BIOGERONTOLOGY

Carnosine-Induced Changes in the Development of Spermatogenic Epithelial Cells in Senescence Accelerated SAM Mice

A. V. Gopko, A. Yu. Kulibin, M. L. Semenova, and S. T. Zakhidov

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 140, No. 8, pp. 206-209, August, 2005
Original article submitted November 9, 2004

Carnosine significantly increased the number of spermatogonia and Sertoli cells in mice prone (SAMP1) and resistant (SAMR1) to accelerated aging and appreciably reduced cell yield in meiosis and spermiogenesis in SAMP1 mice. In experimental SAMP1 mice catastrophic changes in the number of gametes were paralleled by intensive degradation of the spermatogenic epithelium. In SAMR1 mice treated with carnosine highly ordered spermatogenic structure was preserved.

Key Words: *carnosine; SAMP1 mice; SAMR1 mice; spermatogenic cells*

Persuasive evidence of positive phenotypical effects of carnosine is presented in modern reports on this natural agent [1-3]. In cells carnosine acts as an effective antioxidant, and hence, it can be expected that it will exhibit antimutagenic activity in systems with potentially high genetic instability [6,7].

However, studies of the cytogenetic effects of carnosine on the male gametogenesis in senescence accelerated mice showed a paradoxical phenomenon: carnosine significantly increased the release of cells with chromosome aberrations under experimental conditions [5].

We studied delayed effects of long-term treatment with carnosine on quantitative and morphological characteristics of male sex cells in SAMP1 and SAMR1 mice prone and resistant to rapid aging (maximum life span of SAMP1 mice is 15 months, of SAMR1 mice 24 months).

Embryology Department, Biological Faculty, M. V. Lomonosov Moscow State University. ***Address for correspondence:*** kulibin_a_bkrj@front.ru. A. Yu. Kulibin

MATERIALS AND METHODS

Male SAMP1 and SAMR1 mice were kept under common vivarium conditions with free access to water and food. Experimental animals received carnosine (100 mg/kg) from the age of 2 months daily with drinking water [4]. Controls and experimental animals aged 12 months and more were sacrificed by cervical dislocation, paired testicles were removed, one testicle was fixed (whole) in acetic-glycerol mixture and fragments of the other in Bouin's fixative. Cell of different types were counted in a Goryaev chamber after mechanical destruction of spermatogenic tissue and preparation of cell suspension. Sections (5 μ) for histological analysis were stained with hematoxylin and eosin. Experimental material collected in 1998 was analyzed.

RESULTS

In SAMP1 mice treated with carnosine the counts of spermatogonial and Sertoli cells increased by 3.65 and 2.14 times, respectively, compared to controls, while the counts of pachytene spermatocytes, round spermatides,

