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·*Original Article*·

Morphological characteristics of spermatozoa before and after renal transplantation

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Abstract

Aim: To investigate the changes of the spermatozoa ultrastructures before and after renal transplantation in uremic patients. **Methods:** The sperm of five uremic patients before and after transplantation and four healthy volunteers were collected and examined by scanning electron microscopy. **Results:** Abnormal spermatozoa were found in patients pre-transplantation; abnormalities included deletion of the acrosome, absence of the postacrosomal and postnuclear ring, dumbbell-like changes of the head, tail curling, and absence of the mitochondrial sheath in the mid-segment. After renal transplantation, most of the spermatozoa became normal. **Conclusion:** There are many abnormalities with regard to the appearance and structure of the head, acrosome, mitochondria and tail of the spermatozoa in uremic patients. The majority of the spermatozoa returned to normal after renal transplantation, but a few still presented some abnormalities possibly relating to the administration of immunosuppressants. *(Asian J Androl 2005 Mar; 7: 81–85)*

Keywords: uremia; renal transplantation; spermatozoa; scanning electron microscopy

1 Introduction

Previous investigations have indicated that many abnormalites of spermatozoa in uremic patients were corrected after successful renal transplantation [1].

The present study used scanning electron microscopy (SEM) to further investigate the morphological and ultrastructural changes of the spermatozoa in these patients before and after renal transplantation.

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2 Materials and methods

2.1 Clinical characteristics

Our study included five uremic male patients aged 27 to 38 (mean 31) years, all married with healthy children. All had sufferred with chronic glomerulonephritis for a period of 12 to 35 (mean 21.6) months. Hemodialysis (2–3 times per week) was maintained for 2–13 months (mean 7.2) before sperm inspection. Eight to fifteen (mean 12.2) months after successful transplantation the sperm were again collected for inspection. Sperm from four healthy men aged 30–32 (mean 31.3) years (volunteers from the hospital staff) were also examined as controls. All the patients and volunteers were nonsmokers and nondrinkers. Written informed con-

sent was obtained from every subject before the study commenced.

2.2 Methods

Sperm were collected by means of masturbation following at least 3 days of abstention. Sperm were liquefied at room temperature for 30 min, diluted and centrifugalized at 2000 rpm for 10 min. Following removal of supernatants, specimens were washed with normal saline and centrifuged again. The sediment was then aspirated and 2 mL of glutaric dialdehyde (2.5 %) were added. After fixation, drying, desiccation and aurum spraying, the spermatozoa were examined with SEM (Cambridge S260, UK).

3 Results

3.1 Spermatozoa in healthy men

A normal human spermatozoon was tadpole-like with a head and a tail. The head was 4–5 μm long. The length of the main-, mid- and end-segment of the tail was appproximately 45, 5–7 and 5 μm, respectively. Both the head and tail were well developed. The structure of the acrosomal membrane was integrated with a clear demarcation among the postacrosomal and postnuclear ring and each segment of the tail (Figure 1).

3.2 Spermatozoa in uremic patients

Different kinds of deformity in the spermatozoa were observed under SEM. The head presented the following anomalies: (1) absence of the acrosome, indistinct post-acrosomal and postnuclear ring (Figures 2, 3); (2)dumb-bell-like head with irregular contour (Figure 4); and (3) coarseness and unevenness of the surface, absence of the postacrosomal and postnuclear ring (Figure 5). The deformities of the tail included: (1) indistinct mitochondrial sheath in the mid-segment (Figures 2, 4); (2) obvious curl of the tail encircling the head (Figure 5); and (3) mixed deformities (i.e. two or more abnormalities present in a single spermatozoon) (Figures 2–5).

3.3 Spermatozoa in post-transplanted patients

The majority of the spermatozoa was normal after renal transplantation (Table 1). Under SEM both the head and tail could be seen distinctly. The ultrastructure of the acrosome turned towards integrated, the post-acrosomal and postnuclear ring were dimly visible. No obvious abnormalities could be observed (Figure 6). A few spermatozoa showed an irregular configuration in the head but had a normal tail (Figure 7).

