INTRODUCTION

Vasectomy is the surgical ligation and division of the vas deferens carried out as male contraception. It is a widely accepted form of contraception especially in India, China, United States and new Zealand1, 2. It is usually regarded as a permanent surgical procedure with no assurance of successful reversal as far as reproductive function is concerned3, 4. There are some variations on the procedure such as no-scalpel (keyhole) vasectomy which seals only the top end of the vas. With this method sperm are free to spill out from the lower severed end of the vas thus avoiding any build-up of pressure. The likelihood of long-term testicular pain from “backup pressure” seems to be eliminated using this method5, 6. Therefore factors considered in making the choice of the type of vasectomy include post-procedure morbidity, side effects and choice of the client7, 8. Irrespective of the procedure, the ultimate aim of vasectomy is to prevent the ejaculation of spermatozoa following coitus. However, it has been observed by several investigators that following vasectomy, the testes soon begin to undergo structural damage9, 10, 11.

Evidence for post-vasectomy testicular pathology includes testicular atrophy, destruction of epithelium of the seminiferous tubules, absence of spermatozoa, thickening and folding of the tubular membranes and dilatation of the interstitial space12. Other abnormalities described include testicular interstitial fibrosis13, enhanced apoptosis of spermatogonic cells14 and morphological degeneration in the sperm tail and head15, epididymal distension and sperm granuloma formation16 with pathological changes in the vas deferens proximal and distal to the vasectomy site17. There are also data to show increased lymphocyte activity and increased anti-sperm antibody production18, increased pressure of distal epididymis, increased size and number of lysosomes in epididymis and chronic inflammation of epididymal interstitium19. The incidences of testicular and prostate cancer have been reported to increase following vasectomy20, 21 whereas other reports have opposed this view22, 23. The testicular damage following vasectomy is not uniform, but depends on the animal species, technique of vasectomy and the time interval after vasectomy24. Several mechanisms have been proposed as being causally related to the testicular damage following vasectomy. These include increased hydrostatic pressure in the testis, increased production of anti-sperm auto-antibodies, and oxidative stress resulting from increased production of reactive oxygen or nitrogen species25, 26. Increased hydrostatic pressure in the testes is a more plausible explanation10. Conversely anti-sperm antibodies do not seem to play the same role on all species studied so far. In the rabbit it does not seem to be associated with testicular damage following vas deferens obstruction27, whereas in the rat, its production precedes the testicular damage19.

Most men who have chosen vasectomy as the choice of contraception value the freedom from artificial birth control and the fear of unplanned pregnancy. However, some people come to regret the operation and want it reversed, usually because they have a new partner or want more children28, 29. There is paucity of data on the precise cause of testicular damage following vasectomy and the possibility of reversal of the testicular damage. Preventing or reversing the testicular damage following vasectomy may imply a more favorable outcome in those patients asking for vasectomy reversal.

MATERIALS AND METHODS

Animals

The study comprised 80 adult male Sprague-Dawley rats (S-D) obtained from the Laboratory Animal Centre of the College of Medicine of the University of Lagos. They were authenticated by a taxonomist30 in the Department of Zoology of the University of Lagos. Animals were kept in metal cages in the Animal Room of the Department of Anatomy and allowed to acclimatize for two weeks under standard laboratory conditions of temperature 27-30°C, with a photoperiodicity of approximately twelve hours light alternating with twelve hours of darkness. They were fed with commercially available rat chow (Livestock feeds Plc., Ibeja, Lagos Nigeria) and had unrestricted access to water.
**Experimental protocol**

The experimental rats were divided randomized into four groups (I to IV) of 10 S-D rats per group. They were made up of a sham operated group and three different obstructive vasectomy models. Group I: Intact rats; Group II: Ligation applied between caput epididymis and upper pole of the testis; Group III: Ligation applied to both ends (proximal and distal) after division of vas deferens 2-3 cm from caudal epididymis; Group IV: Vas divided 2-3 cm from caudal epididymis and ligation applied only to the proximal end. In addition, five animals in each group received 1 mg/kg melatonin intra-peritoneally.

Each experimental group was controlled matched with 10 S-D rats that underwent right sided orchiectomy used for the estimation of baseline testicular malondialdehyde levels and cytometry (measurement of cells and cellular constituents). After 12 weeks, the testes were removed for MDA estimation, histological processing for testicular morphometric measurements.

**Anesthesia and the surgical procedure**

The anesthesia was administered by an intra-peritoneal injection of 50 mg/kg ketamine hydrochloride. All operations were carried out on the right side through a scrotal incision. In the sham operated group, the testes and spermatic cord were exposed and closed. In the right testis in all experimental groups (p < 0.05). Ligation of the vas deferens of the right testes did not significantly affect the diameter of the corresponding contralateral testis. Ligation of the vas deferens caused a significant decrease in mean seminiferous tubular diameter in all experimental groups (p < 0.05). The seminiferous tubular diameter was least affected when only the proximal end of the divided vas deferens was ligated. Mean testicular tubular diameter was not affected in the intact left testis in all experimental groups (Table 2).

