



ISSN 1313 - 8820
Volume 2, Number 2
June 2010

AGRICULTURAL SCIENCE AND TECHNOLOGY

2010

An International Journal Published by Faculty of Agriculture,
Trakia University, Stara Zagora, Bulgaria

Editor-in-Chief

Tsanko Yablanski
Faculty of Agriculture
Trakia University, Stara Zagora
Bulgaria

Co-Editor-in-Chief

Radoslav Slavov
Faculty of Agriculture
Trakia University, Stara Zagora
Bulgaria

Editors and Sections

Genetics and Breeding

Atanas Atanasov (Bulgaria)
Ihsan Soysal (Turkey)
Max Rothschild (USA)
Stoitcho Metodiev (Bulgaria)

Nutrition and Physiology

Nikolai Todorov (Bulgaria)
Peter Surai (UK)
Zervas Georgios (Greece)

Production Systems

Dimitar Pavlov (Bulgaria)
Dimitar Panaiotov (Bulgaria)
Jordan Staikov (Bulgaria)
Yuliana Yarkova (Bulgaria)

Agriculture and Environment

Georgi Petkov (Bulgaria)
Ramesh Kanwar (USA)

Product Quality and Safety

Marin Kabakchiev (Bulgaria)
Stefan Denev (Bulgaria)

English Editor

Yanka Ivanova (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 manuscript written in English should be submitted as MS-Word file attachments via e-mail to ascitech@uni-sz.bg. Manuscripts must be prepared strictly in accordance with the detailed instructions for authors at the website

<http://www.uni-sz.bg/ascitech/index.html> 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 an 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 request from the Technical Assistance or could be downloaded from the website of the journal. 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.

Subscriptions

Agricultural Science and Technology is published four times a year. The subscription price for institutions is 80 € and for personal subscription 30 € which

include electronic access and delivery. Subscription run for full calendar year. Orders, which must be accompanied by payment may be sent direct to the publisher:

Trakia University
Faculty of Agriculture, Bank account:
UniCredit Bulbank,
Sofia BIC: UNCRBGSF

IBAN: BG29UNCR76303100117681
With UniCredit Bulbank Stara Zagora

Internet Access

This journal is included in the Trakia University Journals online Service which can be found at www.uni-sz.bg.

Copyright

All rights reserved. No part of this publications may be translated into other languages, reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying or any information storage and retrieval system without permission in writing from the publisher.

Address of Editorial office:

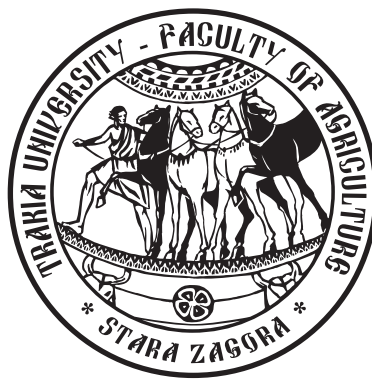
Agricultural Science and Technology
Faculty of Agriculture, Trakia University
Student's campus, 6000 Stara Zagora
Bulgaria
Telephone.: +359 42 699330
+359 42 699446
<http://www.uni-sz.bg/ascitech/index.html>

Technical Assistance:

Nely Tzvetanova
Telephone.: +359 42 699446
E-mail: ascitech@uni-sz.bg

ISSN 1313 - 8820

Volume 2, Number 2
June 2010



AGRICULTURAL SCIENCE AND TECHNOLOGY

2010

An International Journal Published by Faculty of Agriculture,
Trakia University, Stara Zagora, Bulgaria



— PRODUCTION OF FRESH —
SAUSAGES AND OTHER MEAT PRODUCTS



*KEN...
Everywhere
you go*

KEN LTD
6000 Stara Zagora
www.ken.bg
for contacts:
+359 42 64 11 28
60 24 92
fax: +359 42 60 11 46



Genetics and Breeding

Influence of various cryoprotectants on the sperm mobility of Muscovy semen before and after cryopreservation

V. Gerzilov*

Department of Animal Science, Agricultural University, 12 Mendeleev, 4000 Plovdiv, Bulgaria

