to make it return to initial values and changes invoked by the one-time exercise have deepened for a day after the end of the exercise. Tocopherol application significantly reduced CK activity in the plasma of experimental animals, which was lower by 60% in comparison with the control group.

After one day of rest, significant influence of one-time exercise on an increase in the concentration of the CRP protein in the blood was observed, which can be indicative of a slight increase in inflammatory processes in the body due to exercise. A significant impact of the interaction: exercise  $\times$  duration of rest ( $p = 0.03$ ) on the CRP concentration was also observed with differences between sedentary animals and animals subjected to one-time exercise were greater in control groups than in groups getting tocopherol.

No significant influence of the application of  $\alpha$ -tocopherol or one-time intense physical exercise on the level of individual lipid fractions in the plasma collected immediately after the exercise was observed. Only the duration of rest significantly influenced ( $p \leq 0.01$ ) an increase in the level of triglycerides (TG). Their concentration measured in the blood collected one day after the exercise was significantly higher (by 15% on the average) in animals subjected to exercise than in control animals. It can be indicative of the mobilization of lipid reserves due to exercise.

The application of  $\alpha$ -tocopherol caused a significant increase in its concentration in the animal blood plasma measured in the material collected immediately after one-time intense physical exercise and after one day of rest.

Among the tissues, the highest average concentration of  $\alpha$ -tocopherol was observed in the liver (96.9 – 101.7 nM /g of tissue) and the heart (57.6 – 65.2 nM/ g of tissue) while the lowest concentration was found in the muscles (31.7 – 37.3 nM/ g of tissue). A single application of tocopherol and intense exercise did not significantly influence the content of  $\alpha$ -tocopherol in the liver and the heart measured in the material collected immediately after the exercise, contrary to the muscles for which a significant impact of both these factors was observed. After 24 hours of rest, the concentration of tocopherol in the heart and muscles in

control groups subjected to a single exercise significantly decreased (respectively:  $p \leq 0.01$  and  $p \leq 0.004$ ).

The effect of  $\alpha$ -tocopherol application and one-time physical exercise on the activity of antioxidant enzymes measured in the blood and in the tissues was varied. One-time physical exercise and the application of  $\alpha$ -tocopherol significantly increased the activity of SOD in erythrocytes only in those groups where  $\alpha$ -tocopherol was applied. Significant influence of the one-time exercise and tocopherol and the interaction of exercise and tocopherol on SOD activity in a muscle was also demonstrated, involving a decrease in SOD activity in the muscles of animals from groups subjected to exercise that received  $\alpha$ -tocopherol and its increase in control groups. The duration of rest after the exercise turned out to be significant only in the case of the SOD activity in a muscle, which increased 24 hours after the exercise.

No significant influence of the one-time exercise, tocopherol or rest duration after the exercise on the GPx activity in the blood was found. The activity of GPx in the liver increased under the influence of the one-time physical exercise in animals from the control group and, after 24 hours, also in groups that received tocopherol. The activity of GPx similarly increased in groups subjected to the exercise. After 24 hours of rest, differences in the activity of GPx between E and ET groups and K and KT groups disappeared, with a significant decrease in the activity of GPx found in the trained control group ( $p \leq 0.03$ ). Tocopherol significantly influenced the GPx activity only in the muscle, causing an increase in its activity in groups receiving  $\alpha$ -tocopherol.

One-time intense exercise and the application of tocopherol significantly influenced the concentration of lipid peroxides (TBARS) in the gastrocnemius muscle. The level of peroxides in the group subjected to exercise that received tocopherol was lower than in the control group. A single application of tocopherol was not sufficient to effectively counteract the oxidation of lipids in the heart or plasma. What is more, groups fed the vitamin had significantly higher concentrations of TBARS in the plasma.

***Influence of  $\alpha$ -tocopherol acetate and progressive physical exercise on the redox homeostasis of the body***

Progressive physical exercise and the application of  $\alpha$ -tocopherol did not significantly influence the values of haematology indicators and the concentration of glucose and CRP protein in the animal plasma. However, physical exercise caused a significant increase ( $p \leq 0.04$ ) in the activity of creatine kinase (CK) – but only in animals not receiving  $\alpha$ -tocopherol. Kinase activity was lower in groups subjected to the training and receiving  $\alpha$ -tocopherol for 10 days, which indicated explicitly that  $\alpha$ -tocopherol reduced the intensity of muscle fibre degradation processes.

The training also promoted an increased concentration of HDL-cholesterol and a reduction of the concentration of lipid proteins LDL+VLDL in the plasma regardless of the application of  $\alpha$ -tocopherol.

