AGRICULTURAL
SCIENCE AND TECHNOLOGY

2014

An International Journal Published by Faculty of Agriculture,
Trakia University, Stara Zagora, Bulgaria
Scope and policy of the journal

Agricultural Science and Technology (AST) – an International Scientific Journal of Agricultural and Technology Sciences is published in English in one volume of 4 issues per year, as a printed journal and in electronic form. The policy of the journal is to publish original papers, reviews and short communications covering the aspects of agriculture related with life sciences and modern technologies. It will offer opportunities to address the global needs relating to food and environment, health, exploit the technology to provide innovative products and sustainable development. Papers will be considered in aspects of both fundamental and applied science in the areas of Genetics and Breeding, Nutrition and Physiology, Production Systems, Agriculture and Environment and Product Quality and Safety. Other categories closely related to the above topics could be considered by the editors. The detailed information of the journal is available at the website.

Proceedings of scientific meetings and conference reports will be considered for special issues.

Submission of Manuscripts

All manuscripts written in English should be submitted as MS-Word file attachments via e-mail to editoffice@agriscitech.eu. Manuscripts must be prepared strictly in accordance with the detailed instructions for authors at the website www.agriscitech.eu and the instructions on the last page of the journal. For each manuscript the signatures of all authors are needed confirming their consent to publish it and to nominate on author for correspondence.

They have to be presented by a submission letter signed by all authors. The form of the submission letter is available upon from request from the Technical Assistance or could be downloaded from the website of the journal. Manuscripts submitted to this journal are considered if they have submitted only to it, they have not been published already, nor are they under consideration for publication in press elsewhere. All manuscripts are subject to editorial review and the editors reserve the right to improve style and return the paper for rewriting to the authors, if necessary. The editorial board reserves rights to reject manuscripts based on priorities and space availability in the journal.

The journal is committed to respect high standards of ethics in the editing and reviewing process and malpractice statement. Commitments of authors related to authorship are also very important for a high standard of ethics and publishing. We follow closely the Committee on Publication Ethics (COPE), http://publicationethics.org/resources/guidelines

The articles appearing in this journal are indexed and abstracted in: EBSCO Publishing, Inc. and AGRIS (FAO). The journal is accepted to be indexed with the support of a project № BG051PO001-3.3.05-0001 “Science and business” financed by Operational Programme “Human Resources Development” of EU. The title has been suggested to be included in SCOPUS (Elsevier) and Electronic Journals Submission Form (Thomson Reuters).

Address of Editorial office:
Agricultural Science and Technology Faculty of Agriculture, Trakia University Student’s campus, 6000 Stara Zagora Bulgaria
Telephone.: +359 42 699330 +359 42 699446
www.agriscitech.eu

Technical Assistance:
Nely Tsvetanova
Telephone.: +359 42 699446
E-mail: editoffice@agriscitech.eu
**Effect of vitamin E in the diet of pigs on the lipid and protein oxidative stability of meat during storage**

T. Popova*, P. Marinova, M. Ignatova

Institute of Animal Science, 2232 Kostinbrod, Bulgaria

**Abstract.** The changes in lipid and protein oxidative stability in response to vitamin E supplementation in the diet (400 mg/kg feed) during storage of Musculus Longissimus dorsi and Musculus Semimembranosus were studied in ♀Youna x ♂ Pietrain pigs. Muscle samples were stored for 6 days at 4°C after which the storage continued for up to 90 days at –20°C. Lipid oxidation was analysed by determination of 2-thiobarbituric acid reactive substances (TBARS) formed during storage of 48 h, 6 days and 90 days. The oxidation of proteins was determined by measuring the content of carbonyls formed during the same storage periods. The muscles of the pigs from the vitamin E supplemented group showed lower TBARS content for 48 h and 6 days (P<0.05). Significant influence of the vitamin E was observed after frozen storage (P<0.01) as well and lower levels of TBARS were displayed in the muscles from the supplemented group. Contrary to the lipid oxidation, vitamin E supplementation of the diet only tended to decrease the oxidation of the proteins in the muscles during the storage. However, carbonyl formation differed between the muscles. During the refrigerated storage Musculus Longissimus showed higher extent of carbonylation, whereas after frozen storage the carbonyl content was increased in Musculus Semimembranosus. In both muscles lipid and protein oxidation showed the same trends of development in the control and supplemented group, showing maximal content of TBARS and carbonyls after the 90th day of frozen storage.

