



Short Communication

Exogenous melatonin administration is beneficial for male reproductive function during normal ageing

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Keywords:

Melatonin, sperm count, sperm motility, normal ageing

ABSTRACT

Background: A concern in the use of exogenous melatonin as a therapeutic intervention is that it may interfere with reproductive function. Herein, we report that chronic exogenous melatonin administration does not impair male reproductive function during ageing and at old age in male Sprague Dawley rats. **Methods:** Reproductive function was assessed from spermatozoa derived from the cauda epididymis in 3 months, 9 months, 12 months and 24 months old male Sprague Dawley rats after 0.1mg/kg melatonin administration for 8 weeks. Data were analyzed by Student's unpaired t-test. **Results:** Sperm motility and sperm count was preserved in 3 months, 9 months, 12 months and 24 months old rats compared with untreated controls ($P>0.05$). The 24 months old rats showed high levels of sperm motility and count with melatonin treatment. **Conclusion:** We conclude that exogenous melatonin administration preserves male reproductive function during ageing in male Sprague Dawley rats.

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INTRODUCTION

The level of melatonin drops significantly during puberty to allow for sexual maturation (Waldhauser et al., 1988; Karasek, 2007). A high melatonin level during this stage of life has been reported to delay sexual maturation, reduce gonadal development and thus prevent reproductive capacity (Rissman, 1980, Amador et al., 1986). Short photoperiod or long photoperiod with melatonin treatment has also been reported to delay gonadal development in rodent populations (Zucker et al. 1980, Deveson et al., 1992). The role of chronic melatonin administration on reproductive function in pre-pubertal, young, middle-aged, old mice and female rats has been documented (Pierpaoli et al., 1997; López et al., 2005). Gwayi and Bernard reported a dose dependent decrease in all

parameters of sperm motility with melatonin treatment *in vitro* on spermatozoa collected from adult Wistar rats (Gwayi and Bernard, 2002). Melatonin treatment improved epididymal sperm concentration and motility in 6-7 months old homocysteine treated Wistar rats (Sönmez et al., 2007). However, the role of chronic treatment of melatonin in male reproductive function at old age in male Sprague Dawley rats remains unanswered. This paper presents results from a comparative study on the outcome of melatonin administration on male reproductive function during ageing in Sprague Dawley rats.

MATERIALS AND METHODS

Animals and treatment

A total of twenty-two animals were used for the experiment. Animals were raised and kept in the animal house of the Lagos State University College of Medicine. Institutional guidelines for the care and use of animals were followed. Animals were kept in 12hour light: 12 hour dark lighting condition and $24\pm 6^{\circ}\text{C}$. The animals were divided into four different age groups. Each age group had a melatonin treated group and a control group which received ethanolic saline. Melatonin used for this experiment was supplied by Sigma, Aldrich. The treatment groups received 0.1mg/kg melatonin dissolved in 10% ethanolic saline

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while controls received ethanolic saline only. At the termination of the experiment, animals were sacrificed by cervical dislocation. The paired testes and epididymis were quickly dissected out for sperm analysis.

Sperm Count

Reproductive tests were carried out after sacrificing the animals by cervical dislocation. Sperm was drawn from the epididymis of each animal for assessment of morphology and count using the light microscope and the haemocytometer for counting. Spermatozoa in the cauda epididymidis was diluted to 1ml with normal saline. Haemocytometer (Improved Neubauer chamber), was filled with sperm suspension from cauda epididymidis diluted 1: 20 with formal saline. This was charged unto the counting chamber and the sperm cell counted using x40 Objective. To count the number of cells, five squares was read and the average of the count from the five squares was multiplied by 1 (one) million (WHO, 1999).

Sperm Motility

We assessed sperm motility immediately the epididymis was excised according to the method described by (Minaii et al., 2014) with some modification. Briefly, one of the paired epididymis was excised on a warm slide at 37°C. A drop of warm saline 37°C was added and the mixture covered with cover slip. Motility was scored under X40 objective using a light microscope.

Data and Statistical Analysis

Data was reported as +SEM and statistical Analysis was carried out using unpaired t-test with a significance level of P<0.05.