4 Discussion

The abnormalities of the spermatozoa in uremic patients were much more visible by SEM than by light microscopic examination. Prem *et al.* [1] reported the results of sperm analysis in 19 male patients 6 months after maintained hemodialysis; testis biopsy was undertaken

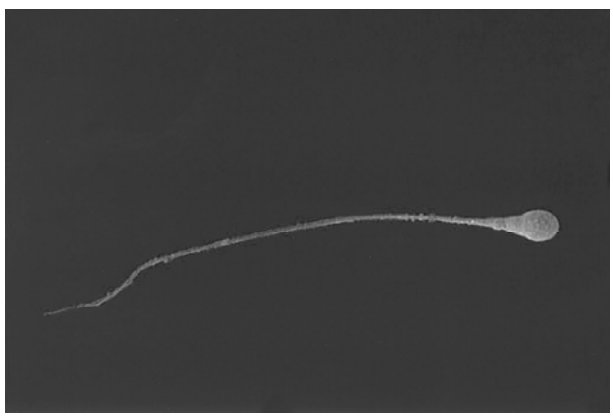

Figure 1. This normal human spermatozoon shows the following characteristics: head and tail well-developed, structure of acrosomal membrane integrated, distinct demarcation among the neck and mid-, main- and end-segment of the tail. The postacrosomal and postnuclear ring of the head and mitochondrial sheath of the mid-segment of the tail are dimly visible (×2120).

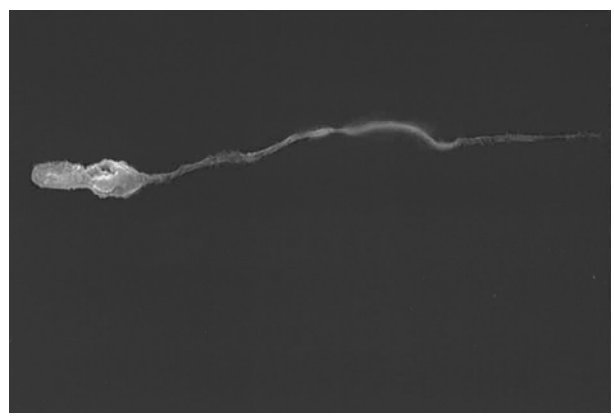

Figure 2. This image shows the following deformaties: deletion of the acrosome, indistinction of postacrosomal and postnuclear ring as well as the mitochondrial sheath of the mid-segment of the tail and poor stretching of the tail (×2360).

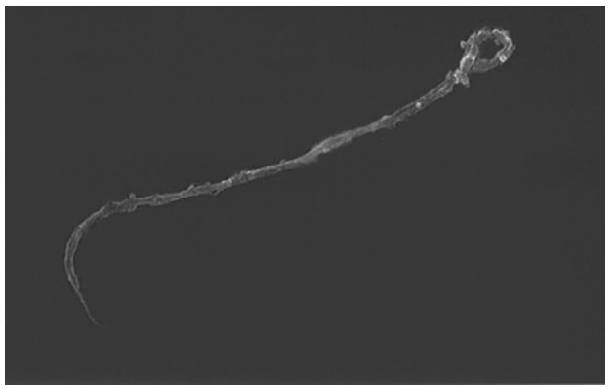

Figure 3. This image shows the acrosome partly deleted, with distinct foam in nuclear and spiculated structure in the curling tail (×3000).

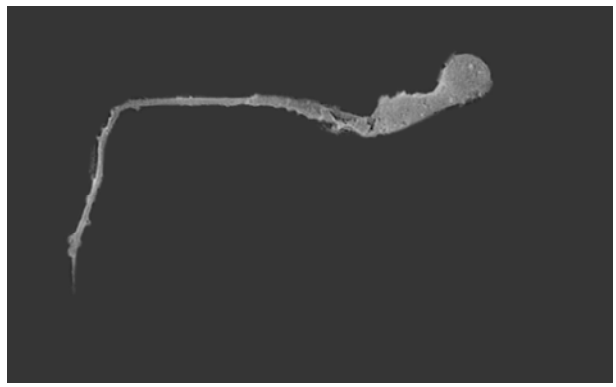

Figure 4. This image shows the following deformities: dumb-bell-like head, structure of membranous foramen, postacrosomal and postnuclear ring as well as the indistinct mitochondria sheath of the mid-segment (×2860).