Progressive exercise caused an increased concentration of  $\alpha$ -tocopherol in the plasma but only in the ET group (receiving  $\alpha$ -tocopherol) while it significantly reduced the content of  $\alpha$ -tocopherol in the liver ( $p \leq 0.001$ ) irrespectively of the application of  $\alpha$ -tocopherol. The feeding of  $\alpha$ -tocopherol to animals for 10 days caused a significant increase in its concentration in the tissues. Significant influence of the supplied  $\alpha$ -tocopherol and training on the increased concentration of  $\alpha$ -tocopherol was found in the heart and muscle. It indicates that, due to exercise,  $\alpha$ -tocopherol is released from the liver while more of it is accumulated in tissues such as the heart and the muscle where the blood flow and, therefore, the flow of oxygen, is intensified. It can also result from an intensified absorption of the supplied  $\alpha$ -tocopherol in organisms subjected to systematic exercise. Obtained results explicitly confirm the influence of systematic exercise on an increase in the content of  $\alpha$ -tocopherol in the heart and muscles.

When evaluating the impact of studied factors on the activity of antioxidant enzymes, significant influence of progressive exercise on the SOD activity in erythrocytes and in the heart was demonstrated (respectively:  $p \leq 0.02$  and  $p \leq 0.01$ ). However, no influence of  $\alpha$ -tocopherol on the SOD activity in erythrocytes was found. Neither progressive exercise nor  $\alpha$ -tocopherol influenced the activity of GPx in the plasma, liver and heart. A more intense GPx activity was only found in the muscle in the trained control group. Probably,  $\alpha$ -

tocopherol triggers adaptation changes in the antioxidant defence system because RFT can be removed by antioxidants and this phenomenon, in turn, reduces the need for an induction of antioxidant enzymes. However, a reduced activity of the enzymatic defence system after physical exercise can be due to a high output of free radicals, significantly exceeding the detoxicating ability of antioxidant enzymes. As a result, the intracellular redox system changes, catalytic centres of enzymes are modified and, as a consequence, their activity is inhibited.

Physical exercise is a strong inductor of lipid peroxidation demonstrated in the research conducted in people and animals. As a part of this work, lipid peroxidation level was determined with the use of the TBARS level measurement, which is considered a reliable marker of oxidation damage resulting from physical exercise. TBARS concentration in the plasma and tissues of animals subjected to progressive exercise significantly varied from one group to another. The factor determining the analysed indicator in the plasma and in the muscle was the supply of  $\alpha$ -tocopherol (respectively:  $p \leq 0.03$  and  $p \leq 0.02$ ) that reduced the lipid peroxidation level. Higher concentrations of TBARS in the plasma and muscle of animals from control groups indicate that the degree of antioxidation defence was reduced in the 10-day period during which rats did not get  $\alpha$ -tocopherol with their diet, which is related to the above-mentioned lower level of  $\alpha$ -tocopherol in the plasma and the intensification of peroxidation processes in the muscle during progressive exercise. The level of lipid oxidation products in the heart was higher in trained groups than in corresponding untrained groups, which is indicative of the significant influence ( $p \leq 0.03$ ) of exercise on an increased TBARS concentration in the heart. TBARS concentration in the liver was higher in the trained groups receiving  $\alpha$ -tocopherol than in the untrained group and did not significantly differ from the control group, which is a sign of the influence of training on the level of lipid peroxidation.

Summing up, it was found that exercise combined with  $\alpha$ -tocopherol deficient diet caused more damage to muscles and an intensification of lipid oxidation processes. The training influenced the activation of enzymatic systems even though the reaction in analysed tissues was varied. In particular, the supply of  $\alpha$ -tocopherol caused its increased concentration in the plasma and in analysed tissues and, as a consequence, influenced a lower level of damage to muscles and the lower oxidation of lipids in the plasma and

muscles of animals supplemented with  $\alpha$ -tocopherol. It allows us to conclude that the supply of  $\alpha$ -tocopherol reduced the oxidative stress level induced due to physical exercise and normalized the redox condition. Based on the obtained results it was found a significant role of administration  $\alpha$ -tocopherol in preventing the effects of free radicals, resulting from disturbed oxidation-reduction balance by physical exercise.

***Comparison of the influence of  $\alpha$ -tocopherol acetate and different physical exercise on the redox homeostasis***

Values of haematological indicators and glucose concentration in the plasma of studied animals did not depend on the analysed factors, i.e. parameters of experiments I and II, varied physical exercise and  $\alpha$ -tocopherol. However, it was found that physical exercise significantly influenced ( $p=0.03$ ) an increase in the concentration of the CRP protein in the blood.

In general, physical exercise caused a significant ( $p=0.001$ ) increase in the CK activity while the supply of  $\alpha$ -tocopherol significantly ( $p=0.01$ ) reduced it. CK activity in the plasma was negatively correlated with the concentration of  $\alpha$ -tocopherol in the muscles, both after one-time and after progressive exercise, which is clearly indicative of a significant role of  $\alpha$ -tocopherol in the protection of muscles.

When compared with one-time exercise, the training caused beneficial changes in the lipid profile of the plasma of rats visible in the higher level of HDL-cholesterol and the lower LD+VLDL.