Abstract. A study for influence of five cryoprotectants - glycerol, dimethyl sulfoxide (DMSO), ethylene glycol, 1,3 propandiol and polyethylene glycol in different concentrations from 3%, 5% and 7% with use of HIA-1 and AU extenders on the mobility of Muscovy spermatozoa after freeze-thaw was carried out. The semen was collected with artificial vagina from 9 one-year-old Muscovy drakes by using a female as a teaser twice a week. The sperm was diluted with HIA-1 and AU extenders with 15% egg yolk (v/v) at the ratio of 1:3 (semen:extender), respectively, and divided equally. Cryoprotectant was added in one of following concentrations 3-5-7% into each semen sample, respectively, a process of equilibration in a refrigerator followed at 4 0 C for 60 min. Afterwards they were directly dropped in concave cavities of dry ice at -790 C for 10 min. The semen pellets were placed in an atmosphere of liquid nitrogen (LN2) vapors for 5 - 10 min and finally they were put in cryotubes and plunged into liquid nitrogen. The pellets were kept frozen in the LN2 container for at least 2 months before being thawed for evaluation. The sperm samples were thawed at 420 C with HIA-1 and AU extenders, respectively. Comparatively the highest sperm mobility was established at using 5% and 7% glycerol and 7% DMSO. Both HIA-1 and AU extenders are suitable to semen dilution. Cryopreservation of Muscovy semen caused damages in the morphological integrity of sperm cells.

Keywords: Muscovy duck, semen, sperm mobility, cryopreservation, cryoprotectant

Abbreviations: LN₂ - liquid nitrogen, spz – spermatozoa, DMSO - dimethyl sulfoxide, ME - metabolizable energy, CP - crude protein

Introduction

The preservation of poultry semen in frozen state has been the subject of intense scientific interest beginning approximately 60 years ago by Polge (1949, 1951) with the discovery of glycerol as a good cryoprotective medium. Despite the fact that this scientific breakthrough was accomplished with rooster semen (Polge, 1951), the overall fertility rates with frozen/thawed poultry semen are highly variable and not reliable enough for use in commercial production or preservation of genetic stocks. According to Long (2006), the greatest progress in commercializing semen preservation has been achieved by the dairy and beef cattle industries, where semen cryopreservation has been optimized, standardized, and automated. This high level of success with bull semen has not been achieved with other livestock species, such as pigs or sheep (Holt, 2000), and the fertility rates of cryopreserved poultry sperm are dramatically lower than any of the domestic mammalian species.

Despite extensive research on poultry semen cryopreservation, there is limited success in applying these procedures either to "on farm" use or to the enhanced management or conservation of rare wild avian species (Blanco et al., 2000). The sperm cryopreservation for *ex situ* management of genetic resources in poultry is still a big problem. According to Blesbois and Brillard (2007), cryobanking for species other than chicken remains extremely limited. The lack of knowledge required to develop the appropriate technology for cryopreservation of sperm in poultry species is another challenge for the years to come. Cryopreservation of Muscovy duck semen is more difficult than Pekin ducks (Tselutin et al., 1995). However, the

experiments indicate that spermatozoa of commercial lines of these two species of ducks are able to be frozen with reasonable success of fertility of frozen-thawed semen (Blesbois, 2007).

One of the most critical steps in the cryopreservation of avian semen is the choice of the cryoprotectant and its use during the process (Tseltin et al., 1999). Many compounds have been tested for their efficiency as sperm cryoprotectants (Holt, 2000), but most extensively – glycerol, dimethyl sulfoxide, dimethyl acetamide, dimethyl formamid, diethyl formamide, ethylene glicol, propylene glicol (Lake and Ravie, 1982; Hammerstedt and Graham, 1992; Surai and Wishart, 1996; Tseltin et al., 1999; Tai et al., 2001; Łukaszewicz, 2001).

The aim of this study is to compare the effect of two HIA-1 and AU extenders, and five cryoprotectants - glycerol, dimethyl sulfoxide, ethylene glycol, 1,3 propandiol and propylene glycol in 3%, 5% and 7% as a final concentration, respectively, on the mobility of Muscovy spermatozoa before and after cryopreservation.

Material and methods

Birds

The experiment was carried out on the Poultry farm of the Agricultural University, Plovdiv with 9 one-year-old Muscovy drakes. During the entire natural breeding season the males were kept individually in cages (0.6/0.8/0.6 m in size) under natural light. The birds were fed with diet consisting of ME - 11.5 MJ/kg and CP – 16.0 %, and daily ration from 200 to 250 g/bird.

* e-mail: vasko@au-plovdiv.bg

Semen collection

The semen was collected individually by placing a female (teaser method) in the cage of the Muscovy drake using an artificial vagina, two times per week (Tan, 1980; Gerzilov, 2000). The artificial vagina consisted of a rubber muff and a graduated test-tube.

