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MAGNETIC RESONANCE

A prospective study on contrast-enhanced magnetic resonance imaging of testicular lesions: distinctive features of Leydig cell tumours

Lucia Manganaro[1] · Valeria Vinci[1] · Carlotta Pozza[2] · Matteo Saldari[1] · Daniele Gianfrilli[2] · Riccardo Pofi[2] · Silvia Bernardo[1] · Vito Cantisani[1] · Andrea Lenzi[2] · Michele Scialpi[3] · Carlo Catalano[1] · Andrea M. Isidori[2]

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Abstract
Objectives Up to 20 % of incidentally found testicular lesions are benign Leydig cell tumours (LCTs). This study evaluates the role of contrast-enhanced magnetic resonance imaging (MRI) in the identification of LCTs in a large prospective cohort study.
Materials and methods We enrolled 44 consecutive patients with at least one solid non-palpable testicular lesion who underwent scrotal MRI. Margins of the lesions, signal intensity and pattern of wash-in and wash-out were analysed by two radiologists. The frequency distribution of malignant and benign MRI features in the different groups was compared by using the chi-squared or Fisher's exact test. Sensitivity, specificity, positive and negative predictive value, and diagnostic accuracy were calculated.
Results The sensitivity of scrotal MRI to diagnose LCTs was 89.47 % with 95.65 % specificity; sensitivity for malignant lesions was 95.65 % with 80.95 % specificity. A markedly hypointense signal on T2-WI, rapid and marked wash-in followed by a prolonged washout were distinctive features significantly associated with LCTs. Malignant lesions were significantly associated with blurred margins, weak hypointense signal on T2-WI ,and weak and progressive wash-in. The overall diagnostic accuracy was 93 %.
Conclusions LCTs have distinctive contrast-enhanced MRI features that allow the differential diagnosis of incidental testicular lesions.
Key Points
- *MRI is able to characterize testicular lesions suggesting a specific diagnosis.*
- *Rapid and marked wash-in is a common feature of Leydig cell tumours.*
- *Markedly hypointense T2-WI signal is significantly correlated with benign lesions.*
- *Blurred margins and weak hypointense T2-WI signal are correlated with malignant tumours.*
- *Weak and progressive wash-in features are present in 85 % of seminomatous lesions.*

Keywords Magnetic resonance imaging · Testicular cancer · Leydig cell tumours · Contrast enhanced MRI · Seminoma

Introduction

Testicular tumours account for 4–6 % of all neoplasms of the male urogenital tract and for 1–2 % of all cancers in young males [1]. A high proportion of small testicular lesions have been proven benign [2, 3]. Leydig cell tumours account for up to 22 % of tumours less than 1.5 cm in size, incidentally found during routine ultrasound (US) [2].

The ability to distinguish benign from malignant non-palpable lesions pre-operatively allows opting for conservative surgery or follow-up in selected cases, with preservation of fertility, endocrine function and relevant savings for health services [4].

✉ Lucia Manganaro
lucia.manganaro@uniroma1.it

[1] Department of Radiology, Sapienza University of Rome, Viale Regina Elena 324, 00161 Rome, Italy

[2] Department of Experimental Medicine, Sapienza University of Rome, Viale del Policlinico 155, 00161 Rome, Italy

[3] Department of Surgical and Biomedical Sciences, Division of Radiology 2, Perugia University, S. Maria della Misericordia Hospital, S. Andrea delle Fratte, 06156 Perugia, Italy

Scrotal US is the preferred modality in the assessment of male reproductive problems increasing the detection of incidentally non-palpable lesions. However, it does not allow a characterization of their nature [5, 6]. A significant improvement in the diagnostic accuracy has been demonstrated by integrating US with contrast-enhanced US (CEUS) [2, 7, 8] and elastosonography [7, 9].

The issue is of particular relevance for monorchid patients and those with bilateral lesions.

Magnetic resonance imaging (MRI) has been recently proposed as an additional diagnostic procedure in the characterization of testicular masses, offering higher sensitivity and specificity than unenhanced US for selected testicular pathologies [10].

The role of MRI in the differential diagnosis of testicular masses, particularly the use of contrast agents, is still controversial. Although dynamic contrast-enhanced subtraction MRI (DCE-MRI) imaging can provide information about testicular perfusion by means of testicular contrast enhancement [11], the clinical value and diagnostic performance of DCE-MRI is not yet established, as was recently demonstrated with CEUS [12].

The aim of this study was to evaluate prospectively the accuracy of qualitative and semi-quantitative contrast-enhanced MRI in the differential diagnosis of non-palpable testicular masses.

Materials and methods

Patients

The local review board approved the protocol, and all patients provided written informed consent. This was a prospective diagnostic accuracy study, conducted at our hospital, between 2006 and June 2012.

All patients included in the CEUS diagnostic study by Isidori et al. [2] with at least one solid, non-palpable testicular lesion, were offered undergoing MRI imaging of the scrotum with gadolinium enhancement. Indications for CEUS are described in Table 1. Only patients with proven histology (reference diagnosis) were considered in the current analysis.