**Results**

Testicular malondialdehyde levels in Sprague-Dawley rats

In all the groups, ligation of the vas deferens caused a significant increase in MDA when melatonin was not given (p < 0.05). Testicular MDA increased as the point of ligation got closer to the testis. At each point of ligation, the testicular MDA was significantly reduced if melatonin was administered (p < 0.05). Testicular MDA was least when only the proximal end of the sectioned vas deferens was ligated (Table 1).

**Table 1: Malondialdehyde levels in adult rat testes (µmol/mg protein) in three obstructive models of the vas deferens ± Melatonin (Mel)**

<table>
<thead>
<tr>
<th>Groups</th>
<th>(-Mel)</th>
<th>(+Mel)</th>
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<tbody>
<tr>
<td>I</td>
<td>0.38±0.24</td>
<td>0.38±0.07</td>
</tr>
<tr>
<td>II</td>
<td>0.92±0.16</td>
<td>0.47±0.23</td>
</tr>
<tr>
<td>III</td>
<td>0.69±0.02</td>
<td>0.42±0.14</td>
</tr>
<tr>
<td>IV</td>
<td>0.40±0.21</td>
<td>0.38±0.02</td>
</tr>
</tbody>
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**I - Intact rats**

**II - Ligation applied between caput epididymis and upper pole of the testis**

**III - Ligation applied to both ends after division of vas deferens 2-3 cm from caudal epididymis**

**IV - Vas divided 2-3 cm from the caudal epididymis and ligation applied only to the proximal end**

Testicular Morphometry in Sprague-Dawley rats

The diameter of the right testis was significantly reduced in all experimental groups compared to the controls (p < 0.05). Ligation of the vas deferens of the right testes did not significantly affect the diameter of the corresponding contralateral testis. Ligation of the vas deferens caused a significant decrease in mean seminiferous tubular diameter in all experimental groups (p < 0.05). The seminiferous tubular diameter was least affected when only the proximal end of the divided vas was ligated. Mean testicular tubular diameter was not affected in the intact left testis in all experimental groups (Table 2).

**Table 2: Bilateral testicular diameters and mean seminiferous tubular diameter (MSTD) following three obstructive models of the vas deferens**

<table>
<thead>
<tr>
<th>Test groups</th>
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<td>I</td>
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<td>III</td>
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<td>IV</td>
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<table>
<thead>
<tr>
<th>Number of rats (n)</th>
<th>5</th>
<th>12</th>
<th>12</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean diameter (mm)</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Right testis</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>15.8</td>
<td>11.2</td>
<td>12.8</td>
<td>14.6</td>
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<tr>
<td>Mean diameter (mm)</td>
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<td></td>
</tr>
<tr>
<td>Left testis</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15.2</td>
<td>14.8</td>
<td>15.0</td>
<td>15.4</td>
<td></td>
</tr>
<tr>
<td>MSTD (µm)</td>
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<td></td>
<td></td>
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<tr>
<td>Right testis</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>198.2</td>
<td>180.3</td>
<td>178.6</td>
<td>176.4</td>
<td></td>
</tr>
<tr>
<td>MSTD (µm) Left testis</td>
<td>196.2</td>
<td>194.7</td>
<td>198.2</td>
<td>198.5</td>
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</table>

**Statistical analysis**

All data were expressed as mean ± standard deviation. Analysis was carried out using analysis of variance (ANOVA) with Schefffe’s post hoc test. The level of significance was considered at p < 0.05.
testicular damage is believed to result from both increased intratesticular pressure and the production of anti-sperm auto-antibodies as well as oxidative stress. This results from increased production of reactive oxygen or nitrogen species. An amplified hydrostatic pressure is believed to be more important in the pre-pubertal than the adult rat, and is suspected to be due to the developed collateral circulation, and the rat testicular microcirculation with a rhythmic pattern of arteriolar dilatation and constriction demonstrated in the mature rat. Anti-sperm antibodies though outside the scope of this study do not seem to have the same significance in all species studied so far. In the rabbit it does not appear to be associated with testicular damage following vasectomy protocols. We investigators who used morphometric parameters to assess drainage of the seminiferous tubules, abnormal changes were not because when the testicular end of the duct is left open, with luminal pressure may be the primary cause of changes in the testis, structure in a six month period.

To conclude that vasectomy alters ipsilateral testicular structure and testicular structure to vasectomy.

REFERENCES