**Keywords:** pigs, vitamin E, 2-thiobarbituric acid reactive substances, carbonyls, storage, meat

**Introduction**

During the transformation of the muscles in meat after the slaughter of the animals, changes in the balance of pro- and antioxidant systems in the tissues occur, which initiates oxidative processes in lipids and proteins in meat. Lipid oxidation is one of the major causes for negative changes in meat quality traits and the formation of compounds that might be harmful for the human health. The oxidizing lipids, together with metal ions and other prooxidants might as well be one of the main factors to provoke oxidation in meat proteins (Estevez et al., 2008) leading to loss of essential aminoacids and decrease of the protein digestibility which influences the nutritional value of meat. The oxidative processes do not stop during storage and often meat is subjected to treatment with various antioxidants, preferably natural, to reduce the intensity and prevent the damage of oxidation. Recently more studies focus the attention on developing feeding strategies or using natural antioxidants as supplements in the animals diets to increase the antioxidant activity of the muscles and to reduce the oxidative processes post mortem. Vitamin E is one of the most important natural antioxidants, playing a fundamental role in the prevention of oxidation in biological systems such as plasma, membranes and tissues. Vitamin E refers to a group of eight fat-soluble compounds that include both tocopherols and tocotrienols (Brigelius-Flohe and Traber, 1999) of which α-tocopherol was reported to have the highest biological activity (Hewavitharana et al., 2004). Antioxidant vitamins are also essential in animal nutrition for maintaining normal health (Fiego et al., 2004) and also supplementation of diet with vitamin E during the growing and finishing periods has been shown to improve quality of meat in pigs (Niculita et al., 2007).

The aim of the study was to examine the effect of vitamin E in the diet of pigs on the oxidative stability of meat lipids and proteins during storage.

**Material and methods**

**Animals and diets**

The experiment was carried out on the experimental farm of the Institute of Animal Science, Kostinbrod. A total of 20 ♀ Youna x ♂ Pietrain pigs were allocated in 2 groups (control and experimental) of 6 females and 4 castrated males per group. The pigs from both groups were fattened using concentrate with a composition presented in Table 1, as the diet of the experimental group was additionally supplemented with vitamin E in amounts 400 mg/kg feed for a period of 10 weeks. The animals had ad libitum access to feed during storage of 48 h, 6 days and 90 days. The oxidation of proteins was determined by measuring the content of carbonyls formed during the same storage periods.

<table>
<thead>
<tr>
<th>Component</th>
<th>Content, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maize</td>
<td>18</td>
</tr>
<tr>
<td>Barley</td>
<td>20</td>
</tr>
<tr>
<td>Wheat</td>
<td>25</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>22</td>
</tr>
<tr>
<td>Sunflower meal</td>
<td>12.9</td>
</tr>
<tr>
<td>Vitamin-mineral premix</td>
<td>0.20</td>
</tr>
<tr>
<td>Lime</td>
<td>1.48</td>
</tr>
<tr>
<td>Salt</td>
<td>0.30</td>
</tr>
<tr>
<td>Lysine</td>
<td>0.10</td>
</tr>
<tr>
<td>Vanilla</td>
<td>0.02</td>
</tr>
<tr>
<td>Nutritive value</td>
<td></td>
</tr>
<tr>
<td>Crude protein</td>
<td>14.27</td>
</tr>
<tr>
<td>Fat</td>
<td>3.96</td>
</tr>
<tr>
<td>Ash</td>
<td>7.74</td>
</tr>
<tr>
<td>Non-nitrogen extractive compounds</td>
<td>64.33</td>
</tr>
</tbody>
</table>

**Table 1. Diet composition**

* e-mail: tlpopova@yahoo.com
and water. The average weight of the pigs at the beginning of the experiment was 49.1±5.71 kg and reached 94.75±5.99 kg and 94.8±4.54 kg at the end of the trial, respectively for the control and the experimental group.

