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ORIGINAL ARTICLE

Male Health

Apparent diffusion coefficient values of normal testis and variations with age

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The usefulness of diffusion-weighted magnetic resonance imaging (DWI) in the evaluation of scrotal pathology has recently been reported. A standard reference of normal testicular apparent diffusion coefficient (ADC) values and their variations with age is necessary when interpreting normal testicular anatomy and pathology. We evaluated 147 normal testes using DWI, including 71 testes from 53 men aged 20–39 years (group 1), 67 testes from 42 men aged 40–69 years (group 2) and nine testes from six men older than 70 years (group 3). DWI was performed along the axial plane, using a single shot, multislice spin-echo planar diffusion pulse sequence and b-values of 0 and 900 s mm⁻². The mean and standard deviation of the ADC values of normal testicular parenchyma were calculated for each age group separately. Analysis of variance (ANOVA) followed by post hoc analysis (Dunnnett T3) was used for statistical purposes. The ADC values ($\times 10^{-3}$ mm² s⁻¹) of normal testicular tissue were different among age groups (group 1: 1.08 ± 0.13 ; group 2: 1.15 ± 0.15 and group 3: 1.31 ± 0.22). ANOVA revealed differences in mean ADC among age groups ($F = 11.391$, $P < 0.001$). Post hoc analysis showed differences between groups 1 and 2 ($P = 0.008$) and between groups 1 and 3 ($P = 0.043$), but not between groups 2 and 3 ($P = 0.197$). Our findings suggest that ADC values of normal testicular tissue increase with advancing age.

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INTRODUCTION

Diffusion-weighted magnetic resonance imaging (DWI) with calculation of apparent diffusion coefficient (ADC) values has been established as a useful functional diagnostic tool in urogenital imaging.^{1–12} A few recently published reports have addressed the diagnostic performance of DWI in the evaluation of various scrotal pathologies.^{13–17} However, data are needed to establish standard references of normal testicular ADC values.

Aging is associated with structural and functional alterations of normal testicular tissue.^{18–20} Although gonadal function declines with age in both men and women, women experience an abrupt loss of ovarian function, while in men a more gradual, incomplete age-related decline in gonadal function occurs, with a high degree of interindividual variability.^{21–25} A decline in several important sex hormones, including testosterone, is observed with advancing age in men.^{18–25} Specifically, in middle-aged men (40–69 years), total and free serum testosterone levels fall by 0.8% and 2% per year, respectively. In older men above the age of 65 or 70 years, significant declines in free testosterone occur.^{21–25} The term ‘andropause’ is considered inappropriate and the terms ‘symptomatic LOH’ (late onset hypogonadism) or ‘symptomatic ADAM’ (androgen deficiency of aging male) are considered more accurate to describe particular symptoms and low serum testosterone levels.^{21–25} The goal of this study was to determine the ADC values of normal testes and to assess the magnitude of variations with age.

MATERIALS AND METHODS

Study population

This was a retrospective review of a consecutive series of 102 magnetic resonance imaging (MRI) examinations of the scrotum, performed from May 2009 to March 2013. The age range of men included was 20–81 years and the mean age was 42 years. The men were referred to the Urology Department for a variety of clinical symptoms (Table 1). The records included clinical and imaging examinations, surgical findings and pathologic results. Radical orchiectomy was performed in 31 patients, testicular biopsy in five cases and lesion excision in two patients. The time interval between MRI examinations and surgery was less than 2 weeks in all cases.

Because of the retrospective nature of the study, the institutional review board did not require approval or patients’ informed consent.