RESULTS

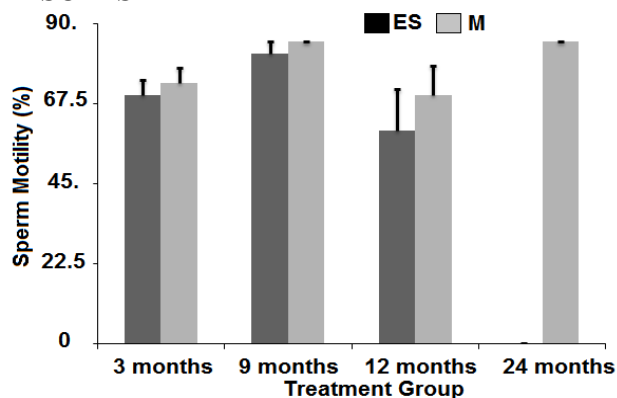


Fig. 1: Sperm Motility Scores in ethanolic saline (ES) and melatonin (M) treated rats in percentages P>0.05 (Melatonin treated 3 months vs Melatonin treated 9months); P>0.05 (Melatonin treated 9 months vs Melatonin treated 12months); P>0.05 (Melatonin treated 12 months versus Melatonin treated 24 months)

DISCUSSION

We studied the role that exogenous melatonin may play in reproductive parameters during ageing and at old age. In this study, exogenous melatonin did not show any adverse effect on sperm motility or sperm count at old age, although there were insignificant reductions in the parameters measured in the younger groups. From these results we speculate that melatonin due to its lipophilicity, hydrophilic and ubiquitous characteristic may improve reproductive profile during old age through its strong antioxidant property and ability to penetrate cellular organelles as reported by Reiter et al., 2002, Messner et al., 1998 and Martin et al, 2000.

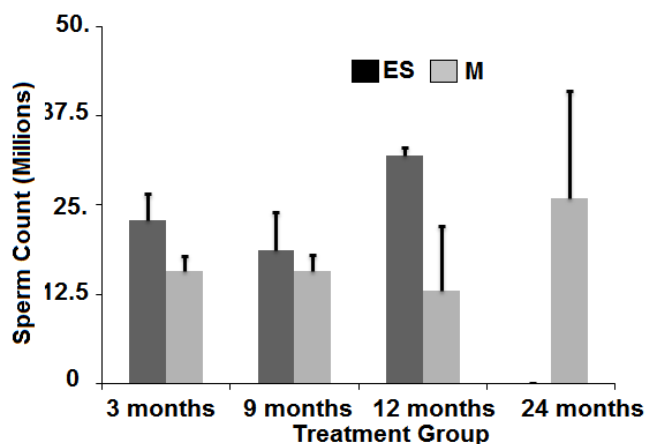


Fig. 2: Sperm Count in ethanolic saline (ES) and melatonin (M) treated rats in million. P>0.05 (Melatonin treated 3 months vs Melatonin treated 9months); P>0.05 (Melatonin treated 9 months vs Melatonin treated 12months); P>0.05 (Melatonin treated 12 months versus Melatonin treated 24 months)

In contrast with reports on melatonin administration in adult Wistar rats by Gwayi and Bernard, 2002, we found no reduction on the sperm motility scores in 9 months and 12 months old male Sprague Dawley rats (Figure 1). However, the reduction in sperm count observed in the 3 months, 9 months and 12 months melatonin treated rats (Figure 2) is consistent with earlier studies on its anti-gonadal function in pubertal rats (Hastings et al., 1985; Olatunji-Bello and Sofola, 2001), although the reduction we observed was insignificant.

It is important to mention that in this study, the 24 months old animals that received ethanolic saline only, all died before the terminal date of the experiment (Figure 1 and Figure 2). This was not the case with the melatonin treated animals which had about 66% survival rate. A limitation to this study is the limited number of animals especially in the older groups due to deaths recorded during the experiments.

As melatonin receptors have been found in mammalian epididymis (Shiu et. al., 1997), we speculate that action of melatonin on sperm motility and count during old age may be an up-regulation of the melatonin receptors in the epididymis or testis. Although the mechanism of its action in male reproductive function during ageing is yet to be elucidated, we found a beneficial role for exogenous melatonin on male reproductive function during ageing and at old age.

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