Slaughtering and sampling
The pigs were slaughtered in a standard slaughterhouse. The carcasses were kept for 24 h at 4°C and after that Musculus Longissimus dorsi (MLD) and Musculus Semimembranosus (MSM) were carefully dissected of each left half of the carcasses. Samples for lipid and protein oxidation measurement were taken from both muscles, wrapped in permeable foil and stored at 4°C for 6 days after which the storage continued at – 20°C for up to 90 days. The oxidation of lipids was analysed by the method of Lynch and Frei (1993), modified by Mercier et al. (1998), measuring the amount of the 2-thiobarbituric acid reactive substances (TBARS) formed during storage for 48h, 6 days and 90 days. The analysis of protein oxidation was done by measurements of the content of carbonyls as nmol DNPH/ mg protein (Olivier et al., 1987) with slight modifications during the same intervals of storage.

Statistical analysis
Data were analysed by two-way analysis of variance by the ANOVA procedure using a model including the main effects of vitamin E supplementation, muscle type and their interaction. The dynamic formation of TBARS and carbonyls in the muscles during storage was analysed by Fisher's LSD method. Statistical evaluation was done by JMP v.7 software package.

Results and discussion

Lipid oxidation
The results for the TBARS formation during the refrigerated storage of the samples showed significant effect of the vitamin E supplementation (Table 2). The values of the TBARS remained lower in the group that received vitamin E after 48 h and 6 days (P<0.01). Significant influence of vitamin E was also observed after frozen storage (P<0.01) and lower levels of lipid oxidation were displayed in the muscles of the pigs from the supplemented group. TBARS formation was affected also by the muscle type, as the values that were determined after refrigerated and frozen storage were lower in MSM (P<0.05).

As a whole the values of TBARS in the muscles of the animals that received vitamin E remained lower compared to the control group. The overall inhibitory effect of vitamin E supplementation of the pig diet on the lipid oxidative processes in meat has been reported in several studies (Buckley et al., 1995; Morrissey et al., 1998; Sahoo and Verma, 1999), as the influence of vitamin E varied between the storage conditions and the amount of vitamin E added.

According to Jensen et al. (1998), when stored at 4°C up to 9 days, meat products obtained from pigs fed 100 – 300 mg all-rac-α-tocopherol/kg feed displayed 22 – 94% lower lipid oxidation. For the frozen meat, stored 13 – 16 weeks, the same authors observed 42-67% increase of lipid oxidatitative stability.

Corino et al. (1999) and Niculita et al. (2007) demonstrated that vitamin E supplementation in the diet of pigs in amounts 100 and 300 mg/ kg diet reduced TBARS values in pork, while Hwang et al. (2012) observed that at amount of 210 IU/ kg the vitamin also decreased TBARS values in pork. On the other hand, Hasty et al. (2002), Lahuky et al. (2005), Bahelka et al. (2011) did not observe significant influence of the vitamin E supplementation on TBARS values in fresh pork loin (24 h of storage), suggesting that it better displays its inhibitory effect on oxidation of meat lipids during longer storage.

Influence of vitamin E supplementation and muscle on the TBARS formation in pigs (mg MDA/kg meat)

<table>
<thead>
<tr>
<th>Time of storage</th>
<th>Group</th>
<th>Muscle</th>
<th>S.E.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Vitamin E</td>
<td>MLD</td>
<td>MSM</td>
</tr>
<tr>
<td>48 h</td>
<td>0.35</td>
<td>0.21</td>
<td>0.30</td>
<td>0.26</td>
</tr>
<tr>
<td>6 days</td>
<td>0.54</td>
<td>0.35</td>
<td>0.52</td>
<td>0.37</td>
</tr>
<tr>
<td>90 days</td>
<td>0.77</td>
<td>0.41</td>
<td>0.69</td>
<td>0.48</td>
</tr>
</tbody>
</table>

The values are presented as least squares means, *P<0.05, ** P<0.01.
During the refrigerated storage the carbonyl content remained lower (Figure 1a) in both groups showed constant increase in the lipid oxidation throughout the storage. Significant differences in the formation depends on the muscle type, storage duration and control group, while in the experimental, significant differences existed between all the measurement intervals. The TBARS contents reached their maximum after 90 day of storage. The same trend of increase was also observed in the carbonylation of the muscle (Figure 1b). In both groups the significant differences in the carbonyl content were found between 48 h and 6 days, as well as 48 h and 90 day (P<0.05).