0007-4888/05/1402-0235

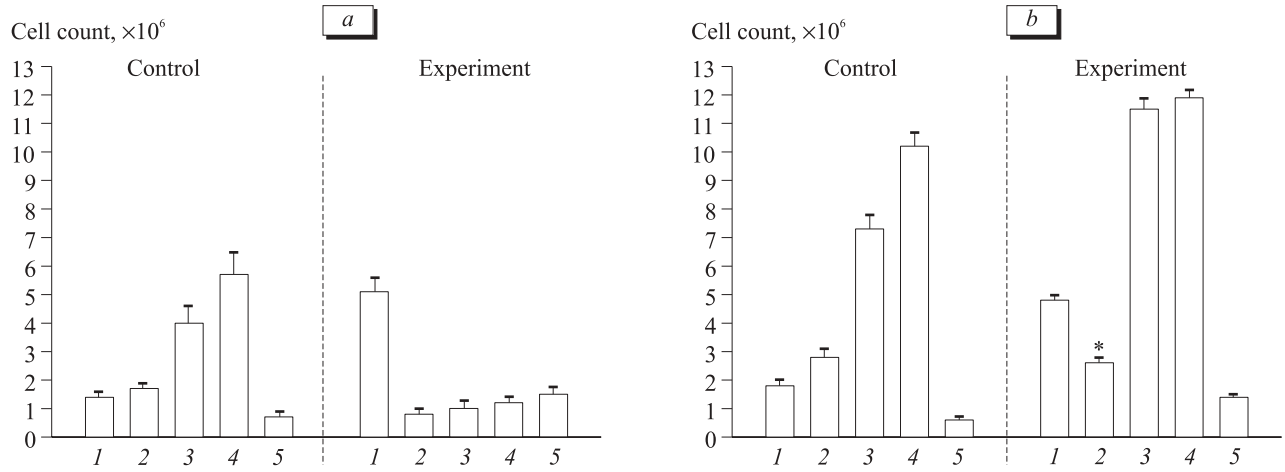

Fig. 1. Quantitative characteristics of spermatogenic epithelium in rapidly aging SAMP1 (*a*) and SAMR1 (*b*) mice. Vertical line: absolute number of cells/testicle. *1*) spermatogonia; *2*) pachytene spermatocytes; *3*) round spermatides; *4*) spermia; *5*) Sertoli cells. *No significant difference *vs.* control at $p<0.05$.

and testicular spermia decreased 2.12-, 4.00-, and 4.75-fold, respectively (Fig. 1). Histological study revealed destructive changes in the spermatogenic epithelium in the majority of testicular tubules in experimental SAMP1 mice. The structure of male gametes in males not treated with carnosine for a long time was ordered, without large foci of destructive changes. On the other hand, abnormal cell forms were detected in many cases (Fig. 2, *a-d*).

The number of spermatogonia and Sertoli cells in SAMR1 mice treated with carnosine by 2.67 and 2.33 times surpassed the normal for these animals (Fig. 1). The counts of pachytene spermatocytes, round sper-

