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ULTRASONOGRAPHY OF THE EXTERNAL REPRODUCTIVE ORGANS IN TOM

Mokhamad Fakhrul Ulum[1]*, Anizza Dyah Kartika Maharani[1], Rizal Eko Kurniawan[1], Arlita Sariningrum[1], Dilla Frastantie[2], Erwin[3], Ligaya ITA Tumbelaka[1], and Deni Noviana[1]
[1]Department of Clinic, Reproduction, and Pathology, Faculty of Veterinary Medicine, Bogor Agricultural University
[2]Program of Reproductive Biology, Postgraduate School, Bogor Agricultural University
[3]Laboratory of Clinic and Surgery, Faculty of Veterinary Medicine, Syiah Kuala University
*Corresponding author: ulum@ipb.ac.id

ABSTRACT

This study aims to delineate the tissue structure of male cat external reproductive organs using ultrasound with different types of consoles and transducers frequencies through indirect method. External organ of 5 tom weighing 2-3 kg was evaluated using ultrasound with stationary console (linear transducer, 10 MHz) and portable console (linear transducer, 6.5 MHz), transcutaneously. The results showed that both console and transducer could not visualize the structure of organs sized less than 10 mm through direct method. However, the indirect method using stationary console with a 10 MHz transducer enable to visualize the organ and depicts clearly the internal organ structure such as penis and its parts, scrotum and its constituent layers, caput and cauda of epididymis, and testicles. Furthermore, portable console with 6.5 MHz transducer was still able to provide an adequate image of those organs. In general, fibrous tissues such as tunica vaginalis, tunica Dartos, funiculus spermaticus, and urethra were visible in ultrasonogram as hyperechoic structures, while soft tissues such as the parts of testicles and penis were visible as hypoechoic structures. Tissue containing fluid such as urethral lumen was visible as anechoic structure. In conclusion, indirect ultrasound imaging method was sufficient to visualize the organs sized less than 10 mm using 3-15 MHz transducer in which was currently widely used in animal health care clinics.

Key words: different frequency, external reproductive organs, imaging methods, tom, ultrasound

ABSTRAK

Penelitian ini bertujuan mencitrakan struktur jaringan organ reproduksi eksternal kucing jantan secara ultrasonografi dengan unit konsole dan frekuensi transduser berbeda dengan metode tidak langsung. Organ eksternal kucing jantan dewasa dengan berat badan 2-3 kg sebanyak 5 ekor dicitrakan menggunakan ultrasonografi konsole stasioner (transduser linier, 10 MHz) dan konsole portable (transduser linier, 6.5 MHz) secara transkutaneus. Hasil pencitraan dari kedua konsole dan frekuensi transduser yang digunakan menunjukkan bahwa metode langsung tidak dapat mencitrakan organ berukuran < 10 mm berikut bagian-bagian dari struktur organ. Metode tidak langsung dengan konsole stasioner dengan transduser frekuensi 10 MHz mampu mencitrakan dengan sangat baik organ berukuran < 10 mm serta bagian-bagian dari organ seperti penis dan bagian strukturnya, skrotum dan lapisan penyusunnya, caput epididimis, testikel dan cauda epididimis. Konsole portable dengan tranduser frekuensi 6.5 MHz dinilai masih mampu memberikan citra yang cukup baik pada organ tersebut. Secara umum, citra ultrasonografi jaringan fibrosa seperti tunika vaginalis, tunika Dartos, funikulus spermatikus, dan uretra tampak hiperekhoik. Jaringan lunak seperti testikel dan bagian-bagian penis tampak hipoekhoik. Jaringan yang berisi cairan seperti lumen uretra tampakan ekhoik. Metode pencitraan ultrasonografi tidak langsung mudah dilakukan untuk mencitrakan organ berukuran < 10 mm menggunakan unit konsole dan transduser frekuensi antara 3-15 MHz yang banyak digunakan di klinik layanan kesehatan hewan saat ini.