Similar to MLD, the TBARS and carbonyl levels showed

Table 3. Influence of vitamin E supplementation and muscle on the carbonyl formation in pigs (nmol/mg protein)

<table>
<thead>
<tr>
<th>Time of storage</th>
<th>Group</th>
<th>Muscle</th>
<th>S.E.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>control</td>
<td>Vitamin E</td>
<td>MLD</td>
<td>MSM</td>
</tr>
<tr>
<td>48 h</td>
<td>4.25</td>
<td>3.95</td>
<td>4.78</td>
<td>3.40</td>
</tr>
<tr>
<td>6 days</td>
<td>7.04</td>
<td>6.44</td>
<td>8.37</td>
<td>5.11</td>
</tr>
<tr>
<td>90 days</td>
<td>10.56</td>
<td>10.15</td>
<td>9.00</td>
<td>11.71</td>
</tr>
</tbody>
</table>

The values are presented as least squares means, *P<0.05, ** P<0.01.

Dynamics of changes in the TBARS and carbonyl formation

The dynamics of changes in the TBARS formation in MLD (Figure 1a) in both groups showed constant increase in the lipid oxidation throughout the storage. Significant differences in the TBARS content were found between 48 h and 90 day (P<0.05) in the control group, while in the experimental, significant differences existed between all the measurement intervals. The TBARS contents reached their maximum after 90 day of storage. The same trend of increase was also observed in the carbonylation of the muscle (Figure 1b). In both groups the significant differences in the carbonyl content were found between 48 h and 6 days, as well as 48 h and 90 day (P<0.05).

Figure 1. Dynamics of change in TBARS (a) and carbonyls (b) content in MLD
*Intervals within group connected with different letters are statistically different (P<0.05)

Similar to MLD, the TBARS and carbonyl levels showed

Figure 2. Dynamics of change in TBARS (a) and carbonyls (b) content in MSM
*Intervals within group connected with different letters are statistically different (P<0.05)
increase during the storage in MSM (Figure 2a and b) in both control and vitamin E supplemented group. Significant difference in the TBARS levels in the control group was found between 48 h and 90" day, while in the experimental group the content of the TBARS differed between 48 h and 6 days, and 48 h and 90 days of storage (P<0.05). In both groups carbonyl content was significantly different between 48 h and 90 day, as well as 6 days and 90 days (P<0.05).

The same trends in the development of the TBARS and carbonyls show that the lipid and protein oxidative processes are possibly coupled. The coupling between lipid oxidation and protein oxidation has been studied to some extent in meat model systems and meat (Lund, 2007). In beef (Lund et al., 2007) and pork (Lund et al., 2008), during chill storage in high-oxygen atmosphere formation of secondary lipid oxidation products was reported to start at earlier storage times than the formation of protein oxidation products. Similar results were observed by Mercier et al. (1998) in turkey meat stored in atmospheric air with an oxygen-permeable film for 9 days. The authors found no significant increase in carbonyl content during the storage period, while a significant increase in TBARS was observed. These observations indicate that lipid oxidation might precede protein oxidation in meat when lipid oxidation is evaluated by TBARS and protein oxidation by the formation of carbonyls. It could be noted that the content of carbonyls formed during storage in MLD are higher in the vitamin E supplemented group, indicating certain prooxidative effect of vitamin E. It was found that the antioxidative effect of vitamin E supplementation might depend on the muscle type. Thus Mercier et al. (1998) found muscle dependent effect of vitamin E in turkeys. The authors reported that after storage, vitamin E decreased the carbonyl content in Musculus Sartorius but no effect was found in Musculus Pectoralis.

**Conclusion**

As a whole vitamin E supplementation in the diet led to significantly lower TBARS formation in the muscles after refrigerated and frozen storage. Throughout the storage periods, MLD displayed higher susceptibility to lipid oxidation. Vitamin E supplementation did not influence the oxidation of proteins to such an extent as the lipid oxidation. Carbonyl formation differed between muscles on all the storage periods. During the refrigerated storage, MLD displayed higher extent of carbonylation, whereas after frozen storage carbonyl content was increased in MSM. In both muscles lipid and protein oxidation showed the same trends of development in the control and experimental group, reaching maximum after 90" day of frozen storage.