MRI protocol

All MR examinations were performed on a 1.5-T Intera scanner (Philips Medical Systems, Cleveland, OH, USA) using a pelvic phased-array coil, a field of view of 240 × 270 mm and an acquisition matrix of 180 × 256 mm. All patients were examined in supine position, with the testes resting on a towel at a similar distance from the coil and the penis draped on the anterior abdominal wall. Axial spin-echo T1-weighted images (repetition time/echo time (TR/TE), 500–650/13–15 ms; scan time: 210 s), and transverse, sagittal and

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coronal fast spin-echo T2-weighted images (TR/TE, 4000/100–120 ms; scan time: 210 s) were analyzed. Images were of 3–4 mm slice thickness, with a 0.5-mm gap. DWI was performed along the axial plane, using a single shot, multislice, spin-echo planar sequence with the following parameters: TR, 3900 ms; TE, 115 ms; number of signals averaged, 1; motion-probing gradient (MPG), 3; matrix, 180 × 256 mm; field of view, 240 × 270 mm; and water excitation with *b*-values of 0 and 900 s mm⁻². An average of 24 slices, with a total acquisition time of 29 s was obtained to cover the scrotal area. The orientation and location of these slices were identical to the conventional transverse images. Full echo information was obtained with a bandwidth of 1, 5774 kHz per pixel, a slice thickness of 3–4 mm and an intersection gap of 0.5 mm. No parallel imaging was used. DW sequences were performed during quiet breathing.

MRI data interpretation

MRI data were interpreted by two radiologists (ACT and AN) and any disagreement was resolved by consensus. DW images were read in conjunction with the transverse T2-weighted images. ADC maps were created on a workstation (MxView; Philips Medical Systems, Cleveland, OH, USA), after zooming the image by a factor of 1.5. We recorded signal intensity mean ADC values of circular regions of interest (ROIs), regions as large as possible placed in the middle of the testis and encompassing the majority of testicular parenchyma. Special care was taken to avoid partial-volume effects and subtraction artifacts. Three different ROIs were placed for each testis and the measurements were averaged. The mean and standard deviation (s.d.) of the ADC values were calculated for each testis.

Statistical analysis

The Kolmogorov–Smirnov test was used to assess normality of the data. Pearson's correlation coefficient (*r*) and linear regression were used to determine the association between age and ADC values. Subjects were classified into three groups according to their age: group 1, young men aged 20–39 years; group 2, middle-aged men 40–69 years and group 3, men 70 years and older. One-way analysis of variance (ANOVA) was

used to determine whether mean ADC differed among age groups. Post hoc analysis (Dunnett T3) was applied to reveal differences in ADC between age groups. Statistical analysis was performed using SPSS version 20.0 (IBM, Inc., Armonk, NY, USA) and reviewed by a biostatistician. In all cases, a *P* < 0.05 was considered statistically significant.

RESULTS

Fifty-six testes in 56 men were not included in data measurement, because of the presence of intratesticular mass lesions, including 28 malignancies, 26 benign lesions and two cases with no histologic confirmation (patients lost to follow-up). In one of these cases, measurements of the ADC of the contralateral testis were not possible, because of its small size and high position. The final diagnoses are presented in **Table 2**.

In 46 patients, both testes (*n* = 92) were characterized as normal; 42 of these patients had testes of normal size and position within the scrotum and four had testes with a high position within the scrotal sac. Two patients had one testis of smaller size than the contralateral testis. In four patients, tubular ectasia of the rete testis was detected; in two of these, the condition was bilateral.

Therefore, 147 (55 and 92) testes from 101 men were evaluated, including 71 testes from 53 men aged 20–39 years (group 1), 67 testes from 42 men aged 40–69 years (group 2) and nine testes from six men older than 70 years (group 3). Testes appearing homogeneously hyperintense on both T2-weighted and DW images and slightly hypointense on the ADC maps and/or with no abnormal intratesticular lesions found during subsequent follow-up study were considered normal. The mean ± s.d. of ADC values (× 10⁻³ mm² s⁻¹) of normal testicular parenchyma were 1.08 ± 0.13 in men aged 20–39 years (group 1; **Figure 1** and **Figure 2**), 1.15 ± 0.15 in men aged 40–69 years (group 2; **Figure 3**) and 1.31 ± 0.22 in men 70 years and older (group 3; **Figure 4**).