Subjecting animals to the training and the longer period of  $\alpha$ -tocopherol supply caused a statistically significant increase in the concentration of  $\alpha$ -tocopherol in the plasma and analysed tissues when compared to animals subjected to one-time exercise. It was observed that, with the 10-day diet with  $\alpha$ -tocopherol and training combined, the concentration of  $\alpha$ -tocopherol in the liver was reduced while it increased in the muscle. It indicates that physical exercise causes a release of tissue reserves of  $\alpha$ -tocopherol and its redistribution among tissues in order to deliver it to tissues more engaged in the physical exercise.

When analysing the influence of experimental conditions on the activity of antioxidant enzymes (SOD and GPx) in the blood and tissues, they were only found to significantly influence the activity of SOD in erythrocytes and the heart and on GPx in the blood. The activity of enzymes was higher in groups subjected to one-time exercise in comparison with trained animals. The supply of  $\alpha$ -tocopherol significantly ( $p < 0.0001$ ;  $p = 0.01$ ) increased the SOD activity in erythrocytes and reduced GPx activities in the muscle. The activity of antioxidant enzymes was higher in groups subjected to one-time exercise in comparison with trained animals, which is a sign of the intensified oxidative stress and the activation of defence mechanisms during such exercise. In turn, the lower activity of analysed enzymes in the blood of trained animals in comparison with animals subjected to one-time exercise can indicate their adaptation to exercise or the activity of non-enzymatic defence systems, which partly confirms hypothesis 2.

It was demonstrated in this work that both one-time and progressive exercise caused an increased TBARS concentration in the material collected immediately after the exercise, in particular, in the plasma, heart and muscle. The concentration of lipid peroxides in the plasma of animals subjected to the training was higher in comparison with animals subjected to one-time intense exercise. In turn, the supply of  $\alpha$ -tocopherol significantly reduces the concentration of TBARS in the plasma ( $p \leq 0.04$ ) and muscles ( $p \leq 0.008$ ). However, no influence of  $\alpha$ -tocopherol on TBARS concentrations in the heart was observed, which can result from a too low dose of  $\alpha$ -tocopherol or an insufficient concentration of other co-antioxidants in a cell as well as from very intense oxidation processes in that tissue.

The concentration of lipid peroxides in the plasma did not correlate with their concentration in tissues. Additionally, it was observed that the increased TBARS concentration in the heart during the training was positively correlated with the SOD and GPx activity in the heart, and the increased TBARS concentration in the liver during the one-time intense exercise promoted a reduced GPx activity in the liver. Dependencies between TBARS concentration and the activity of antioxidant enzymes in rats subjected to physical exercise of varied intensity and duration confirm the differentiated response of cells to the appearing oxidative stress and are indicative of the mobilization of the enzymatic defence system. Such response depends on the type, intensity and duration of physical exercise. Thee research indicated that one-time intense physical exercise strengthens oxidative stress

and causes damage to the muscles while the one-time supply of  $\alpha$ -tocopherol reduces the damage level in muscles and the peroxidation of lipids, which partly corroborates the first research hypothesis. Additionally, it was demonstrated that  $\alpha$ -tocopherol was reducing both TBARS concentrations in the muscles and their damage levels measured with the activity of creatine kinase in both types of exercise.

### 3.4. CONCLUSIONS

Obtained results confirm the physical exercise in untrained organisms intensified oxidative stress in the tissues and increased damage levels in the muscles. One-time intense physical exercise caused a significant increase in the activity of antioxidant enzymes in the blood and heart in comparison with systematic training. The duration of exercise influenced the oxidation of lipids in the plasma, with higher concentrations observed in progressive training. In particular, the supply of  $\alpha$ -tocopherol reduced damage to the muscles and lipid peroxidation irrespectively of the type of exercise. Therefore,  $\alpha$ -tocopherol is of significant importance for previously untrained subject, especially when it comes to a reduction of oxidative damage to muscles. The training combined with the supply of  $\alpha$ -tocopherol acetate caused an increase in the  $\alpha$ -tocopherol concentrations in the plasma, heart and muscles resulting from the mobilization of lipid reserves in the body and  $\alpha$ -tocopherol redistribution between tissues. The observed redistribution of  $\alpha$ -tocopherol between tissues and the influence of  $\alpha$ -tocopherol on the reduction of damage to muscles is another proof of its antioxidative properties.

Obtained results are exploratory and enrich existing knowledge of the influence of physical exercise on the untrained young subject and its requirement for antioxidants. Observed dependencies between  $\alpha$ -tocopherol and the activity of antioxidant enzymes in the blood and tissues require further research.

In particular, research results increased knowledge of the oxidative stress induced by physical exercise in young, growing organisms. In the existing literature, the research of physical exercise conducted in animal or human models mainly refers to adult or aging bodies. Observed dependencies can be used to develop dietary recommendation regarding the consumption of vitamin E by children and adolescents who begin sports training, which can be considered a practical component of the presented work.

#### 4. OVERVIEW OF THE OTHER SCIENTIFIC ACHIEVEMENTS

My first achievement was the master thesis titled *Determination of the content of resistant starch in selected food products* in which I adapted the Champ method in the Faisant modification to the conditions of the laboratory of the Chair of Nutritional Assessment. This work was the first one in Poland to determine resistant starch as one of the tractions of dietary fibre (**II D 34 and 35**).