**Acknowledgements**

This material is based upon work supported by Project BG051PO001-3.3.06-0033 "Support for the development of doctoral graduate students and young researchers" OP "Human resources development", Ministry of Labour and Social Policy, European Social Fund (ESF).

**References**


Review

Effect of cubicle technological parameters on welfare and comfort of dairy cows
D. Dimov

Genetics and Breeding

Identification of promising genotypes for hybridization in spring barley
B. Dyulgerova, D. Dimova, N. Dyulgerov

Comparative evaluation of triticale cultivars grown in the region of Dobrudzha
A. Ivanova, N. Tsenov

Stress tolerance to drought of inbred maize lines
P. Vulchinkova

Sericin content in raw silk from Bombyx mori L. cocoons with different fluorescent characteristics
M. Panayotov

Genetic variability in two-rowed spring barley
B. Dyulgerova, N. Dyulgerov, M. Dimitrova-Doneva

Testing of new Bulgarian sunflower hybrids under the conditions of Northeast Bulgaria
II. Phenological specificity
G. Georgiev, P. Peevska, E. Penchev

Maize hybrids testing in system “Plus”
S. Vulchinkov, V. Valkova, D. Ilchovska, P. Vulchinkova

Effect of gamma-irradiation on the fatty acid composition and susceptibility to powdery mildew (Erysiphe cruciferarum) of oilseed rape plants
M. Petkova, M. Dimova, D. Dimova, S. Bistrichanov

Nutrition and Physiology

Investigations on haematological parameters and bone marrow morphology in broiler chickens with experimental aflatoxicosis
I. Valchev, D. Kanakov, Ts. Hristov, L. Lazarov, N. Grozeva, Y. Nikolov

Light microscopy of the adipose tissue distribution along the coronary branches in the myocard of the New Zealand White rabbit
D. Vladova, D. Yovchev, R. Dimitrov, M. Stefanov, P. Hristov

Clinical and haematological studies on subclinical lactational ketosis in dairy goats
R. Binev, V. Marutsova, V. Radev

Effect of linseed and sunflower oils in the diet on the growth parameters in rainbow trout (Oncorhynchus mykiss W.) cultivated in a recirculating system
G. Zheliazkov
Production Systems

Intensity of spike increase in *Triticum aestivum* L. cultivars depending on some agronomy factors 437
M. Nankova, A. Ivanova, N. Tsenov

Effect of the complex suspension foliar fertilizers Lactofol and Amalgerol premium on grain yield from soybean (*Glycine max* (L.) Merr.) under the conditions of Dobrudzha region 445
G. Milev, R. Todorova

Seasonal dynamics of virus pathogens important for *Foeniculum vulgare* 451
B. Dikova, H. Lambev

Fertility and hatchability of Japanese quail eggs under semi arid conditions in Nigeria 455
A. Raji, S. Mbap, G. Mohammed, I. Kwari

Agriculture and Environment

Algae cenoses with dominate *Homoeothrix varians* Geitler and *Homoeothrix crustaceae* Woronichin in the Veleka River, Bulgaria 460
K. Velichkova, I. Kiryakov

Relationship between soil salinity and *Bassia hirsuta*, *Salicornia europaea* agg. and *Petrosimonia brachyata* distribution on the territory of Pomorie lake and Atanasovsko lake 465
M. Todorova, N. Grozeva, L. Pleskuza, Z. Yaneva, M. Gerdgikova

Environmental friendly methods of inducing resistance against *Cucumber mosaic virus* in pepper 471
N. Petrov

Product Quality and Safety

Meat quality traits in Japanese quails with regard to storage conditions and duration 475
P. Vasilieva, H. Lukanc, A. Genchev

Analytical features of an optimized method for HPLC analysis of some polyphenolic acids and flavonoids in tomato fruits 480
D. Georgieva, I. Tringovska, A. Atanasova, V. Kmetov

Effect of vitamin E in the diet of pigs on the lipid and protein oxidative stability of meat during storage 486
T. Popova, P. Marinova, M. Ignatova

Variability of individual coagulation ability and qualitative composition of milk from Kalofer Longhaired goats 491
G. Kalaydzhiiev, A. Vuchkov, T. Angelova, D. Yordanova, V. Karbashev, J. Krastanov, D. Dimov, N. Oblakov, S. Laleva, Y. Popova

Short Communication

Influence of the distillation rate on the quality and quantity of essential oil from *Rosa alba* L. 494
A. Dobreva
Preparation of papers

Papers shall be submitted at the editorial office typed on standard typing pages (A4, 30 lines per page, 62 characters per line). The editors recommend up to 15 pages for full research paper (including abstract references, tables, figures and other appendices).