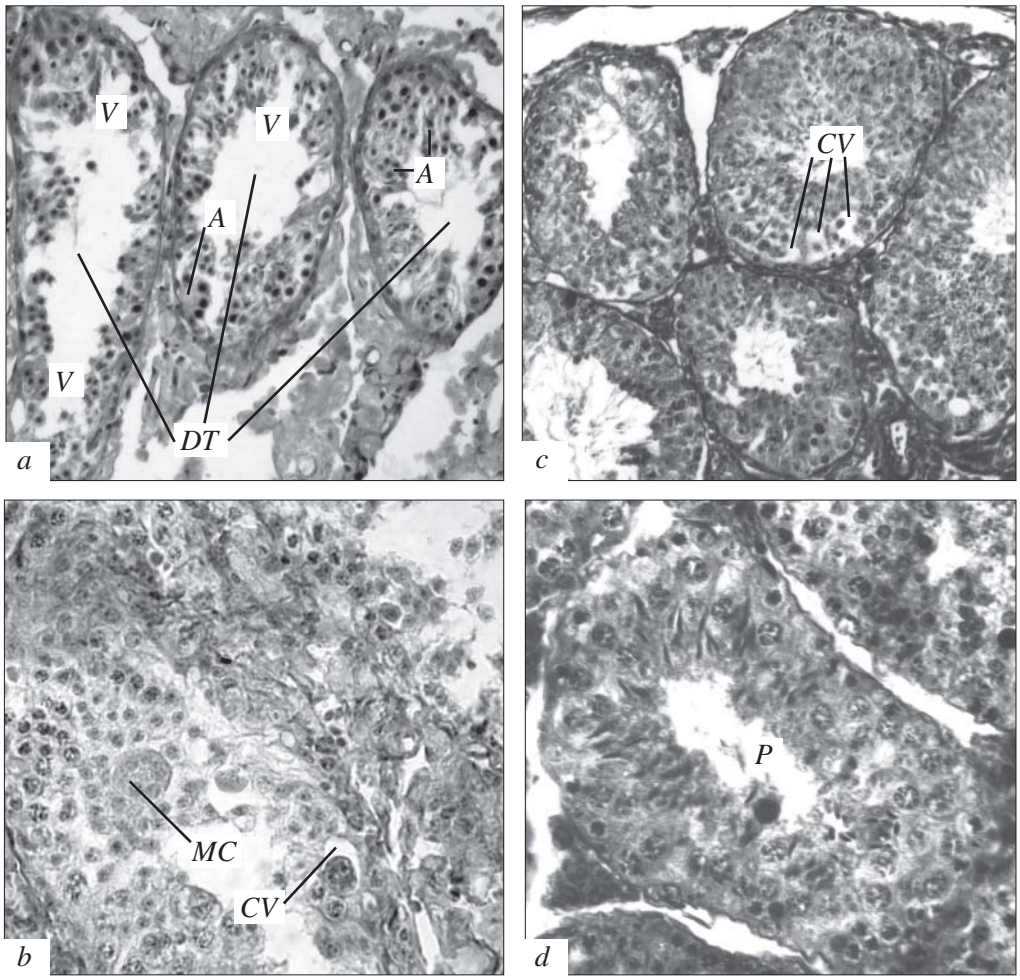

Fig. 2. Sections of testicles from experimental (*a*, *b*) and control (*c*, *d*) SAMP1 mice. *DT*: destructive tubules, with just few sex cells of higher stages of development; *V*, *A*: vacuolation and atomization of spermatogenic epithelium; *MC*: multinuclear cell at the stage of round spermatides; *CV*: cytoplasmic vacuoles in sex cells; *P*: pyknosis; ×160 (*a*, *c*), ×400 (*b*, *d*). Hematoxylin and eosin staining.