Semen evaluation

Only good quality ejaculates (color – pearly-white; cleanliness – free of any contamination with cloacal products; volume – above 0.3 ml; sperm mobility – above 65%, sperm concentration – above 1×10^9 sperm cells/ml) were used for cryopreservation. The pooled

semen was divided in two parts and diluted with HIA-1 and AU-extenders by Gerzilov (2003) with added 15% egg yolk (v/v) at a ratio 1:3 (semen:extender), respectively (Figure 1).

The HIA-1 extender consists of 0.25 g D-glucose, 0.25 g D-fructose, 0.07 g saccharose, 0.50 g sodium citrate, 9.00 g sodium chloride, and 100 mL double distilled water. The osmolarity was 290 mOsmol/kg and pH - 7.00. The AU extender consists of 0.40 g D-glucose, 0.80 g D-fructose, 0.80 g saccharose, 0.90 g sodium citrate, 0.84 g sodium glutamate, 0.40 ml glycolol, 0.04 g ethylenediamine tetra acetic acid disodium salt dihydrate, and 100

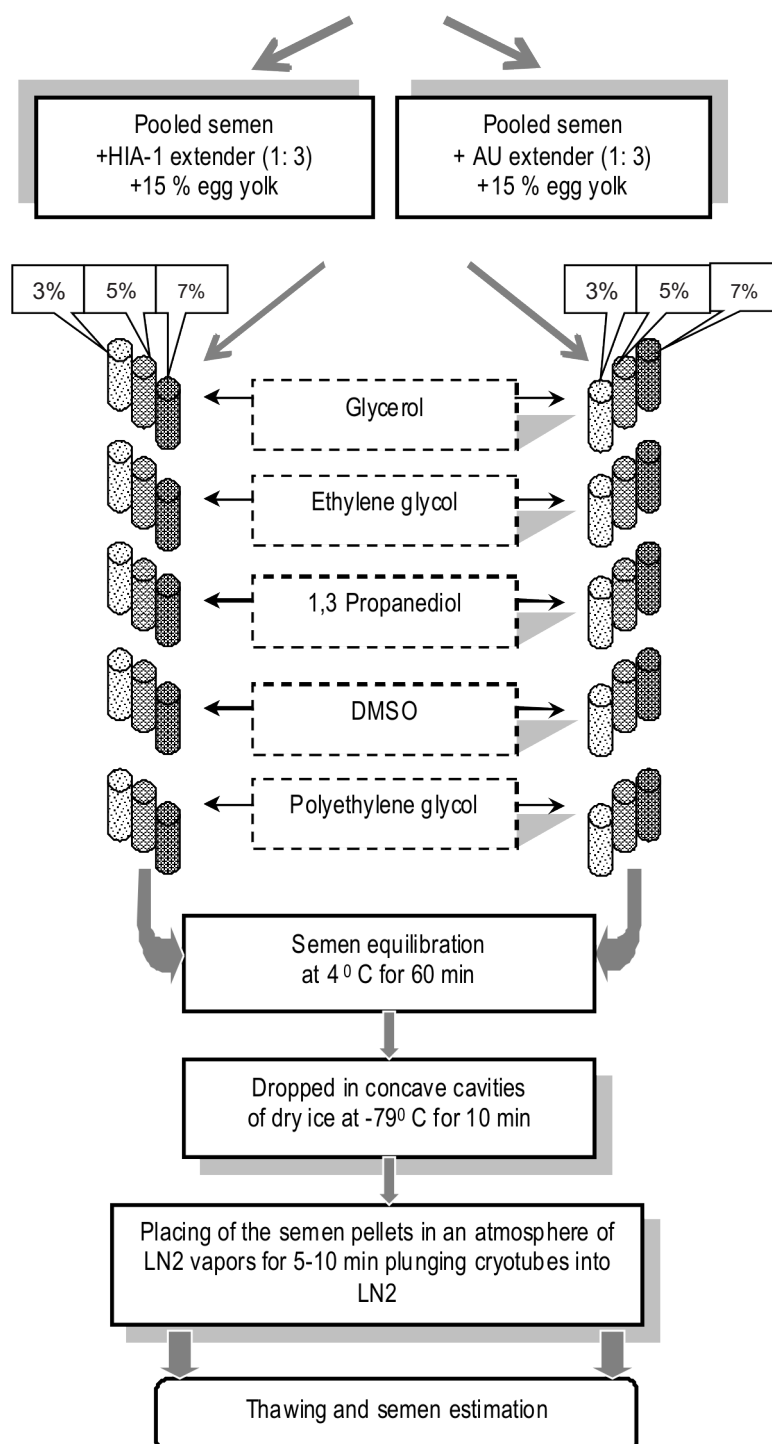


Figure 1. Design of cryopreservation of Muscovy semen

ml double distilled water. The osmolarity was 320 mOsmol/kg and pH - 7.00.