Subjects underwent routine blood tests, hormonal investigations (LH, FSH, testosterone, inhibin B, SHBG, E2) and tumour markers (beta-HCG, alfa-FP, CEA, LDH, PLAP), and were scheduled to undergo scrotal MRI. Thereafter tissue-sparing surgical enucleation was offered for histological confirmation to all patients, independently of the CEUS and MRI results. Malignant tumours were immediately or subsequently treated with radical orchiectomy.

The time interval between CEUS, MRI and surgery was less than 3 weeks. The exclusion criteria were: absence of internal enhancement after contrast administration on MRI and absence of histological diagnosis. None had cardiac insufficiency.

Table 1 Reason for referral for scrotal ultrasound and MRI

Reason for scrotal ultrasound and MRI	No. (%) of patients, n=44 n=115
Infertility	26 (59.1)
Andrological screening/other	6 (13.6)
Testicular pain	4 (9.1)
Previous orchiectomy for contralateral tumour[a]	6 (13.6)
Klinefelter syndrome	1 (2.3)
Hypogonadism	1 (2.3)

[a] Six patients had previous surgery of the testis for testicular tumour; this was the first scan for the remaining 38 patients

MRI protocol

Patients did not require any preparation, except measuremnet of serum creatinine prior to administration of intravenous medium contrast. All patients were examined in the supine position. The penis was draped to the anterior abdominal wall. Testes were placed at a similar distance, with a towel placed between them. All the examinations were performed on a 1.5 T system (Siemens Magnetom Avanto, Erlangen, Germany), with the use of a phased array multichannel (32 channels) body coil. The MRI protocol included: orientation sequences: T2-weighted (T2-W) HASTE (Half Fourier Single Shot Turbo Spin Echo) (slice thickness (SL)=6 mm; repetition time (TR)=1000 msec; echo time (TE)=85 msec; flip angle (FA)=150); morphological sequences: T2-W Turbo Spin Echo High resolution sequences (SL=3.00 mm; TR =3659-5740 msec; TE=95-110 msec; FA=40-150; field of view (FOV)=240 x 240; Matrix=256 x 224); T1-weighted (T1-W) Flash 2D sequences (Fast low-angle single shot Gradient-Echo) with breath hold, with and without fat saturation (SL=5.5 mm; TR=137–240 msec; TE=5 msec; FA 70; Matrix=256 x 205; FOV=350 x 350); T1-WI in and out of phase (SL=3 mm; TR=125 msec; TE=2-5 msec; FA=70; Matrix=256 x 243; FOV=450 x 450).

A medium contrast agent (Gadobenate Dimeglumine, Multihance Bracco 0,1 mL/kg) was administered intravenously; we applied VIBE (volumetric interpolated breath-hold examination) sequences (SL=4 mm; FA=12; FOV=280 x 280; Matrix=256 x 224; TR=4 msec; TE=2 msec) in a dynamic protocol acquiring a pre-contrast scan followed by a post-contrastoghraphic series of six consecutives phases of 20 s each. Finally we completed the study with the elaboration of pre- and post-contrast sequences thus obtaining subtracted images. Examination was completed with a coronal plane scan of the abdomen in order to identify associated

pathologies. The total acquisition time of the examination was 30 min.

Image analysis

Radiologists used the Sony LMD-2451 MD monitor (resolution 1220 x 1920 pixels) for the prospective reading of mass lesion characteristics. MRI image interpretation was performed by two radiologists with more than 15 years and 5 years of experience in genitourinary imaging, respectively, blinded to the surgical or histopathological results, and discrepancies were resolved by consensus. Lesions were characterized on the basis of pre-contrast visualization of the lesion, wash-in and wash-out at semi-quantitative analysis, and pattern of contrast-enhancement.

Signal intensity with respect to normal testicular parenchyma on T2-weighted images (T2-WI) (markedly hypointense, slightly hypointense, isointense and slightly hyperintense), signal intensity on phase T1-weighted images (T1-WI) (hypointense, isointense, slightly hyperintense), and margins of the lesions (well-defined, blurred or irregular) were evaluated.

The time-signal intensity curve (TIC) from contrast-enhanced MRI was obtained by manually placing a region of interest (ROI) within the lesions with care to exclude areas of haemorrhage and necrosis and with the aid of corresponding T1-WI and T2-WI; an identical ROI was placed on the adjacent parenchyma.

According to the shape of the TICs, the wash-in pattern was categorized as rapid or marked (enhancement peak within 1 min after agent injection), reflecting a high degree of vascularisation, and weak and progressive (when the previous condition did not occur).

The wash-out pattern was divided into rapid (decline abrupt of the signal intensity), slow and delayed (progressive decline of signal intensity), and absent (persistent or progressive enhancement, with continuous increase in signal intensity).

Contrast enhancement was classified as homogeneous, non-homogeneous or rim enhancement.