Having graduated from the Master's studies, I worked in the Food Technology Laboratory of the National Food and Nutrition Institute in Warsaw, where I was determining *trans* isomers of essential fatty acids in food products using the method of gas chromatography combined with mass spectrometry (GC-MS). The work in the team of doctor Marek Daniewski made it possible for me to learn the modern precise chemical analysis, which significantly broadened my knowledge and enriched my analytical skills. It was the first institution in Poland to tackle this subject, implementing the evaluation of the content of *trans* isomers in products as a new component of the nutritional evaluation of fats. The obtained data constituted the source database of the content of groups of fatty acids in products, being the basic component of the ingested food ration in a period when the consumption of vegetable fats and confectionery significantly increased in Poland. Obtained results indicated that confectionery fat used in the production of confectionery as well as sweets that are very popular among children and adolescents constituted a significant source of *trans* isomers. Results of the published papers referring to this subject contributed to an improvement of the quality of fat used in food production, the introduction of the "content of trans isomers" to the information on packages and, in particular, to the dissemination of knowledge about the noxious influence of *trans* isomers on the body and the need to reduce their consumption.

Additionally, I participated in the research of the application of chemical positive ionisation for the identification of methyl esters of essential fatty acids with the use of the gas chromatography combined with mass spectrometry.

Most important publications on the discussed subjects: **II D 27-31, II D 33**.

In 2000 I started PhD studies at the Faculty of Human Nutrition and Consumer Sciences at the Warsaw University of Life Sciences during which my scientific interests

focused on the assessment of changes in the quality of nutrition occurring in Poland in the 20-year period when we passed from the centrally planned economy to the free market economy, which promoted changes in the consumption of nutritional components. This issue became the subject of the doctoral thesis titled: *Assessment of the nutritional quality in Poland in years 1980-2000*. The results obtained indicated that the quality of nutrition in Poland changed significantly. It was found that the consumption of most groups of food products dropped in the analysed 20-year period and, as a consequence, the nutritional value of the daily consumption was reduced. The quality of nutrition of the Polish society measured with the INQ (Index of Nutritional Quality) indicator deteriorated, especially after the introduction of the free market economy. In 2004 I received my PhD degree.

I reported on the above-mentioned results to the PhD International Conference in Miskolc (Hungary) in 2001 and 2003 (**III B 18 and 19**). Selected results were also published in: **II D 23-26** and presented at the following conferences: **III B 14-17**.

I began my professional career in the Chair of Nutritional Assessment at the Faculty of Human Nutrition and Consumer Sciences as an assistant in October 2004. After a year (October 2005) I was appointed to the position of an assistant professor.

The profile of my research interests was changing with the progress of my scientific work. As a whole, the subject areas of my research so far has been concentrating on the following issues:

- The role of antioxidants in the maintenance of homeostasis in the body subjected to physical exercise,
- Nutrition of selected population groups in the aspect of the prevention of diet-dependent diseases.

#### **4.1. THE ROLE OF ANTIOXIDANTS IN THE MAINTENANCE OF HOMEOSTASIS IN THE BODY SUBJECTED TO PHYSICAL EXERCISE**

An important topic taken up in 2007 analysed the influence of antioxidants, in particular,  $\alpha$ -tocopherol on oxidative stress induced by physical exercise in growing

organisms, previously untrained. The research was conducted on the animal model, i.e. male rats subjected to the training on a treadmill. The literature lacks data concerned with oxidative damage induced by physical exercise in young organisms. This type of research most frequently uses adult, aging or ill organisms (e.g. with diabetes, cardiovascular disease). Oxidative damage to tissues in young organisms is suspected of having graver consequences than in adult organisms due to the less developed defence mechanisms. In the light of the fact that the issue of physical exercise and the supply of antioxidants on the oxidation and reduction balance and its health consequences in growing organisms has not been explored, it seems that the topic is very significant if we take it into account that many boys start training intensely when still very young.

The goal of the first stage of the research was the assessment of the influence of physical exercise and the supply of varied doses of  $\alpha$ -tocopherol on the level of oxidative stress in growing rats (male). Within the work performed a biological experiment, determination of biomarkers of oxidative stress, i.e. the concentration of  $\alpha$ -tocopherol in the plasma and tissues, lipid peroxides and the antioxidant potential in the blood plasma. The research conducted on 48 male Wistar rats divided into 8 groups fed differentiated doses of  $\alpha$ -tocopherol (0; 0.5; 1.0; 4.0 mg/d). A half of the animals were subjected to 15-minute exercise on a treadmill (belt speed 20m/s) for 10 days. In order to attain the goal of the research, I adapted the method for the determination of  $\alpha$ -tocopherol in the plasma and tissues with the use of the high-efficiency liquid chromatography method (HPLC-UV) and the spectrophotometric method for the determination of lipid peroxides (TBARS method) and antioxidant status (ABTS method) in the plasma. Results showed that the concentration of  $\alpha$ -tocopherol in the plasma was increasing with the supplied doze and was highest when 4 mg of  $\alpha$ -tocopherol was supplied. Physical exercise significantly differentiated the analysed groups; the concentration of  $\alpha$ -tocopherol in the blood plasma of animals subjected to exercise was significantly higher. Additionally, physical exercise caused an intensified mobilization of  $\alpha$ -tocopherol from the liver to the blood in groups subjected to exercise, which was reflected in higher values of  $\alpha$ -tocopherol concentrations in the plasma and lower ones in the liver. The increased concentration of  $\alpha$ -tocopherol in the plasma of these animals was positively correlated with the value of the antioxidant status. The concentration of lipid peroxides in the plasma in groups subjected to physical exercise was negatively correlated