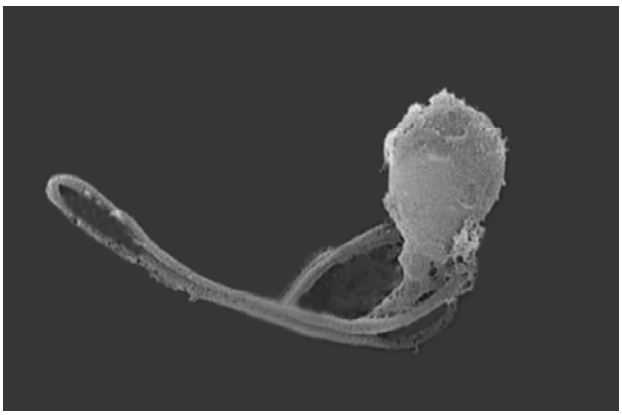

Figure5. This image shows the coarse surface of the head; the postacrosomal and postnuclear ring are invisible. The severe curl of the tail enveloping the head (×6290).

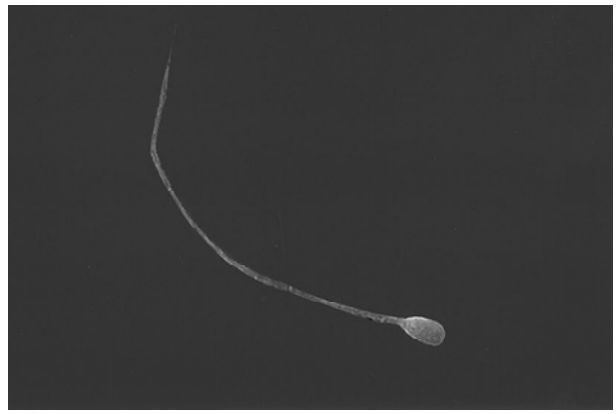

Figure 6. The acrosomal structure is integrated; the postacrosomal and postnuclear ring as well as the mitochondrial sheath of the mid-segment are dimly visible (×1710).

in eight patients with azoospermia or oligozoospermia. Of these there were three patients with hypo-spermatogenesis, four with retardation of spermatozoal maturity and one with dysplasia of gonobasts. Wu *et al.* [2] reported that the quality of the sperm was greatly decreased in uremic patients, which might be related to the absence of the mitochondrial sheath found in this study. Yogev *et al.* [3] reported that in five of six uremic rats, spermatogenic systems and fertility basically remained normal other than a slight reduction in the blood testosterone level after nephrectomy. The dissimilarity in results of these studies may be due to species difference. Prem *et al.* [1] examined the sperm in 19 patients 6 months after renal transplantation. They found the quality of the sperm was improved, and the density and activity of the

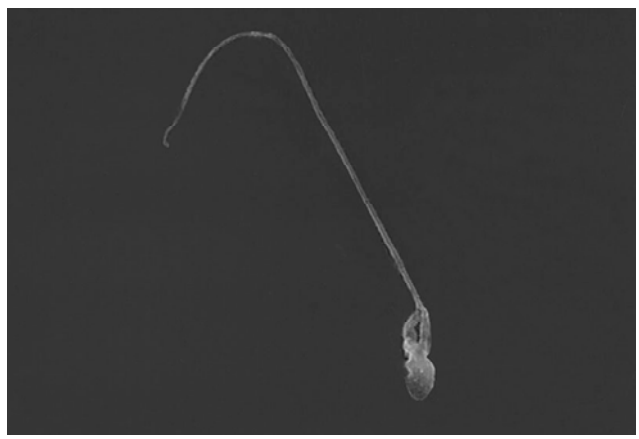

Figure 7. Irregularity of head, postacrosomal and post nuclear ring indistinct. Tumescence of neck-segment. Demarcation among mid-, main- and end-segment of tail distinct (×1850).