The manuscript should be structured as follows: Title, Names of authors and affiliation address, Abstract, List of keywords, Introduction, Material and methods, Results, Discussion, Conclusion, Acknowledgements (if any), References, Tables, Figures.

The title needs to be as concise and informative about the nature of research. It should be written with small letter/bold, 14/without any abbreviations.

Names and affiliation of authors

The names of the authors should be presented from the initials of first names followed by the family names. The complete address and name of the institution should be stated next. The affiliation of authors are designated by different signs. For the author who is going to be corresponding by the editorial board and readers, an E-mail address and telephone number should be presented as footnote on the first page. Corresponding author is indicated with *.

Abstract should be not more than 350 words. It should be clearly stated what new findings have been made in the course of research. Abbreviations and references to authors are inadmissible in the summary. It should be understandable without having read the paper and should be in one paragraph.

Keywords: Up to maximum of 5 keywords should be selected not repeating the title but giving the essence of study.

The introduction must answer the following questions: What is known and what is new on the studied issue? What necessitated the research problem, described in the paper? What is your hypothesis and goal?

Material and methods: The objects of research, organization of experiments, chemical analyses, statistical and other methods and conditions applied for the experiments should be described in detail. A criterion of sufficient information is to be possible for others to repeat the experiment in order to verify results.

Results are presented in understandable tables and figures, accompanied by the statistical parameters needed for the evaluation. Data from tables and figures should not be repeated in the text. Tables should be as simple and as few as possible. Each table should have its own explanatory title and to be typed on a separate page. They should be outside the main body of the text and an indication should be given where it should be inserted.

Figures should be sharp with good contrast and rendition. Graphic materials should be preferred. Photographs to be appropriate for printing. Illustrations are supplied in colour as an exception after special agreement with the editorial board and possible payment of extra costs. The figures are to be each in a single file and their location should be given within the text.

Discussion: The objective of this section is to indicate the scientific significance of the study. By comparing the results and conclusions of other scientists the contribution of the study for expanding or modifying existing knowledge is pointed out clearly and convincingly to the reader.

Conclusion: The most important consequences for the science and practice resulting from the conducted research should be summarized in a few sentences. The conclusions shouldn't be numbered and no new paragraphs be used. Contributions are the core of conclusions.

References: In the text, references should be cited as follows: single author: Sandberg (2002); two authors: Andersson and Georges (2004); more than two authors: Andersson et al. (2003). When several references are cited simultaneously, they should be ranked by chronological order e.g.: (Sandberg, 2002; Andersson et al., 2003; Andersson and Georges, 2004). References are arranged alphabetically by the name of the first author. If an author is cited more than once, first his individual publications are given ranked by year, then come publications with one co-author, two co-authors, etc. The names of authors, article and journal titles in the Cyrillic or alphabet different from Latin, should be transliterated into Latin and article titles should be translated into English. The original language of articles and books translated into English is indicated in parenthesis after the bibliographic reference (Bulgarian = Bg, Russian = Ru, Serbian = Sr, if in the Cyrillic, Mongolian = Mo, Greek = Gr, Georgian = Geor., Japanese = Ja, Chinese = Ch, Arabic = Ar, etc.)

The following order in the reference list is recommended:

Journal articles: Author(s) surname and initials, year. Title. Full title of the journal, volume, pages. Example:


Books: Author(s) surname and initials, year. Title. Edition, name of publisher, place of publication. Example:


Book chapter or conference proceedings: Author(s) surname and initials, year. Title. In: Title of the book or of the proceedings followed by the editor(s), volume, pages. Name of publisher, place of publication. Example:


The Editorial Board of the Journal is not responsible for incorrect quotes of reference sources and the relevant violations of copyrights.