with the dose of  $\alpha$ -tocopherol; in groups not subjected to exercise, this dependency was reversed, which makes it possible to presume that an excessive supplementation of  $\alpha$ -tocopherol in untrained organisms can intensify oxidative stress (**II A 2, II D 19**).

Research on the role of tocopherol in reducing oxidative stress I had continued as head of the faculty grant on: *The role of differentiated doses of  $\alpha$ -tocopherol in the maintenance of DNA stability* (**II I 2**). The aim of the research was to determine the influence of differentiated doses of  $\alpha$ -tocopherol on its concentration in the liver and on DNA protection measured with the quantity of oxidized guanine (8-OhdG) in the liver of rat. This tissue was selected because of its role in the storage of  $\alpha$ -tocopherol and energy substrates (glucose and fat) for muscles engaged in physical exercise. The beneficial influence of physical exercise on the body and the role of antioxidants in the prevention of oxidation damage of skeletal muscles as a result of intensified physical activity have been described for many years, but molecular mechanisms in the tissues not directly engaged during exercise in young organisms have not been explored; this is why the research topic is very important. The obtained results showed that an increased supply of  $\alpha$ -tocopherol that reduced oxidation damage to the DNA in the liver was very important in young, growing organisms subjected to the training.

In the following years, the influence of antioxidants, i.e.  $\beta$ -carotene,  $\alpha$ -tocopherol and ascorbic acid on oxidative stress due to physical exercise was being explored as a part of the research project financed by the Committee for Scientific Research titled: ***Interactions of antioxidant vitamins in DNA protection from oxidative stress*** (PB MNiSW N N312 410037; project manager: dr hab. Agata Wawrzyniak, prof. SGGW; 2009–2012) in which I was a co-author (**II I 1**). The research used the animal material, i.e. 144 male rats who were fed the above-mentioned antioxidants individually in the following doses:  $\alpha$ -tocopherol - 2 mg/d, ascorbic acid – 12 mg/d,  $\beta$ -carotene – 1 mg or 2 or 3 of them combined for 14 days. A half of the animals from each group (n=8) was trained on a treadmill for 15 minutes daily in order to induce oxidative stress. After biological experiment, I determined the concentration of  $\alpha$ -tocopherol in the plasma and liver of the collected biological material using the HPLC-UV method and lipid peroxides in rat testicles. The research made it possible to deepen knowledge of the role of studied antioxidants in the protection of DNA and lipids from oxidation damage and was indicative of the role of  $\alpha$ -tocopherol as a biomarker of oxidative

stress. It was demonstrated that  $\alpha$ -tocopherol counteracted the oxidation of lipids in the plasma (determined with the use of the TBARS method) and reduced oxidation damage to the DNA measured as the quantity of 8-OHdG. Additionally, obtained results indicated that the simultaneous supply of antioxidants reduced the expression of the p53 protein that plays the key role in the adjustment of the cellular cycle by way of the initiation of the process of repair or promoting the apoptosis of the cell. The conducted research was the first in the world to study the influence of the simultaneous supply of  $\beta$ -carotene,  $\alpha$ -tocopherol and ascorbic acid on the expression of the p53 protein and obtained results deepened knowledge of molecular mechanisms related to the protective activity of supplemented vitamins (**II A 1; II D 4; III B 7, 10-12**).

The project also included the research of the influence of physical exercise and the supply of  $\alpha$ -tocopherol, ascorbic acid and  $\beta$ -carotene on the level of tocopherol in the plasma of rats. Results showed that physical exercise caused a significant increase in the concentration of  $\alpha$ -tocopherol in the plasma of trained animals and the highest level of  $\alpha$ -tocopherol was observed in groups of trained animals getting all of the vitamins or the combination of  $\beta$ -carotene and vitamin C. It confirmed that the combined supply of these compounds is more effective due to their mutual interactions. I presented the results of this part of the research at the 22<sup>nd</sup> Polish National Bromatology Symposium in Wisła in 2012 where I was granted a distinction (**III B 9**).