The diluted semen was distributed equally in 15 sterile glass tubes again. A cryoprotectant - glycerol, dimethyl sulfoxide, ethylene glycol, 1,3 propandiol and polyethylene glycol in 3%, 5% and 7% as a final concentration, respectively, was supplementd to each tube as a final concentration. The semen samples were equilibrated in a refrigerator at 4^o C for 60 min, and then they were directly dropped in concave cavities of dry ice at -79^o C for 10 min. Semen pellets were placed in an atmosphere of LN₂ vapors for 5 - 10 min and finally they were put in cryotubes and plunged into liquid nitrogen. The pellets were kept frozen in the LN₂ container for at least 2 months before being thawed for evaluation. The sperm samples were thawed at 42^o C with HIA-1 and AU extenders (1:3 v/v), respectively.

The sperm mobility (%) of the pooled semen, diluted,

equilibrated and frozen/thawed semen was evaluated by visual determination of the spermatozoa with progressive motions using a Nikon Alphaphot-2YS2 microscope (10x40)

Statistical analyses

Data were subjected to one-way analysis of variance (ANOVA) followed by Student t-test to determine the level of significance among mean values. Results are expressed as a mean and standard error.

Results and discussion

Sperm mobility remained high 76.75±1.36 % and 77.50±3.54 % after the dilution with HIA-1 and AU extenders, respectively. There is no effect on the sperm mobility after the addition of

Table 1. Sperm mobility in semen before freezing and after thawing

Cryoprotectant	%	HIA-1 extender			AU extender		
		Diluted semen x ± S _x	Equilibrated semen x ± S _x	Frozen/thawed semen x ± S _x	Diluted semen x ± S _x	Equilibrated semen x ± S _x	Frozen/thawed semen x ± S _x
Glycerol	3	74.50±2.43	71.25±2.01	6.67±7.36	76.00±1.41	68.50±2.12	17.00±2.12
	5	76.67±0.65	72.67±4.08	35.33±5.31	78.00±1.22	73.33±2.48	31.33±3.63
	7	74.50±2.43	70.75±1.06	29.25±4.17	73.33±2.04	70.67±1.47	25.33±6.72
Ethylene glycol	3	70.50±5.51	71.67±4.08	11.67±4.08	72.50±3.53	70.50±3.54	8.00±2.82
	5	74.75±1.91	70.75±0.87	19.00±8.16	76.00±1.41	70.00±2.12	9.00±1.41
	7	76.25±2.76	72.00±1.41	18.25±8.00	78.00±2.83	71.67±5.40	17.33±1.78
1.3 Propandiol	3	70.50±5.51	61.00±8.49	6.50±4.95	76.00±1.41	73.50±4.95	6.33±1.08
	5	74.75±1.91	62.50±10.61	15.00±7.07	73.00±1.54	72.50±3.54	6.50±2.12
	7	76.25±2.76	65.00±6.12	10.00±7.01	76.25±2.76	68.50±2.12	8.00±2.83
DMSO	3	72.67±1.78	71.67±2.04	isolated spz	73.67±1.00	66.00±1.41	11.50±7.78
	5	71.67±5.40	68.33±2.04	22.00±13.91	75.00±7.01	67.50±3.54	21.00±5.66
	7	71.75±5.13	68.25±3.28	25.00±7.71	72.50±3.54	65.00±7.07	23.50±2.12
Polyethylene glycol	3	71.50±2.12	70.50±0.71	isolated spz	73.67±0.82	69.67±1.78	isolated spz
	5	71.00±1.23	68.50±2.12	isolated spz	71.50±2.12	71.50±2.12	isolated spz
	7	66.50±4.95	64.00±1.41	7.00±4.24	71.00±1.23	69.00±4.14	4.50±0.71

cryoprotectants in different concentration to the diluted semen (Table 1.) After the use of HIA-1 extender, sperm mobility was in the range of 66.50±4.95% to 76.67±0.65% and 71.00±1.23% to 78.50±2.12% in the use of AU extender. In both cases the lowest values in sperm mobility were in 7% concentration of polyethylene glycol. The highest values of sperm mobility were obtained in 5% concentration of glycerol. Sperm mobility after the equilibration was decreased but the differences were not significant.