For each patient, on T2-WI the observers subjectively scored the degree of lesion-to-testis contrast according to the following five-point scale: 1=signal intensity of lesion is lower than those of surrounding normal testis; 2=signal intensity of lesion is equal to those of surrounding normal testis; 3=signal intensity of lesion is slightly higher than those of surrounding normal testis; 4=signal intensity of lesion is noticeably higher than those of surrounding normal testis; 5=signal intensity of lesion is markedly higher than those of surrounding normal testis. The mean visual scores for lesion-to-testis contrast was then calculated and compared.

Statistical analysis

Groups were compared using odds ratio (OR) confidence intervals for categorical variables. The frequency distribution of malignant and benign MRI features in the different group was compared by using the chi-squared or Fisher's exact test. Subgroup analysis was performed to limit the comparison to the largest groups of histotypes. Sensitivity, specificity and diagnostic accuracy of MRI findings in the assessment of benign and malignant testicular lesions were calculated.

Interobserver agreement was analysed by means of Kappa statistics. A positive correlation was considered to be indicated by a k value greater than 0, poor correlation by values of 0.00–0.01, low correlation by values of 0.01–0.20, moderate correlation by values of 0.21–0.40, good correlation by values of 0.41–0.60, substantial correlation by values of 0.61–0.80, and almost perfect agreement by values greater than 0.81 [13].

Results

Clinical data

Of the 115 patients who presented a non-palpable testicular lesion, 69 agreed to undergo a gadolinium-enhanced scrotal MRI. Of these, 25 patients were excluded because they showed no internal enhancement at MRI (6/69) or refused to undergo surgery (19/69) and thus no definite histology was available [2]. Therefore the final study group consisted of 44 patients (Fig. 1). The reason for referral is reported in Table 1.

Twenty-three patients had malignant tumours (20 seminomas, two embryonal carcinomas and one mixed germ cell tumour) and 21 had benign tumours (19 Leydig cell tumours and two Sertoli cell tumours). All 44 subjects underwent enucleation followed by radical orchiectomy in all 23 malignancies. The malignancies tended to be larger than benign tumours (mean diameter 0.9±0.31 cm vs. 0.6±0.28 cm, respectively, $p<0.001$;Table 2). Full hormonal data were available for 20 of the 44 patients. Of these, 34 % of the patients had a FSH level of more than 7 mIU/mL (16.8±17.0, normal range 1.38–9.58), 13 % had a decreased testosterone level (4.4±1.6 ng/mL, normal range 2.8–11) and 20.5 % had an elevated LH level (8.5±8.4 mIU/mL, normal range 1.8–8.16). Inhibin B, SHBG, E2 and tumour markers were normal at baseline in all 44 patients.

MRI qualitative results

Most of the histologically proven benign lesions appeared hypointense on T2-WI compared to malignant lesions (17/21, 81 %, vs. 8/23, 34.8 %, $\chi2$ 9.537, $p=0.03$). The remaining benign lesions appeared slightly hypointense (14.3 % vs. 52.2 %, $\chi2$ 7.013, $p=0.011$) and only in one case hyperintense