Another sub-topic was the influence of  $\beta$ -carotene,  $\alpha$ -tocopherol and ascorbic acid supplementation and of physical exercise on the level of lipid peroxides in testicles of rats. The topic is very important in the light of the problem related to the growing percentage of sterile young men who, more frequently than women, training. Perhaps the phenomenon is also related to the early commencement of sports training at school. The determined level of lipid peroxidation in testicles indicated that physical exercise intensified the oxidation of lipids, with their concentration higher by 11-32% in groups of trained rats in comparison with untrained groups. I also demonstrated that the effectiveness of the supplementation of 2-3 vitamins was higher when it comes to the prevention of oxidation damage of lipids in testicles of rats (**III B 8**). Obtained results constitute a material for further studies.

My achievements in the field of research I used to develop approaches to conducting research and preparing the dissertation discussed above.

For the last few years, I have focused on the assessment of the consumption of antioxidant vitamins in selected population groups. Antioxidant vitamins play the key role in the maintenance of redox homeostasis and, if supplemented in sufficient quantities, can counteract the effects of an excessive production of free radicals. In turn, disturbances of the oxidation balance underlie the appearance of diet-dependent diseases such as: tumours, coronary disease, diabetes or neurodegenerative diseases. Therefore, the research assessing the consumption level of these compounds are of vital importance to the development of dietary recommendations aimed at the prevention of above-mentioned diseases.

When evaluating the consumption of antioxidant vitamins (A, C, E) in a group of students (n=145), I indicated their appropriate quantities in food rations except for the group of smokers in whose case the consumption was demonstrated to be too low when compared with the existing recommendations, which increases the risk of diseases related to oxidative stress in that group **(II D 14)**.

Research under my supervision was conducted among children aged 10-12 from sports classes (n=67). It was demonstrated that the average supply of vitamin E in the analysed group of pupils was insufficient; it amounted to 6.4 mg/day. In turn, the average consumption of vitamin C in the analysed group amounted to 129 mg/day, which demonstrates its sufficient consumption. I observed similar dependencies in the research involving women practicing fitness exercises (n=52) whose food rations covered the demand for vitamin E only in 44% of women; it can be disadvantageous in the light of the antioxidant role of that vitamin. However, the consumption of vitamin C was appropriate. The obtained results, which I treat as pilot ones, are indicative of the need to introduce further extended research of quantities of antioxidants consumed in groups of physically very active people **(II D 3; II K 1; III B 3)**.

In 2014, I received the Award of HM Rector of the Warsaw University of Life Sciences for this research area (Estimation of health opportunities and threats resulting from the consumption of antioxidant vitamins).

#### 4.2. NUTRITION OF SELECTED POPULATION GROUPS IN THE ASPECT OF THE PREVENTION OF DIET-DEPENDENT DISEASES

The nutritional assessment in different groups of people, with a particular stress on children and adolescents was an important topic of the conducted research. The nutrition monitoring is a important aspect of the prevention of diet-dependent diseases and promotes to create of nutritional recommendations.

When assessing kindergarten food rations, many irregularities were found irrespectively of the status of such institutions (public/non-public kindergartens). Five decade menus served in kindergartens in Warsaw, Dziekanów, Żyrardów, Łomianki, Nowa Iwiczna and four menus served in kindergartens in Zbuczyna, Krakow and Siedlce (4 public kindergartens and 4 non-public ones) were evaluated. The evaluation of kindergarten food rations showed dietary mistakes related to an excessive supply of energy, protein, vitamins A and B<sub>2</sub> in comparison with the requirements. Kindergarten food rations did not cover needs for vitamin C, D and calcium. Results were presented in: **II D 16, III B 2**.

Dietary mistakes of older children can result from the lack of understanding of nutritional recommendations communicated to them, e.g. the food pyramid. This is why the subject consisted of the understanding and practical application of recommendations contained in the food pyramid for children was taken. The research was conducted among schoolchildren (10-11 years; n=100). It was demonstrated that nutritional recommendations presented in the form of a pyramid are not entirely comprehensible for children. Only 40% of respondents could use the information contained in the pyramid to plan a correct daily menu. It was also a problem to classify product in individual product groups comprising the food pyramid. Both results referring to the evaluation of kindergarten food rations and results concerned with the understanding and practical application of nutritional recommendations by older children (**II D 6 and 9; III Q 2.1**) indicated the need to intensify the activity focused on nutritional education both among children and employees of educational institutions, especially as regards the practical application of nutritional recommendations. Obtained results were disseminated as a part of the training courses and as publications (**III I 5 and 6; III M 1**).

The next aspects of nutrition in selected population groups was to determine nutrition of young women and men. Obtained results showed the existence of numerous

nutritional mistakes. It was demonstrated that only 12% of students (n=143) consumed an appropriate quantity of energy from total fats and the recommended level of cholesterol was significantly exceeded (by 82%) in food rations of men, which increased the risk of cardiovascular diseases. In turn, deficiencies of iron and folates were found in women aged 21-28 (n=121); their respective consumption amounted to 9.7 mg/d and 270 µg/d. Such a low supply of iron and folates resulting from an insufficient energy value of food rations as well as the structure of consumed products is particularly dangerous for women at reproductive age, especially in the context of anaemia and the prevention of neural tube defects in the offspring (**II D 17, 18, 20**).