The data followed a normal distribution as evaluated using the Kolmogorov–Smirnov test. A positive correlation (*r* = 0.236, *P* < 0.001) between ADC and age was found. Similarly, linear regression analysis revealed a strong age-ADC relationship (*P* < 0.001), indicating that age can be considered a predictor of ADC (**Figure 5a**). ANOVA revealed differences in mean ADC among age groups (*F* = 11.391, degrees of freedom (df) = 2, *P* < 0.001). Post hoc analysis (Dunnett T3) showed

Table 1: Clinical characteristics of the study population

Clinical symptoms	Number
Vague scrotal pain (in 5 cases following recently treated epididymitis/epididymo-orchitis)	27
Painless scrotal enlargement and/or palpable mass	21
Painless scrotal enlargement (in one patient after recent testicular biopsy)	9
Sonographically detected intratesticular mass lesion in asymptomatic patients	19
Sonographically detected intratesticular mass in patient with supraclavicular lymphadenopathy	1
Signs of epididymo-orchitis	11
Subfertility	2
Elevated serum alpha-fetoprotein levels	2
Recent scrotal trauma	1
Partial thrombosis of corpus cavernosum	1
Follow-up after surgery (one case after removal of a large paratesticular hematoma, one patient with surgically removed retroperitoneal germ cell tumor and 3 cases with undescended testis)	5
Multiple hypoechoic intratesticular lesions in a patient with retroperitoneal angiosarcoma	1
Fournier's gangrene	1
Carcinoma of the glans penis	1

Table 2: Diagnoses of intratesticular mass lesions

Diagnosis	Number
Malignant	28
Seminomas	17
Nonseminomatous germ cell tumors	10
Intratesticular metastases	1
Benign	26
Benign Sertoli cell tumor	1
Acute epididymo-orchitis	11
Post-biopsy changes	1
Posttraumatic hematoma	1
Postsurgical changes	1
Testicular fibrosis	2
Hemorrhagic necrosis	1
Hemorrhagic necrosis and atrophy	1
Undescended testis	2
Epidermoid cyst	1
Negative for malignancy	4

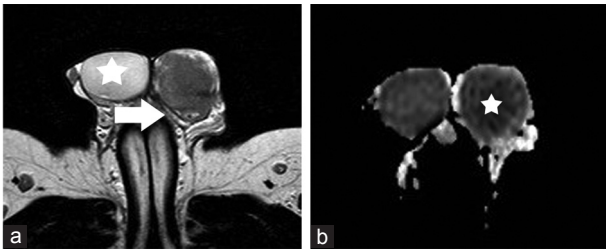


Figure 1: Left testicular seminoma in a 27-year-old man. (a) Transverse T2-weighted image depicts relatively homogeneous left testicular mass, mainly hypointense when compared to the normally hyperintense right testicular parenchyma (asterisk). The neoplasm is seen extending to the paratesticular space (arrow). (b) Transverse ADC map ($b = 900 \text{ s/mm}^2$) shows marked hypointensity of the tumor (asterisk), when compared with the normally hypointense contralateral testis. The ADC values of the right testis were $0.98 \times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$. ADC: apparent diffusion coefficient.

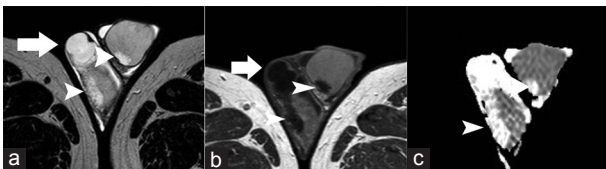


Figure 3: Right spermatocele and bilateral TERT in a 64-year-old man, referred for scrotal enlargement. Imaging findings were typical for the diagnosis of TERT in this case. Sonographic follow-up revealed no change in the lesions. Transverse (a) T2-weighted and (b) post-contrast T1-weighted images show multilocular cystic mass (arrow) of the right paratesticular space, findings suggestive of the presence of a spermatocele. A small hydrocele is seen bilaterally. Both testes appear normal, except for the presence of TERT bilaterally (long arrow), detected as multilocular cystic spaces in the mediastinum testis, hyperintense on T2-weighted images and not enhancing after gadolinium administration. (c) Transverse ADC map ($b = 900 \text{ s/mm}^2$). Cystic ectasia of the rete testis appeared hyperintense on the ADC maps (arrowheads). The ADC values of the normal testicular parenchyma were $1.30 \times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$ (right testis) and $1.32 \times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$ (left testis). ADC: apparent diffusion coefficient; TERT: tubular ectasia of the rete testis.