The mobility in the thawed semen ranged from several isolated sperm cells using polyethylene glycol to 35.33 ± 5.31% using glycerol, respectively. The thawed semen had the highest mobility using 5% and 7% glycerol and 7% DMSO in comparison with all other cryoprotectants. The cryoprotentants - 1,3 propandiol and polyethylene glycol were extremely unsuitable and toxic. The

cryopreservation process induced a significant decrease of sperm mobility vs. diluted and equilibrated semen (P<0.001).

In our previous works we established high percentage of dead and abnormal spermatozoa in unfrozen semen (Gerzilov et al. 2009; Kazachka et al., 2009). The morphological damages of sperm cells affected the membrane integrity, midpiece and mitochondrial helix mainly. Xia et al. (1988) established similar changes in rooster sperm. According to Maeda et al. (1984), the most radical change in Muscovy sperm was the complete separation of acrosome from the apical part of the nucleus. In general, avian spermatozoa are more sensitive to the freezing/thawing process and fertility rates of cryopreserved poultry sperm are dramatically lower than any of the domestic mammalian species (Donoghue and Wishart, 2000; Long, 2006).

Conclusion

Comparatively highest sperm mobility was established at using 5% and 7% concentration of glycerol and 7% concentration of DMSO. Both HIA-1 and AU extenders are suitable as semen diluents. Cryopreservation of Muscovy semen caused damages in the morphological integrity of sperm cells.

References

- Blanco JM, Gee G, Wildt DE and Donoghue AM**, 2000. Species Variation in Osmotic, Cryoprotectant, and Cooling Rate Tolerance in Poultry, Eagle, and Peregrine Falcon Spermatozoa. *Biology of Reproduction*, 63, 1164–1171.
- Blesbois E and Brillard JP**, 2007. Specific features of in vivo and in vitro sperm storage in birds. *Animal*, 1, 10, 1472–1481.
- Blesbois E**, 2007. Current status in avian semen cryopreservation. *World's Poultry Science Journal*, 63, 213–222.
- Chalah T, Seigneurin F, Blesbois E and Brillard JP**, 1999. In vitro comparison of fowl sperm viability in ejaculates frozen by three different techniques and relationship with subsequent fertility in vivo. *Cryobiology*, 39, 185–191.
- Donoghue AM and Wishart GJ**, 2000. Storage of poultry semen. *Animal Reproduction Science*, 62, 213–232.
- Gerzilov V, Kazachka D, Jeleva S, Petrov P, Sabev M and Nikolov I**, 2009. Effect of cryoprotectants on the mobility and the morphology integrity of muscovy spermatozoa. *Proceedings XXI International Poultry Symposium PB WPSA "Science for poultry practice – poultry practice for science"*, 7-9 September 2009, Piechowice, Poland.
- Gerzilov V**, 2000. A method for obtaining of semen from the species Muscovy duck (*Cairina moschata*). *Animal Science*, 37, 4, 56–63, (Bg).
- Gerzilov V**, 2003. A study of sperm production of Muscovy ducks (*Cairina moschata*) and the possibility for receiving of Mule ducks by artificial insemination of Pekin ducks (*Anas platyrhynchos*). PhD Thesis, p. 150, (Bg).
- Hammerstedt RH and Graham JK**, 1992. Cryopreservation of poultry sperm: The enigma of glycerol. *Cryobiology*, 29, 26–28.
- Holt WV**, 2000. Basic aspects of frozen storage of semen. *Animal Reproduction Science*, 62, 3–22.
- Kazachka D, Jeleva S, Sabev M, Nikolov I, Stefanov R, Gerzilov V and Miteva K**, 2009. Ultrastructural studies of spermatozoa from the species *Cairina moschata* during cryoconservation. *Comptes rendus de l'Academie bulgare des Sciences*, 62, 3, 361–364.
- Lake PE and Ravie O**, 1982. Dimethylacetamide as a cryoprotectant for fowl spermatozoa. *Poultry Science*, 61, 1497–1498.
- Long JA**, 2006. Avian semen cryopreservation: What are the biological challenges. *Poultry Science*, 85, 232–236.
- Łukaszewicz E**, 2001. DMF effects on frozen gander semen. *British Poultry Science*, 42, 308–314.
- Maeda T, Terada T and Tsutsumi Y**, 1984. Morphological observations on frozen and thawed muscovy spermatozoa. *British Poultry Science*, 25, 3, 409–13.
- Polge C**, 1951. Functional survival of fowl spermatozoa after freezing at -70° C. *Nature*, 167, 949–950.
- Polge C, Smith A and Parkes A.**, 1949. Revival of spermatozoa after vitrification and dehydration at low temperatures. *Nature*, 164, 666.
- Schramm GP and Hubner R**, 1988. Effects of different cryoprotectives and deep freeze techniques on the reproductive potential of turkey sperm after prolonged storage. *Veterinary Medicine*, 43, 426–427.
- Surai PF and Wishart GJ**, 1996. Poultry artificial insemination technology in the countries of the former USSR. *World's Poultry Science Journal*, 52, 227–243.
- Tai JJJ, Chen, JC, Wu KC, Wang SD and Tai C**, 2001. Cryopreservation of gander semen. *British Poultry Science*, 42, 384–388.
- Tan NS**, 1980. The training of drakes for semen collection. *Annali Zootechnica*, 29, 2, 93–102.
- Tselutin K, Narubinal L, Mavrodina T and Tur B**, 1995. Cryopreservation of poultry semen. *British Poultry Science*, 36, 805–811.
- Tselutin K, Seigneurin F and Blesbois E**, 1999. Comparison of cryoprotectants and methods of cryopreservation of fowl spermatozoa. *Poultry Science*, 78, 586–590.
- Xia LJ, Lalli MF, Ansah GA and Buckland RB**, 1988. Ultrastructure of fresh and frozen-thawed spermatozoa of high and low fertility lines of chickens. *Poultry Science*, 67, 819–825.