The other works in nutritional assessment field focused on the comparing the consumption of drinks containing caffeine (coffee, drinks such as coca cola, energy drinks) and isotonic drinks in the group of pupils of junior high schools (n=90) and university students (n=100). The obtained results indicated that they were popular in both groups. 42% of respondents were consuming isotonic drinks with male respondents consuming more of them, probably due to their more intense physical activity. The most frequently declared reasons for the consumption of isotonic drinks included an improvement of physical capability and the quenching of thirst (**II D 5, 7; III B 4**).

Drinks such as coca cola were another type of a popular drink containing caffeine, consumed by 97% of junior high school pupils and 93% of students. The consumption of coffee and energy drinks was increasing with age. Sex was a significantly differentiating factor when it comes to the consumption of energy drinks; men declared a more frequent consumption with differentiated consumption determinants. Junior high school pupils more frequently declared that they consumed energy drinks during and after physical exercise to improve physical capacities while students mostly consumed them when learning in order to improve the concentration and mental capacity. It was confirmed by results of subsequent research involving students (n=92), where a significantly higher consumption of energy drinks was demonstrated during the examination session in comparison with the consumption throughout the academic year (**II D 10, 13**).

The consumption of drinks containing caffeine or, as in the case of energy and isotonic drinks, other bioactive substances and vitamins from the B group requires the monitoring of consumed quantities because their combined supply with various products

can cause an excessive consumption and disadvantageous health impact. Analysed students experienced stimulation (73%), palpitation (32%) and insomnia (26%) after the consumption of energy drinks. Persons consuming energy drinks considered them effective (89%), tasty (42%) and dangerous for health (44%). A great majority of respondents (81%) used to mix energy drinks with alcohol. Every fourth respondent did not read the information about the content of consumed drinks, which undoubtedly demonstrates the need to improve nutritional awareness. Further research is also required with regard to the observed dependency between the consumption of energy drinks in connection with physical exercise, especially in younger age groups.

Research comparing the nutrition of university students (n=92) in Poland and Norway was also conducted. It was found that the nutrition of the Polish students did not meet recommendations more and, when combined with poor physical activity, increased the risk of diet-dependent diseases in the future (**II D 2, III B 1**).

Results of the research conducted are clearly indicative of the need to intensify the activities promoting the practical ability to apply knowledge gained as well as promoting physical activity and healthy dietary habits regardless of the age. To improve the health condition of the Polish society in the future, the scope of the dietary education should be extended among children, adolescents and students at all types of universities. Even though further research is required, the obtained results also indicate that it is necessary to propagate nutritional recommendations to reduce the risk of diet-dependent diseases later.

Cardiovascular diseases constitute one of the main causes of death both in Poland and in Europe, especially among men. An increased risk of cardiovascular diseases (CVD) depends on many factors; nutritional ones include the quantity of consumed fat, cholesterol, the quantity and type of fatty acids, fibre, antioxidant vitamins, potassium, calcium and magnesium. Therefore, another research focused on the nutrition and selected aspects of nutritional state of patients with cardiovascular diseases, aged 30 – 90 (n=95). In particular, an incorrect body weight was found in 82% of respondents along with an excessive supply of fat and an insufficient supply of calcium, magnesium, potassium and antioxidant vitamins. The lipid profile in the blood of respondents was incorrect. Despite the application of medications reducing lipid levels, the concentration of LDL-cholesterol and total cholesterol exceeded target levels for people with CVD. Additionally, the research demonstrated that

20% of patients did not meet any dietary recommendations for people with CVD while more than 40% of them was not aware of the role of the nutrition in the secondary prevention of cardiovascular diseases (**II D 8; III Q 2.2**). In the light of the role of nutrition both in the primary and secondary prevention, it seems necessary to take action in order to improve this situation and educate the patients.

In Table 1 a list of published works together with points was presented.

**Table 1. A list of published papers with the Ministry of Science and Higher Education and IF points (\*according to the journal list for publishing years)**

CATEGORY	Number of papers			Sum*
	Before PhD	After PhD	All	
Original publications Journal Citation Report (A list)				
Nutrition Research		1	1	20 IF = 2,142
Polish Journal of Veterinary Sciences (15, 20)	-	2	2	35 IF= 0,435 (2009) IF= 0,565 (2011)
Publications in reviewed journals (B list)				
Polish Journal of Food and Nutrition Sciences (9, 10)	-	2	2	19
Bromatologia i Chemia Toksykologiczna (4 i 6)	2 (4)	2 (6)	4	20
Postępy Higieny i Epidemiologii (7)		1	1	7
Roczniki PZH (4, 6, 7, 9)	2 (4)	5 (9), 3 (7), 3 (6)	13	92
Żywnienie Człowieka i Metabolizm (3,4)	4 (3), 1 (4)	1 (4)	6	20
Nowiny Lekarskie (6)	-	1	1	6
Other publications				
Kwartalny Biuletyn Polskiego Towarzystwa Dietetyki	1		1	
	10	21	31	219
Publications in scientific monographs in Polish:				
to 10 pages (4 pkt.)		6	6	24
Publications in scientific monographs in basic discipline language:				
to 10 pages (5pkt.)		1	1	5
Abstract in reviewed journals:				
Annals of Nutrition & Metabolism	-	1	1	
Abstract in reviewed conferences materials:				
	4	11	15	
				248