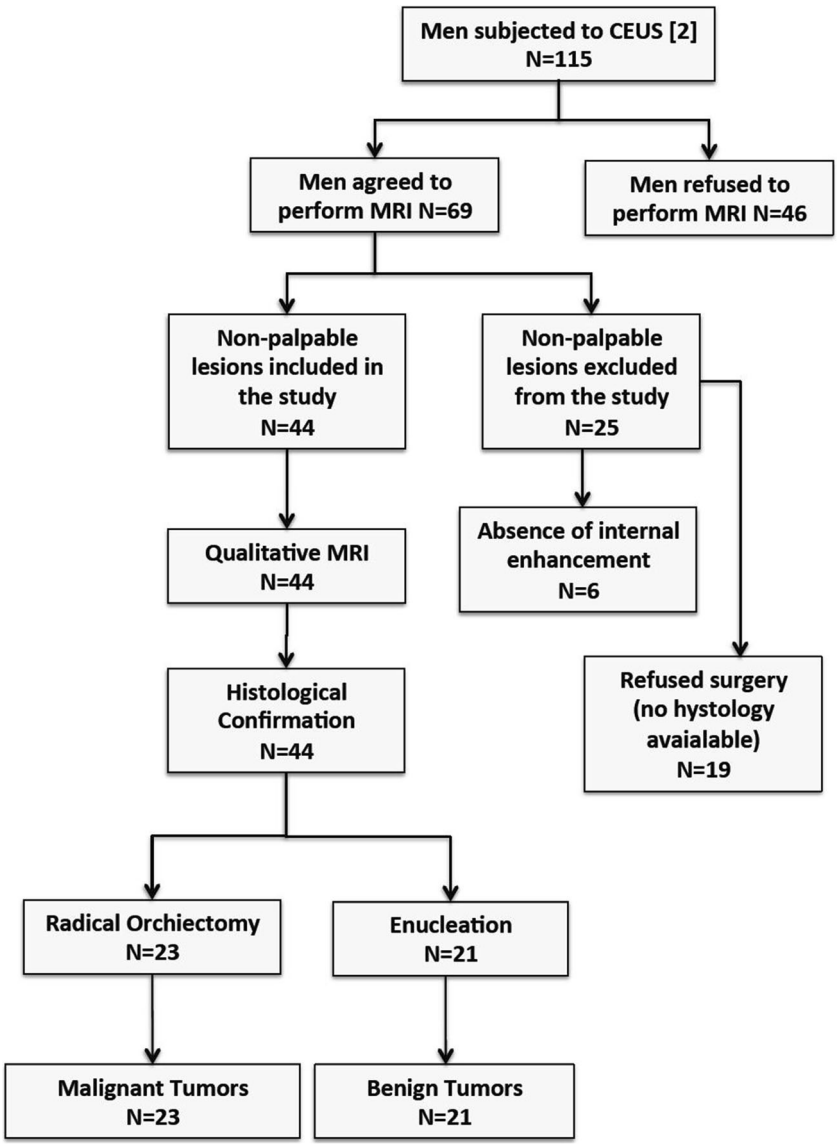

Fig. 1 STARD (Standards for Reporting of Diagnostic Accuracy) flow diagram of the study [2]

(one case of benign vs. one case of malignant lesions, $\chi 2$ 0.004, p=N.S.). Well-defined margins were observed in 14 of the 21 benign lesions (66.7 %) compared with five of the 23 malignant lesions (21.7 %) ($\chi 2$ 9.031, p=0.003). Blurred

Table 2 Clinical data (mean± standard deviation) of all patients and lesions according to the reference diagnosis

				Testicular volume (mL)	
Final diagnosis	N	Age (y)	Diameter (cm)	R	L
All patients	44	34.45±8.97	0.8±0.34	12.6±5.05	12.6±5.93
Malignant tumours	23	33.96±5.13	0.9±0.31*	12.9±5.43	12.6±5.68
Pure seminoma	20	33.57±5.84	1.0±0.32	10.50±4.65	12.32±6.29
Embryonal carcinoma	2	29±4.24	1.2±0.28	19.2±3.18	13.7±2.61
Mixed germ cell tumour	1	38	1.0	19.5	5.4
Benign tumours	21	35±11.98	0.6±0.28	12.3±4.89	12.6±6.30
Leydig cell tumour	19	36.0±13.91	0.6±0.26	12.9±4.95	13.7±6.32
Sertoli cell tumouur	2	32.6±5.50	0.4	11.9±2.96	9.7±3.25

* p<0.001 malignant tumours vs. benign tumours

R right testis, *L* left testis

margins were more typical of malignant lesions (14/23, 61 % vs. 5/21, 23.8, $\chi 2$ 6.145, p=0.013).

A homogeneous enhancement was observed predominantly in benign lesions (18/21, 85.7 % vs. 7/23, 30.4 %, $\chi 2$ 13.672, p<0.001), whereas rim enhancement was present only in malignant tumours (4/23 vs. 0/21, $\chi 2$ 4.017, p=0.109).

MRI semi-quantitative results

Benign lesions (Fig. 2) had a more rapid and intense wash-in (18/21, 85.7 %, vs. 5/23, 21.7 %, $\chi 2$ 18.008, p <0.001) and a prolonged wash-out (15/21, 71.4 % vs. 0/23, $\chi 2$ 24.926, p< 0.001), whereas malignant tumours (Figs. 3 and 4) showed a weak and progressive wash-in (18/23, 78.3 % vs. 3/21, $\chi 2$ 18.008, p<0.001) and an absent wash-out (21/23, 91.3 % vs. 6/21, 28.6 %, $\chi 2$ 18.221, p<0.001) (Tables 3 and 4). Subgroup comparisons of MRI features were also performed for the two most numerically representative histotypes: Leydig cell tumours (LCTs, 19 cases) and seminomas (20 cases). The results are summarized in Tables 3 and 4.

Comparison between MRI and histology

MRI findings led to a correct pre-operative characterization in 86 % of tumours (39 of 44 cases): 17/21 of the benign and 22/23 of the malignant tumours. Of 18 lesions (41 %, 18/44) diagnosed as benign at MRI, 17 were found to be benign and one malignant at final histology. Of the 26 lesions (59 %, 26/44) initially diagnosed as malignant at MRI, 22 were confirmed while four lesions turned out to be benign at final histology.

The sensitivity of scrotal MRI in diagnosing Leydig cell tumour was 89.47 % (95 % CI: 66.82– 98.39), and the specificity 95.65 % (95 % CI: 77.98–99.27, Table 5).

The sensitivity of scrotal MRI in diagnosing malignant lesions was 95.65 % (95 % CI: 77.98–99.27), and the specificity 80.95 % (95 % CI: 58.08–94.44). The diagnostic accuracy was 93 %.

The Kappa value for the interobserver agreement obtained from the qualitative analysis was 0.854 (p<0.001, CI 95 %), characterizing a very good agreement between observers.