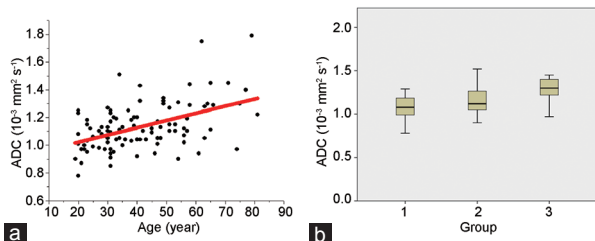


Figure 5: (a) Scatter plot and linear regression fit of ADC vs age. (b) Boxplots of ADC values for each age group. ADC: apparent diffusion coefficient.

differences between groups 1 and 2 ($P = 0.008$) and between groups 1 and 3 ($P = 0.043$), but not between groups 2 and 3 ($P = 0.197$). **Table 3** provides mean ADC values, standard deviations and post hoc analysis results for the age groups. Boxplots in **Figure 5b** depict the ADC distribution for each group.

DISCUSSION

Although ultrasonography is the imaging modality of choice for assessing scrotal lesions, MRI can provide important diagnostic information, especially in cases of inconclusive or nondiagnostic sonographic findings.^{26–39} DWI with evaluation of ADC has been shown to be an important diagnostic tool, providing quantitative information regarding structural tissue changes at a cellular level, which helps in

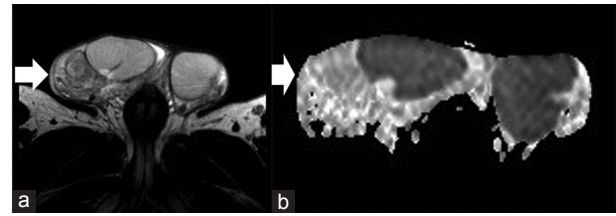


Figure 2: Right epididymo-orchitis in a 31-year-old man. (a) Transverse T2-weighted image depicts enlargement and hypointensity of the right epididymis (arrow). Both testes appear normally hyperintense. (b) Transverse ADC map ($b = 900 \text{ s/mm}^2$) shows hypointensity of both testes. The right epididymis (arrow) was hyperintense on the ADC maps, due to inflammation. The ADC values of the normal left testicular parenchyma were $1.09 \times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$. ADC: apparent diffusion coefficient.

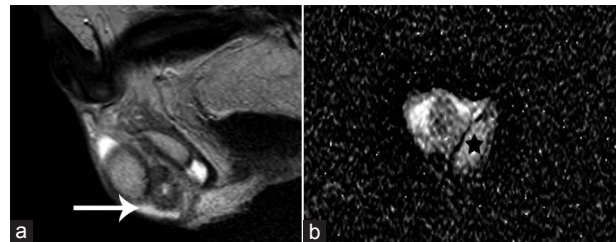


Figure 4: Right epididymo-orchitis in a 79-year-old man. (a) Sagittal T2-weighted image depicts enlargement and hypointensity of the right epididymal tail (arrow). (b) Transverse DW image ($b = 900 \text{ s/mm}^2$). The ADC values of the normal contralateral testis (asterisk) were $1.79 \times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$. ADC: apparent diffusion coefficient; DW: diffusion-weighted.

Table 3: Mean ADC values, standard deviations (s.d.) and post hoc analysis results for the age groups

Group	ADC values ($\times 10^{-3} \text{ mm}^2 \text{ s}^{-1}$)		P value (post hoc)		
	Mean	s.d.	1	2	3
1	1.09	0.14			
2	1.13	0.13	0.45		
3	1.28	0.24	0.01	0.05	

ADC: apparent diffusion coefficient

tissue characterization.^{1–12} There are a few published reports on the clinical applications of DWI in the evaluation of scrotal pathology, including detection and localization of impalpable testes, diagnosis of testicular torsion and differentiation between normal, benign and malignant scrotal contents.^{13–17}