## II. TEACHING ACTIVITY

During PhD studies, I gave classes in the following subjects: Food Analysis, Biological Assessment of Food at full-time and part-time studies (field of study: food technology and human nutrition). After obtaining the doctoral degree and starting work in the Department of Human Nutrition at the Warsaw University of Life Sciences, I started to lecture in the following subjects: Food Analysis, Basics of Nutritional Assessment, Basics of Nutrition, Current Trends in Human Nutrition to students of food technology and human nutrition, human nutrition and food assessment, dietetics as well as gastronomy and hotel management.

By decision of the Faculty Council, I was entrusted with the giving of lectures related to Nutritional Assessment Basics for the full-time and part-time students of the Faculty of Human Nutrition and Consumer Sciences.

I am author of labs programs of Basics of Nutritional Assessment for full-time and part-time students at the faculties such as Food Technology and Human Nutrition, Human Nutrition and Food Assessment as well as Dietetics. I also prepared instructions for labs regarding: *The application of reference values to the assessment of a nutrition (assessment of the macro nutrients, vitamins, mineral components, salt, water intake) on an individual and group levels and assessment of nutritional state using anthropometrical indicators.*

In 2014, I drew up labs program in Current Trends in Human Nutrition for master studies at the faculty of Human Nutrition and Food Assessment.

Since 2008 I have been a supervisor 25 engineer theses and 11 master theses. I have also been taking part in diploma exams at various studies at the Faculty and I have been engineering and master theses' reviewer.

In 2010 and 2012, I led the workshop for stewards on: *The adequate nutrition of children and composition of nutritional menus for employees of primary schools and kindergartens responsible for child nutrition* organized under the patronage of the City of Warsaw Education Department. As a part of this training, I conducted practical classes in the planning of kindergarten menus for about 100 people employed in Warsaw institutions (**III A 2**). In 2013, I participated in the Polish national educational and research program titled *Mądre żywienie, zdrowe pokolenie* for which I developed a lesson for junior high and high school pupils titled Breakfast for health (**III A 3**) and in the Polish national campaign *Z miłości*

*do zdrowia* in 2014 (coordinator: Carrefour) for which I prepared leaflets about principles of healthy nutrition (**III A 4**). As of January 2015, I have been participating in the MNiSW program (Innovative Economy) – the University of Young Inventors – the project titled *Good Nutritional Practice of Junior High School Pupils* for which I prepare and conduct workshops for junior high pupils on rational nutrition and research of nutrition (**III A 5**). Additionally, I am the leader of the dietary block of the Polish national project of the Carrefour Foundation “ABC of Healthy Nutrition” that began at the end of 2014 (**III A 6**).

To improve my professional skills, I participated in the training courses and conferences: **III Q 3**.

In recognition of my educational achievements, I received the 1<sup>st</sup> grade Award of HM Rector in 2012 and a diploma of merit in 2014.

### **III. POPULARIZING SCIENCE ACTIVITIES**

My teaching activities outside the university focused mainly on conducting workshop for children and a youth about the principles of rational nutrition and lectures on the role of diet in maintenance of genome stability. Details are presented in Appendix no.3, point **III.I. 7**

### **IV. ORGANIZATIONAL ACTIVITIES**

In addition to research and teaching activities, I try to get involved in organizational activities of the Faculty and the University. So far, I performed the following functions:

- in 2005-2006, I participated in the work of the Faculty Admissions Committee
- I was a member of the Organizing Committee of the IX National Nutrition Workshop in Rogów (2006) and the Scientific-Training Conference *O zdrowie i prawidłowy rozwój dzieci i młodzieży oraz pełnosprawność psychofizyczną osób starszych* (2008)
- since 2006 I have been participating in the work of the Olympiad Examination Committee of Agricultural Knowledge and Skills; block Human Nutrition and Household,

- since 2008 I have been participating in the Days of Warsaw Agricultural University, in which I have been preparing materials and stand for anthropometric measurements and develop competitions for children on nutrition,
- in 2010 – 2014 I was a vintage supervisor of full-time students of the Faculty of Human Nutrition and Consumer Science
- since 2013 I have been a member of the Committee on Faculty Affairs Promotion Department,
- in 2013 I participated as member of Examination Committee of the Review of Students Scientific Associations SGGW
- since 2006, I am a member of the Polish Society of Nutritional Sciences.

**V. COOPERATION WITH INSTITUTIONS, ORGANIZATIONS AND SCIENTIFIC SOCIETIES**

While working at the Faculty of Human Nutrition and Consumer Science WULS, I have been working with the Faculty of Animal Science, the Faculty of Veterinary Medicine, and the Faculty of Production Engineering WULS, which resulted in joint publications (**II A 1-3, II D 2, 11, 12, 22**). Moreover, now I have been participating in research in the national project ABC Healthy Nutrition.