Discussion

Our study prospectively evaluated the accuracy of qualitative and semi-quantitative contrast-enhanced MRI in the differential diagnosis of non-palpable testicular masses. For the first time, we described that in a large prospective series of patients, Leydig cell tumour of the testis presents distinct MRI features that enable an accurate pre-operative diagnosis in 93 % of the cases. Furthermore, our study provides a qualitative and semi-quantitative analytical description of gadolinium contrast kinetics in benign and malignant tumours, demonstrating typical features of Leydig cell tumours and further improving the differential diagnosis with seminomas.

US is considered the gold standard in the evaluation of scrotal pathology. Occasionally, during a routine US examination performed for other reasons, non-palpable testicular

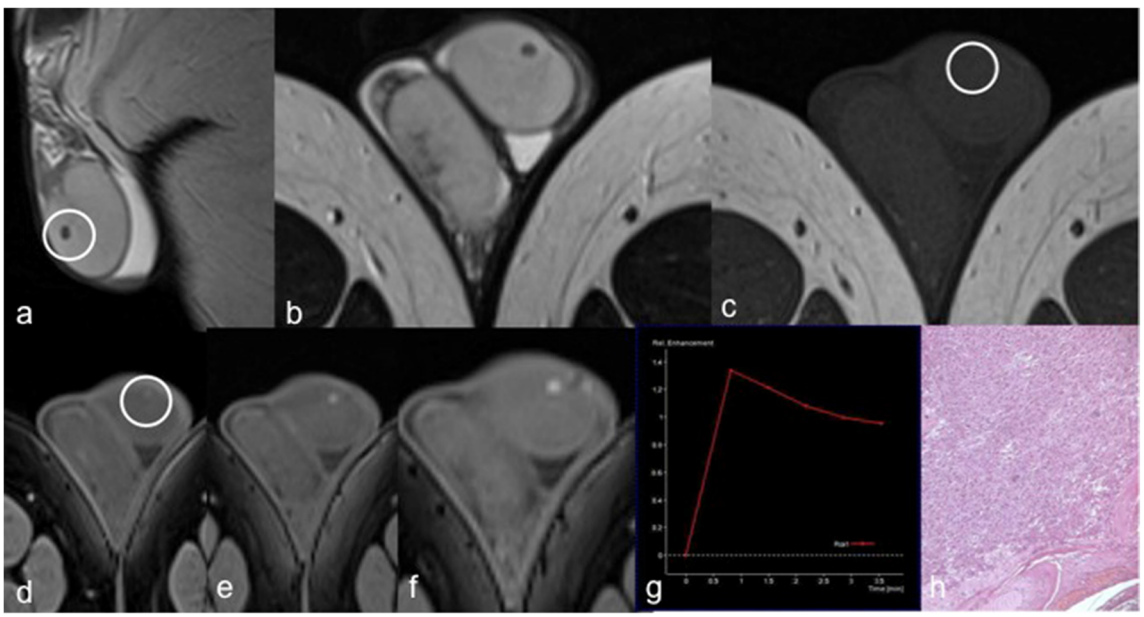

Fig. 2 Leydig cell tumour in the medium part of the left testes in a 38-year-old man. T2 WI on the sagittal (**a**) and axial (**b**) planes: the lesion is well defined and markedly hypointense. Isointense on T1 WI (**c**). VIBE (volumetric interpolated breath-hold examination) subtracted T1 WI after contrast administration (**d**, **e**, **f**) shows a rapid and marked wash-in. The intensity-time curve (**g**) shows a peak within the first minute. Histological examination of the lesion (10x) with homogeneous tumour cells arranged in a row is shown in (**h**)

