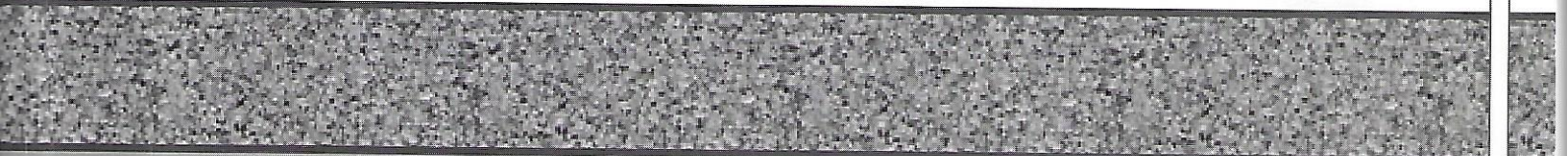




Biological Food Safety & Quality

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Morphological and morphometric characteristics of testicles of immunocastrated pigs

G. Ušćebrka¹, M. Baltić², S. Stojanović¹, J. Aleksić²

¹University of Novi Sad, Faculty of Agriculture, Department of Veterinary Medicine, Novi Sad, Serbia;

²University of Belgrade, Faculty of Veterinary Medicine, Belgrade, Serbia (slobodan113@gmail.com)

Introduction

Surgical castration of male pigs is still common practice in order to avoid the unpleasant smell of meat, and also the aggressive behavior of sexually mature male pigs (Fredriksen et al., 2009, Lundström et al., 2009). However, surgical castration has disadvantages in terms of technological quality of meat. Also, surgical castration without anesthesia is flawed from the standpoint of animal welfare. Alternative methods for the prevention of unpleasant odor of boar meat are of primary interest in pork production, as well as the satisfaction of demands of animal welfare. One of them is immunocastration, non-surgical castration by immunization in which are the male pigs immunized against GnRH (gonadotropin-releasing hormone).

This form of castration is used extensively in many countries, including Australia, New Zealand, Brazil, and its use was approved in Switzerland in 2008 and in the EU in 2009 (Skrllep et al., 2010). Immunocastration leads to a reduction in weight of testicles and accessory sex glands, and to physiological changes which are reflected in reduced production of substances (androstenedione and skatole), which are responsible for the odor of boar meat (Brunius, 2011). The aim of this study was to examine the effectiveness of vaccines based on monitoring of the absolute and relative testicular weight, and morphometric and microscopic characteristics of the testicles of control and immunocastrated boars.

Materials and Methods

The experiment was carried out on two groups of 30 pigs. Groups included non-castrated male pigs (C group) and immunocastrated pigs (I group). The first vaccination of pigs was carried out in the eighth week of age (56 days after birth), and the second one five weeks before slaughter (i.e., with 144 days of age). The pigs were slaughtered at 179 days of age. Both groups of pigs originated from one boar (Duroc and Pietrain mixed-breed) and the same line sows (Landrace and Yorkshire mixed-breed). Pigs of both groups were fed and housed in the same way. Immunization of pigs was conducted by the subcutaneous application (behind the ear base) 2x2 ml (2 ml of the first vaccination, 2 ml of the second vaccination) Improvak vaccine (Pfizer Ltd., Formerly CSL Limited, Parkville, Victoria, Australia). Immediately prior to slaughter of pigs, total body weight was determined, and after the slaughter, the total mass of the testicles was determined. Based on these parameters, average body and testicular weight was determined, as well as relative testicular weight.

From each group a tissue sample of testicles were taken. The tissue samples were fixed in a 10% buffered formalin solution, followed by a sequence of dehydration and clearing. The samples were then embedded in paraffin then cut into serial 5 µm thick sections using a microtome. Histological preparations were stained with hematoxylin-eosin. The following parameters were observed in the analysis of histological preparations of the tissue samples: percentage of germinal epithelium and lumen in tubuli seminiferi, as well as percentage of parenchyma and blood vessels in testicular interstitium. For stereological analyses of percentage of the observed parameters, measurements were performed using the M42 testing system which consists of 21 line segments and 42 points in a testing area. Microscopy was performed using a light microscope Leica DMLS with a Leica DC 300 digital camera, and the software package IM 1000 (Leica Imaging Systems Ltd, Cambridge, UK).

The statistical significance of differences obtained in measurements were determined using one-way ANOVA. The statistical significance of differences was expressed as significant at $P < 0.01$. Statistical processing of data was carried out using the software package Statistica for Windows ver. 7.0.