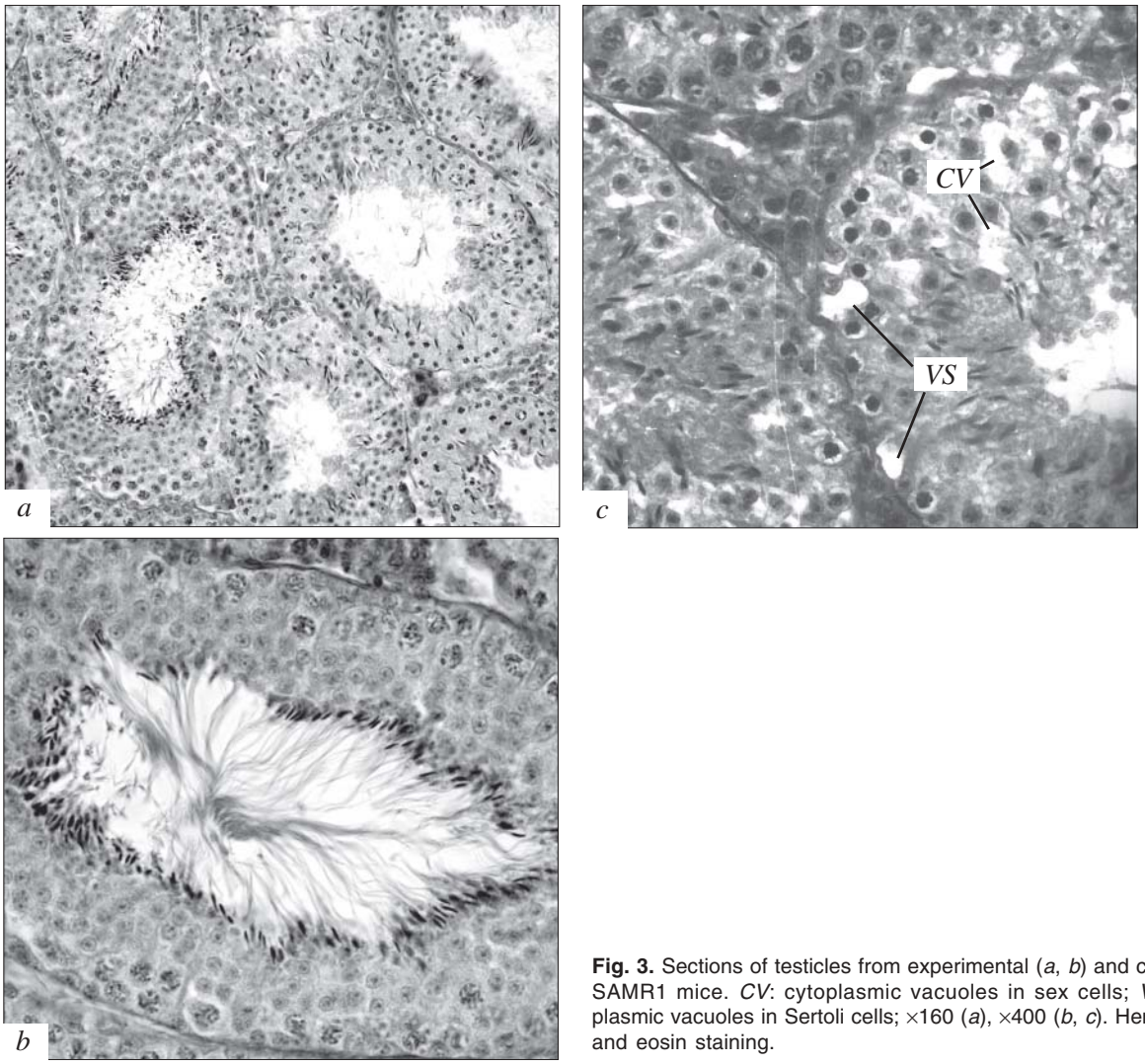

Fig. 3. Sections of testicles from experimental (*a*, *b*) and control (*c*) SAMR1 mice. *CV*: cytoplasmic vacuoles in sex cells; *VS*: cytoplasmic vacuoles in Sertoli cells; ×160 (*a*), ×400 (*b*, *c*). Hematoxylin and eosin staining.

matides, and spermia did not change or, despite the significance of changes, changed negligibly. Morphological study of the spermatogenic epithelium in experimental SAMR1 mice showed a lesser number of disorders compared to the control group (Fig. 3, *a-c*).

The spermatogenic system of senescence accelerated mice is genetically unstable, and cell systems burdened with mutations and hence, enthropic protoplasmic environment, can be modified in any direction under the effect of bioactive compounds and other external energy sources. For example, X-raying of testicles in young rat pups did not accelerate degeneration of damaged gonocytes, but promoted their long-term survival [8].

Presumably, carnosine changed the proliferation/apoptosis balance in SAMP1 and SAMR1 mice via stimulation of mitotic division or by attenuating the functions of proteins and enzymes mediating the mechanism of cell selection (death).

More detailed interpretation of these findings is hardly possible, because we do not know the response of spermatogenic cells to carnosine, for example, in 22-24-month-old SAMR1 mice (maximum threshold life span) and in normally aging animals. However, our experiments confirmed nonlinear effect of carnosine on the development of male gonads of rapidly aging mice.

REFERENCES

1. *Biokhimiya*, **65**, No. 7, 883-1024 (2000).
2. A. A. Boldyrev, *Carnosine* [in Russian], Moscow (1998).
3. A. A. Boldyrev, *Biokhimiya*, **65**, 884-890 (2000).
4. S. Gallant, M. L. Semenova, and M. Yuneva, *Ibid*, 1018-1021 (2000).
5. S. T. Zakhidov, A. V. Gopko, M. L. Semenova, *et al.*, *Byull. Eksp. Biol. Med.*, **134**, 89-92 (2002).
6. S. T. Zakhidov, O. F. Gordeeva, and T. L. Marshak, *Izv. Rossiisk. Akad. Nauk*, Series *Biology*, No. 1, 23-30 (2001).
7. S. T. Zakhidov, T. L. Marshak, I. V. Uryvaeva, *et al.*, *Ontogenez*, **6**, 444-456 (2002).
8. G. G. Shchegolev, *Arkh. Anat. Gistol. Embriol.*, **XIII**, No. 2, 315-326 (1934).