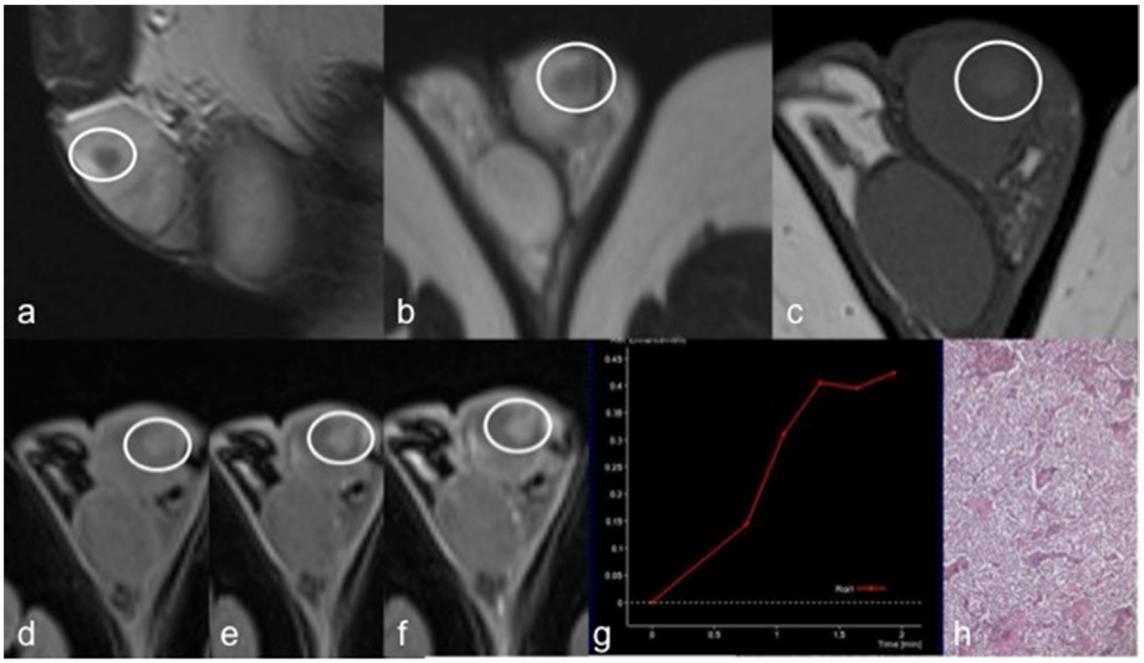

Fig. 3 Seminomatous lesion in the superior part of the left testes of a 35-year-old man. T2 WI on the sagittal (**a**) and axial (**b**) planes: the lesion appears slightly hypointense and with feathered margins. T1 WI on the axial plane shows a weak hyperintensity of the lesion (**c**). VIBE (volumetric interpolated breath-hold examination) subtracted T1 WI shows a weak and progressive enhancement (**d**, **e**, **f**), which is well visible on the intensity-time curve (**g**). Histological examination (10x): the neoplasm contains abundant infiltration of lymphocytes (**f**)

lesions are identified; among these, approximately 10 % are tumours, half of which are malignant [14–16].

US is able to identify a scrotal mass and to differentiate its origin (intra- or extratesticular) with a sensitivity range between 98 % and 100 % [17–19]. However, this imaging modality is considered insufficient in discriminating benign and malignant testicular neoplasms due to their frequent overlying appearance, particularly in case of smallest lesions.

Isidori et al. demonstrated that in experienced centres, CEUS can improve the diagnostic accuracy of unenhanced US in the differential diagnosis of testicular non-palpable lesions [2, 20, 21]. Similarly, the possibility of using MRI as the third level imaging in the study of testicular masses has been investigated, in particular if they are of difficult characterization [22].

Cramer et al. [23] demonstrated the value of MRI in the detection of testicular tumours; moreover, Adham et al. [24] found that it may be helpful in the histological characterization of testicular tumours. In addition, Schultz-Lampel et al. [25], evaluating 88 patients with clinical suspicion of testicular

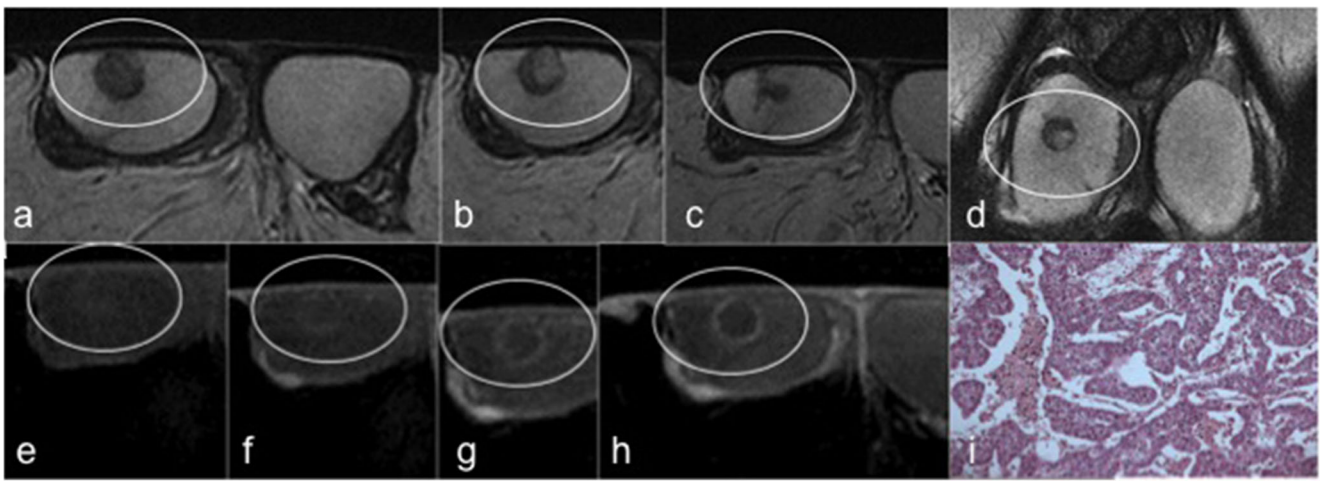

Fig. 4 Embryo cancer on the right testes of a 30-year-old man. (**a**, **b**, **c**) T2 WI. On a different axial plane, the lesion is non-homogeneous with a hypo-isointense rounded area. Four consecutive scans of post-contrast VIBE (volumetric interpolated breath-hold examination) sequences (**e**, **f**, **g**, **h**) show the weak contrast enhancement only seen on the peripheral margin. Histological examination (10x) with atypical cells and necrotic areas (**i**)