Animal welfare

Studies performed on experimental animals should be carried out according to internationally recognized guidelines for animal welfare. That should be clearly described in the respective section “Material and methods”.

Instruction for authors

Preparation of papers

Papers shall be submitted at the editorial office typed on standard typing pages (A4, 30 lines per page, 62 characters per line). The editors recommend up to 15 pages for full research paper (including abstract references, tables, figures and other appendices).

The manuscript should be structured as follows: Title, Names of authors and affiliation address, Abstract, List of keywords, Introduction, Material and methods, Results, Discussion, Conclusion, Acknowledgements (if any), References, Tables, Figures.

The title needs to be as concise and informative about the nature of research. It should be written with small letter/bold, 14/without any abbreviations.

Names and affiliation of authors

The names of the authors should be presented from the initials of first names followed by the family names. The complete address and name of the institution should be stated next. The affiliation of authors are designated by different signs. For the author who is going to be corresponding by the editorial board and readers, an E-mail address and telephone number should be presented as footnote on the first page. Corresponding author is indicated with *.

Abstract should be not more than 350 words. It should be clearly stated what new findings have been made in the course of research. Abbreviations and references to authors are inadmissible in the summary. It should be understandable without having read the paper and should be in one paragraph.

Keywords: Up to maximum of 5 keywords should be selected not repeating the title but giving the essence of study.

The introduction must answer the following questions: What is known and what is new on the studied issue? What necessitated the research problem, described in the paper? What is your hypothesis and goal?

Material and methods: The objects of research, organization of experiments, chemical analyses, statistical and other methods and conditions applied for the experiments should be described in detail. A criterion of sufficient information is to be possible for others to repeat the experiment in order to verify results.

Results are presented in understandable tables and figures, accompanied by the statistical parameters needed for the evaluation. Data from tables and figures should not be repeated in the text. Tables should be as simple and as few as possible. Each table should have its own explanatory title and to be typed on a separate page. They should be outside the main body of the text and an indication should be given where it should be inserted.

Figures should be sharp with good contrast and rendition. Graphic materials should be preferred. Photographs to be appropriate for printing. Illustrations are supplied in colour as an exception after special agreement with the editorial board and possible payment of extra costs. The figures are to be each in a single file and their location should be given within the text.

Discussion: The objective of this section is to indicate the scientific significance of the study. By comparing the results and conclusions of other scientists the contribution of the study for expanding or modifying existing knowledge is pointed out clearly and convincingly to the reader.

Conclusion: The most important consequences for the science and practice resulting from the conducted research should be summarized in a few sentences. The conclusions shouldn’t be numbered and no new paragraphs be used. Contributions are the core of conclusions.

References: In the text, references should be cited as follows: single author: Sandberg (2002); two authors: Andersson and Georges (2004); more than two authors: Andersson et al. (2003). When several references are cited simultaneously, they should be ranked by chronological order e.g.: (Sandberg, 2002; Andersson et al., 2003; Andersson and Georges, 2004). References are arranged alphabetically by the name of the first author. If an author is cited more than once, first his individual publications are given ranked by year, then come publications with one co-author, two co-authors, etc. The names of authors, article and journal titles in the Cyrillic or alphabet different from Latin, should be transliterated into Latin and article titles should be translated into English. The original language of articles and books translated into English is indicated in parenthesis after the bibliographic reference (Bulgarian = Bg, Russian = Ru, Serbian = Sr, if in the Cyrillic, Mongolian = Mo, Greek = Gr, Georgian = Geor., Japanese = Ja, Chinese = Ch, Arabic = Ar, etc.)

The following order in the reference list is recommended:

Journal articles: Author(s) surname and initials, year. Title. Full title of the journal, volume, pages. Example:


Books: Author(s) surname and initials, year. Title. Edition, name of publisher, place of publication. Example:


Book chapter or conference proceedings: Author(s) surname and initials, year. Title. In: Title of the book or of the proceedings followed by the editor(s), volume, pages. Name of publisher, place of publication. Example:


The Editorial Board of the Journal is not responsible for incorrect quotes of reference sources and the relevant violations of copyrights.

Animal welfare

Studies performed on experimental animals should be carried out according to internationally recognized guidelines for animal welfare. That should be clearly described in the respective section “Material and methods”.