Results

The results indicate that there were no differences in body weight between C group and I group. Absolute and relative testicle weight was higher in the C group. From the morphometric parameters, the presence of the germinal epithelium in the tubuli seminiferi, as well as the presence of blood vessels in the testicular interstitium were higher in group C, while the presence of lumen in tubuli seminiferi as well as percentage of parenchyma in testicular interstitium were higher in I group (Table 1).

Table 1. Body weight, absolute and relative testicular weight and morphometric parameters of testicles of C and I group

	Control group (C)	Immunocastrated group (I)
Body weight (kg)	111.37	109.40
Absolute testicular weight (g)	574.67*	234.83
Relative testicular weight (g / kg)	5.15*	2.14
Percentage of germinal epithelium in tubuli seminiferi (%)	67.37*	36.25
Percentage of lumen in tubuli seminiferi (%)	32.63	63.75*
Percentage of parenchyma in interstitium (%)	91.11	95.66*
Percentage of blood vessels in interstitium (%)	8.89*	4.34

* Means differ significantly ($P < 0.01$)

Histological analysis of the tissue sections points to a clear morphological specificity of C and I group. Testicular microstructure of animals of group C indicated a good level of development of all of its structural components: a well developed interstitium with numerous of Leydig cells with plenty of lipid droplets, well marked system of capillaries and blood vessels; germinal epithelium in tubuli seminiferi developed well and in these tubules all stages of spermatogenesis and spermiogenesis could be observed, so that in lumen numerous spermatozoids were detected (Figure 1).

Morphological structure of testicles of I group resembled the structure of the testicles of sexually immature animals. In the interstitium, a lower number of blood vessels were observed, and Leydig cells had a smaller diameter with a much smaller number of lipid droplets. In some testicular tubules from animals of I group, germinal epithelium was reduced to only two layers of cells, and in some places complete absence of germinal epithelium was detected. Lumina of tubuli seminiferi were much larger and the presence of spermatozoids was not observed in them (Figure 2).

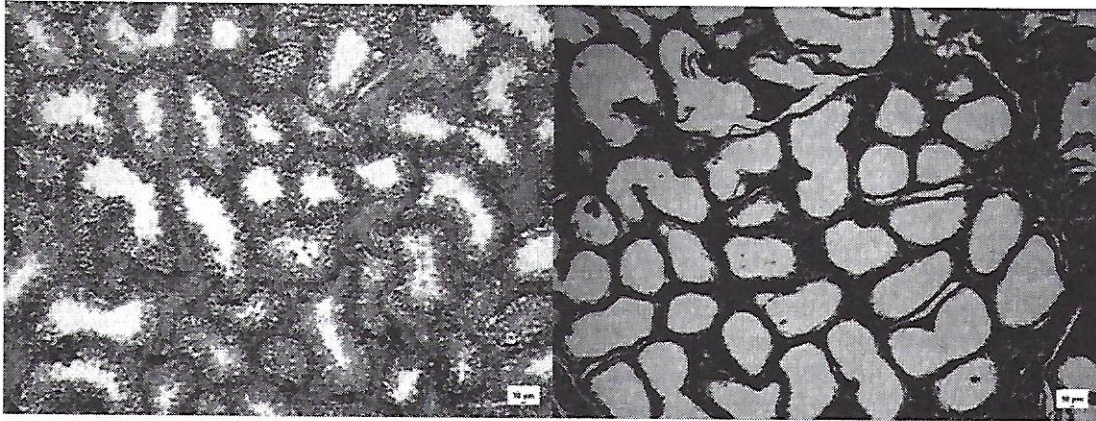


Figure 1. Tubuli seminiferi of C group.
Scale bar = 10 μ m.

Figure 2. Tubuli seminiferi of I group.
Scale bar = 10 μ m.

Conclusions

Based on the obtained parameters, the following conclusions can be derived:

- There were no significant differences in body weight between groups, while the absolute and relative testicular weights were significantly lower in immunocastrated animals
- In immunocastrated animals absence of spermatozoids in lumina of tubuli seminiferi was detected, which indicates cessation of spermiogenesis, and in some tubules, cessation of spermatogenesis.
- Percentage of germinal epithelium in tubuli seminiferi, as well as percentage of blood vessels in interstitium was significantly lower in immunocastrated animals, which indicated lower activity of testicles of immunocastrated animals

Acknowledgement

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