Table 3 Frequencies of qualitative features MRI. Data are presented as percentages (number)

Modality	Feature		Benign tumour N=21	Malignant tumour N=23	p-value	Leydig cell tumour N=19	Seminomas N=20	p
MRI	T2-W	Markedly hypointense	81 (17)	34.8 (8)	0.003	89.4 (17)	35 (7)	0.002
		Slightly hypointense	14.3 (3)	52.2 (12)	0.012	5.3 (1)	60 (12)	0.003
		Hyperintense	4.8 (1)	4.3 (1)	0.948	5.3 (1)	5.0 (1)	0.055
		Isointense	0 (0)	8.7 (2)	/	/	/	/
	T1-W	Slightly hyperintense	23.8 (5)	56.5 (13)	0.032	26.3 (5)	60 (12)	0.038
		Isointense	76.2 (16)	43.5 (10)	0.032	73.7 (14)	40 (8)	0.038
	Margins	Well defined	66.7 (14)	21.7 (5)	0.004	68.4 (13)	10 (2)	0.001
		Blurred	23.8 (5)	60.9 (14)	0.016	26.3 (5)	70 (14)	0.009
		Irregular	9.5 (2)	17.4 (4)	0.454	5.3 (1)	20 (4)	0.198
DCE-MRI	Enhancement	Homogeneous	85.7 (18)	30.4 (7)	0.001	94.7 (18)	30 (6)	0.001
		Non-homogeneous	14.3 (3)	52.2 (12)	0.012	5.3 (1)	60 (12)	0.003
		Rim enhancement	0 (0)	17.4 (4)	/	0 (0)	10 (2)	/
	Wash-in	Rapid and intense	85.7 (18)	21.7 (5)	<0.001	94.7 (18)	15 (3)	<0.001
		Weak and progressive	14.3 (3)	78.3 (18)	<0.001	5.3 (1)	85 (17)	<0.001
	Wash-out	Slow and delayed	71.4 (15)	0 (0)	/	78.9 (15)	0 (0)	/
		Rapid	0 (0)	8.7 (2)	/	0 (0)	10 (2)	/
		Absent	28.6 (6)	91.3 (21)	<0.001	21.1 (4)	90 (18)	<0.001

cancer, reported that MRI was able to correctly characterize all cases, both malignant and benign, with the limitation, in that particular series, that nearly all benign lesions were attributable to paratesticular masses.

Tsili et al. [26] highlighted the parameters that mostly oriented towards characterization such as margins, signal intensity of T2-WI and, in a less significant way, of T1-WI, especially for lesions that are not definable as seminomas.

Table 4 Diagnostic performance of MRI in the differential diagnosis of testicular lesions

Modality		Feature	Benign vs. malignant tumours		LCTs vs. seminomas	
			χ^2	p-value	χ^2	
MRI	T2-W	Markedly hypointense	9.537	0.003[a]	12.216	0.001[a]
		Slightly hypointense	7.013	0.011[a]	13.137	<0.001[a]
		Hyperintense	0.004	1.000[a]	0.001	1.000[a]
		Isointense	1.913	0.489[a]	/	/
	T1-W	Slightly hyperintense	4.859	0.027	4.496	0.034
		Isointense	4.859	0.027	4.496	0.034
	Margins	Well defined	9.031	0.003	14.050	<0.001[a]
		Blurred	6.145	0.013	7.442	0.006
		Irregular	0.577	0.666	1.893	0.342[a]
DCE-MRI	Enhancement	Homogeneous	13.672	<0.001	17.252	<0.001
		Non-homogeneous	7.013	0.011	13.137	<0.001[a]
		Rim enhancement	4.017	0.109	2.003	0.487[a]
	Wash-in	Rapid and intense	18.008	<0.001	24.927	<0.001
		Weak and progressive	18.008	<0.001	24.927	<0.001
	Wash-out	Slow and delayed	24.926	<0.001	25.658	<0.001[a]
		Rapid	1.913	0.489	2.003	0.487[a]
		Absent	18.221	<0.001	18.837	<0.001[a]

[a] Fisher exact test

LCTs Leydig cell tumours

Table 5 Diagnostic performance of MRI in diagnosing Leydig cell tumour

	Diagnostic performance of MRI, % (95 %CI)	
	Diagnosing Leydig cell tumours	Diagnosing malignant lesions
Sensitivity	89.47 (66.82–98.39)	95.65 (77.98–99.27)
Specificity	95.65 (77.98–99.27)	80.95 (58.08–94.44)

Diffusion-weighted imaging (DWI) and apparent diffusion coefficient (ADC) values are potentially valuable in the differentiation between normal, benign and malignant scrotal lesions, allowing complementary information for morphological and contrast-enhanced imaging. Qualitative evaluation of DWI may also be helpful, even if the quantitative ADC values are not useful.