The ADC values of biological tissues are influenced by many factors. The motion of water molecules is restricted by interactions with tissue compartments, cell membranes, intracellular organelles, cytoskeleton and macromolecules.^{1–12} Hyperintensity and hypointensity of normal testicular parenchyma on DW sequences and ADC maps is explained by the complex histology of normal testicular tissue. The presence of densely packed seminiferous tubules lined by a compact fibroelastic connective tissue sheath results in restricted diffusion of water molecules.^{17,31} Another factor that contributes to restricted diffusion is the presence of the interstitial stroma, which fills the spaces between the seminiferous tubules and contains fibroblasts, blood vessels, lymphatics and Leydig cells.^{17,31}

The evaluation of testicular diffusivity usually needs a comparison of the ADC between the affected testis and the contralateral unaffected testis. A standard reference of ADC values of normal testicular parenchyma could be valuable, serving as a baseline when interpreting scrotal pathology. Intersubject differences in the ADC of normal

testicular parenchyma were noted in this report. The magnitude of variations was comparable to report differences between malignant and benign intratesticular lesions.¹⁷ However, a previous study found that the ADC values of intratesticular malignancies were significantly lower than those of normal testicular tissue and benign lesions.¹⁷

An increase in the ADC of normal testicular parenchyma was observed with advancing age. Differences were found between ADC values in young men (group 1), when compared with middle-aged men (group 2, $P = 0.008$) and old men (group 3, $P = 0.043$). No differences were observed when comparing the ADC between groups 2 and 3 ($P = 0.197$). Testicular volume and testicular parenchyma volume decrease with advancing age.^{18–20} These changes mainly involve the seminiferous tubules, which decrease in length and diameter because of loss of both the germ cells and the Sertoli cells. These histologic changes probably account for the increase in ADC with advancing age. Aging also leads to thickening of the tunica propria, increased intertubular connective tissue with peritubular fibrosis, progressive marked hyalinization and atrophy of some of the seminiferous tubules. An increase in the Leydig cell population also occurs.^{18–20} The above changes probably result in the relatively less marked increase in the ADC observed in old men, compared to middle-aged men.

Sonography currently remains the primary method of examination in assessing scrotal lesions.^{26,27} However, imaging of the scrotum has been significantly refined during recent years.^{13–17,28–45} The goal of imaging is to improve the diagnosis and management of men with acute scrotal symptoms or a palpable mass and to reduce the number of unnecessary radical surgical procedures. Contrast-enhanced ultrasonography has been proposed as an alternative modality in cases of inconclusive sonographic findings, and has proven useful in patients with testicular infarction and trauma, in the detection and characterization of testicular masses and when testicular torsion cannot be ruled out.^{40–43} Functional MRI techniques, including DWI, dynamic contrast-enhanced subtracted MRI and MR spectroscopy have added important diagnostic information in the interpretation of scrotal pathology.^{13–17,36,44,45}

There were limitations in this study. First, it was a retrospective review that included only a small number of men older than 70 years. Prospective studies of larger numbers of men of different ages are needed to better define standard references of ADC values of normal testicular parenchyma. Another potential criticism is the lack of histologic confirmation of the so-called 'normal testes' in our report. Finally, only a single evaluation of the MRI data by two radiologists in consensus was performed, therefore interobserver variability was not assessed.

Our study concluded that the ADC values of normal testicular tissue increase with advancing age. A standard reference of normal testicular ADC values is necessary when interpreting normal testis anatomy and pathology on DWI.

AUTHOR CONTRIBUTIONS

ACT conceived of the study, participated in its design, in data analysis and interpretation and helped to draft and revise the manuscript. DG participated in data acquisition, analysis and interpretation. AS participated in data acquisition, analysis and interpretation. AN participated in data acquisition, analysis and interpretation. LGA participated in the design of the study and performed the statistical analysis. NS conceived of the study and helped to draft and revise the manuscript. MIA conceived of the study, participated in its design and helped to draft and revise the manuscript. All authors read and approved the final manuscript.

COMPETING INTERESTS

All authors declare no competing interests.

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