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:

Simm G, Lewis RM, Grundy B and Dingwall WS, 2002. Responses to selection for lean growth in sheep. *Animal Science*, 74, 39-50

Books: Author(s) surname and initials, year. Title. Edition, name of publisher, place of publication. Example: **Oldenbroek JK**, 1999. Genebanks and the conservation of farm animal genetic resources, Second edition. DLO Institute for Animal Science and Health, Netherlands.

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:

Mauff G, Pulverer G, Operkuch W, Hummel K and Hidden C, 1995. C3-variants and diverse phenotypes of unconverted and converted C3. In: Provides of the Biological Fluids (ed. H. Peters), vol. 22, 143-165, Pergamon Press. Oxford, UK.

Todorov N and Mitev J, 1995. Effect of level of feeding during dry period, and body condition score on reproductive performance in dairy cows, IXth International Conference on Production Diseases in Farm Animals, Sept. 11 – 14, Berlin, Germany, p. 302 (Abstr.).

Thesis:

Penkov D, 2008. Estimation of metabolic energy and true digestibility of amino acids of some feeds in experiments with muscus duck (*Carina moshata*, L). Thesis for DSc. Agrarian University, Plovdiv, 314 pp.

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



CONTENTS

Genetics and Breeding

- Influence of various cryoprotectants on the sperm mobility of Muscovy semen before and after cryopreservation** 57
V. Gerzilov

- Adaptive possibility and yield stability of varieties of oil-bearing roses** 61
N. Kovatcheva, K. Rusanov, I. Atanasov

Nutrition and Physiology

- Shelter policies in the management of canine aggression** 64
A. Arnaudova, I. Varlyakov

Production Systems

- Daily dynamics of the essential oils of *Rosa damascena* Mill. and *Rosa alba* L.** 71
A. Dobрева, N. Kovacheva

- Effectiveness of the insecticide "Mido 20 SL" in the fight with the green rose aphid populations (*Macrosiphum Rosae* L)** 75
H. Lambev

- Potassium fertilization on cotton** 78
G. Panayotova, N. Valkova

Agriculture and Environment

- Microbial communities in areas affected by formation of calcrete in the Thracian plane** 84
S. Bratkova, K. Nikolova, K. Gesheva

- An approach for Fusarium infected corn kernels recognition using linear discrete models** 90
P. Daskalov, V. Mancheva, Ts. Draganova, R. Tsonev

- Mechanism-based category formation of aldehydes for acute aquatic toxicity and mutagenicity** 96
Y. Koleva

Quality and Safety

- Gas-chromatography and organoleptic analysis of the essential oil of *Agastache foeniculum* (Pursh.) Kuntze** 102
G. Zhekova, A. Dzhurmanski, A. Dobрева

- Comparative studies on the fatty acid composition of White brined cheese, marketed in the town of Stara Zagora** 105
N. Naydenova, K. Davidova, T. Iliev, G. Mihaylova