Table 1. Scanning electron microscopic observation on morphologic changes of sperm in uremic patients before and after renal transplantation.

Patient (age)	Before renal transplantation		After renal transplantation	
	Head	Tail	Head	Tail
1 (27)	Morphological structure extremely irregular, deficit of acrosome, disappearance of postacrosomal and postnuclear ring.	Curled tail, poor stretching, had partial spinous structure.	Acrosomal structures tend to be complete, Postacrosomal and post nuclear ring indistinct.	Stretching of all segments basically normal.
2 (28)	Dumb-bell shaped head with deficit of acrosome and irregular edge.	Mitochondrial sheath indistinct, part of it was uneven in thickness.	Acrosomal structure basically complete with regular edge.	Mitochondrial sheath may be seen indistinctly, delimitation between segments basically normal.
3 (30)	Uneven surface, disappearance of postacrosomal and postnuclear ring.	Evident curled tails, some of them had short tails, deficit of mitochondria.	Smooth surface, acrosomal structure complete, postacrosomal and postnuclear rings are indistinct.	Delimitation between segments clear without curling.
4 (32)	Deficit of acrosome, external acrosomal membrane and inner acrosomal membrane.	Deficit of mitochondrial sheath, part of it was uneven in thickness and had spinous structure.	Acrosomal structure, basically normal.	Mictochondrial sheath may be seen indistinctly, all segments basically normal.
5 (38)	Irregular spinous protrusion, deficit acrosome.	Terminal segment invisible, had partial deficit.	Acrosomal structure rather regular, enlargement of neck.	Complete mitochondrial sheath, delimitation between neck, mid-, main- and terminal segments rather clear.

sperm were greatly increased. Testicular biopsy was carried out in seven patients; of those spermatogenesis returned to normal in four and maturity retardation of the spermatozoa remained in three [1, 4]. Holdsworth *et al.* also found successful renal transplantation played a significant role in the improvement of the testicular function impaired by uremia [5]. Our study showed that the configuration of the spermatozoa returned distinctly to normal following renal transplantation. These findings demonstrated that after transplantation the restoration of the configuration of the spermatozoa did occur to a certain extent. Nevertheless, there still remained certain abnormalities in a few spermatozoa as mentioned also by Prem *et al.* [1]. On the other hand we think the side-effects of immunosuppressant cyclosporin A (CsA) should also be considered. In the animal experiment there was no damage to the testicles at a CsA dose of 10 mg·kg^{-1}, but when the dose was increased to $\geqslant$ 20 mg·kg^{-1}, a decrease in testicular weight, activity and quantity of the spermatozoa and degeneration, and necrosis of Leydig and Sertoli cells would occur [6]. Li *et al.* reported the effects of different CsA dosage on rat testes by transmission electron microscopy. They found the development of the seminiferous epithelia and spermatozoa were inhibited at a dose of 10 mg·kg^{-1}, the structural changes of the seminiferous epithelia and spermatozoa, degeneration and necrosis of the spermatocytes and spermatoblasts followed by abnormal development of the spermatozoa occurred at a dose of 20 mg·kg^{-1}[7]. Hanbermen *et al.* reported that of nine post-transplanted patients treated with CsA at a dose of 3 mg·kg^{-1}·day^{-1}, sperm analysis was normal in eight patients [8]. Thus there exists a correlation between the recovery of a spermatozoa deformity and the CsA dosage. It was demonstrated that a blood concentration of CsA greater than 600 μg·L^{-1} was the potential toxic concentration [9]. Therefore, we suggest that a lower dose CsA administration and a close monitoring of the CsA blood concentration are essential strategies for the restoration of the spermatozoa function after renal transplantation.

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