According to others [27, 28], in our experience the wide individual variation of ADC measurements and the difficulty in suggesting the ADC cut-off value as an effective parameter for tissue characterization limited the use of DWI in the evaluation of testicular lesions. Our data confirm that a markedly hypointense signal on T2-WI is significantly associated with a benign nature of the lesion, and this feature may be suggestive of a Leydig cell tumour (80 % of all benign and 90 % of Leydig cell tumours included in our series). Furthermore, a typical pattern of contrast enhancement, characterized by homogeneous impregnation ($p=0.001$) and rapid and marked wash-in ($p<0.001$), was found in 94.7 % of Leydig cell tumours, accompanied by a slow and late wash-out ($p<0.001$). This behaviour, with the appropriate distinction, is similar to what was observed in contrast-enhanced US [2], and has been attributed to a high degree of vascularization of Leydig cell tumours, local estrogen generation and an increased level of endocrine-gland vascular endothelial growth factor (EG-VEGF) expressed by Leydig cells [29–31]. Malignant lesions were significantly associated with blurred margins ($p=0.013$) and a weak hypointense signal on T2-WI ($p=0.011$). Seminomas showed this feature in 70 % ($p=0.009$) and 60 % ($p=0.003$) of the cases, respectively. Another feature found in this study is represented by a weak signal hyperintensity on T1 in 60 % of cases of seminoma ($p=0.038$). Furthermore, seminomatous lesions showed weak and progressive wash-in ($p<0.001$) in 85 % of cases, with an absent wash-out ($p<0.001$). Our pattern differs from those reported in other studies in which seminomas are characterized by an enhancement with a rapid increase of signal intensity followed by gradual washout of the contrast medium [12]. The Sertoli cell tumours in our series showed a DCE-MRI behaviour similar to seminomas, and therefore were erroneously considered malignancies. Because of their rarity, a conclusive statement cannot be made; however, recent data suggest that Sertoli cell tumours do not express EG-VEGF to the high level seen in LCTs [29]. An important finding of this study was the very high frequency of Leydig cell tumours. This supports the hypothesis that Leydig cell dysfunction has a role in testicular dysgenesis syndrome [28] and it is sustained by the finding of subclinical hormonal failure (high LH or reduced testosterone) found in 20.5 % and 13 % of our patients, respectively [32].

Our study has some limitations: first, our series included patients with small (<1.5 cm) non-palpable lesions evaluated by CEUS in a previous study [2], which already, and independently of MRI findings, improved the differential diagnosis and the individualized management of small testicular lesions. Despite these results, MRI was performed blinded to CEUS results to evaluate prospectively the accuracy of qualitative and semi-quantitative contrast-enhanced MRI in the differential diagnosis of non-palpable testicular masses. Secondly, the semi-quantitative analysis, which depends on the placement of the ROI, could be affected by the small size of the tumour. Third, we can't ignore that the case mix may have affected the sensitivity (one false-negative case) and the specificity (four false-positive cases) of MRI. We considered four benign tumours to be malignant (two Sertoli cell tumours and two LCTs) because they showed two features that we attributed to malignant neoplasms: weak and progressive wash-in, absent wash-out and a slight hypointense signal on T2-weighted images. Nevertheless, to our knowledge the present series is the largest prospective collection of MRI imaging of Leydig cell tumours. The only false-negative case of the study (one embryonal carcinoma) was misdiagnosed because of its well-defined margins and rim enhancement, which had made radiologists lean towards the nature of an active inflammation, supported by the clinical history of the patient who reported fever, elevated white cell count, pain in the scrotal region and negative markers. In our sample, both cases of embryonal carcinomas showed a pattern of rim enhancement, requiring a differential diagnosis toward inflammatory lesions. Even if this feature cannot be standardized, it may find an explanation in the aggressive nature of this tumour, which is often characterized by areas of haemorrhage and necrosis, producing a central non-enhancing

area; moreover, in agreement with our data [33, 34], even at CEUS, embryonal carcinomas were characterized by poor or absent internal contrast enhancement [15].

In conclusion, the results achieved with our protocol (further improvements can be obtained by use of specified perfusion sequences and 3D high resolution T2 sequences), demonstrated that MRI might be proposed in addition to unenhanced US and CEUS when it is necessary to decide the best therapeutic procedure for a testicular mass not yet characterized. Our study demonstrated that MRI is able not only to characterize benign and malignant testicular lesions, with a diagnostic accuracy of 93 %, but also suggests a specific diagnostic hypothesis when specific imaging features are present. Well-defined margins, a markedly hypointense signal on T2-WI, and a rapid and marked wash-in followed by a slow and late wash-out, should orient toward the diagnosis of Leydig cell tumour. On the other hand, blurred margins, a slightly hypointense signal on T2-WI and a slightly hyperintense signal on T1-WI, associated with a weak and progressive wash-in followed by an absent wash-out, are highly specific for the diagnosis of seminoma.

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Methodology: prospective, diagnostic study, performed at one institution.

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