Kata kunci: beda frekuensi, organ reproduksi eksternal, metode pencitraan, kucing jantan, ultrasonografi

INTRODUCTION

Ultrasonography could visualize structure the architectural details of soft tissues and organs. The image produced in sonogram is created using high-frequency sound wave and is displayed as real-time slices of images similar to histological images (Pavlin and Foster, 1998). High-frequency sound wave of 20 MHz is commonly used in the medical services for dermatological examination (Gniadecka and Quistorff, 1996). This method known as dermatosonography (Jemec *et al.*, 2000) while 30-100 MHz was used for eye examination (Pavlin and Foster, 1998). However, such high-frequency transducer is not available in most medical facilities. The most commonly available transducer in medical facilities are 3-15 MHz transducers (Mantis, 2008) that used for obstetrical examination and imaging of other internal organs.

The sound wave produced and captured by transducers consists of several parts namely Fresnel zone, focal zone and Fraunhofer zone (Noviana *et al.*, 2012; Mattoon and Nyland, 2014). Target organ examination is performed at focal zone to obtain an image with adequate resolution. However, the limitation of 3-15 MHz transducers is the location of Fresnel zone more than 10 mm, hence the target organ could not be depicted clearly. Ultrasonography examination of scrotum in humans (Hebert *et al.*, 2012) and large animals (Constantinescu, 2005) with a scrotal size of > 30 mm is easy because most of the organ is located at the focal zone (Mantis, 2008). However, difficulty arises when examining small animals and pets that owing small sized of organs (Dyce *et al.*, 2009). Although ultrasonography has been introduced since 1978 (O'Grady *et al.*, 1978), reports on normal and abnormal image of small mammals with tissue width < 20 mm are rare (Redrobe, 2001).The health status of small mammals in zoos or their habitat is very important for its survival (Carey and Johnson, 1995), hence there is a need for new approaches on the application of ultrasonography imaging.

Internal organs sized < 10 mm such as ovaries (Davidson and Baker, 2009a), testicles and penis (Davidson and Baker, 2009b) and their parts are

difficult to be visualized through ultrasonography. Reproductive organs examination for pets and wild animals inside conservation areas are important to monitor their health status and to diagnose certain abnormalities. Although there has been reports of ultrasonography imaging on the reproductive organs of male dogs (Davidson and Baker, 2009b) and thin tail sheep (Ulum *et al.*, 2013), there has been no detailed report on external reproductive organs of toms. New ultrasonography method is needed to visualize tissues and organs sized < 10 mm using the currently available diagnostic equipment. This study reports the result of ultrasonography on external reproductive organs of tom which included scrotum, testicles, penis and prepuce, using consoles equipped with transducers that having difference frequencies through indirect imaging method. This method could be used as a model for ultrasonography of small mammals with tissue width of < 20 mm. The result of this study would improve diagnostic imaging not limited on pets and small mammals but also for conserved animals such as reptiles, amphibians, fishes and other small exotic animals.

MATERIALS AND METHODS

Research Animal

The animals used in this study were 5 clinically healthy tom aged 1-2 years old weighing 2-3 kg with scrotal circumference was 6.0-7.5 cm and scrotal diameter ±10 mm. Uncooperative tom was anesthetized to aid the imaging process. As much as 0.025% atropine sulfate at dose of 0.02-0.04 mg/kg body weight was injected subcutaneously 10-15 minutes before anesthetized (Mann *et al.*, 2011). The tom was then anesthetized using 10% ketamine at 10 mg/kg body weight and 2% xylazine at 2 mg/kg body weight intramuscularly (Fossum, 2013). After anesthesia, the tom was laid on their left comfortably on the examination bed. The tom that did not need anesthesia was handled and restrained physically with firmly but comfortably during imaging at their preferred position (laid on its back, recumbent or prone).

Ultrasonography Imaging

Ultrasonography imaging of external reproductive organ of tom was performed using a stationary ultrasonography console unit (Mirror 2, Landwind Medical, China) with 10 MHz linear transducer and a portable ultrasonography console (SonoVet, Meditech Equipment, China) with 6.5 MHz linear transrectal transducer (Table 1). The hairs on the scrotum, prepuce and perineum were not shaved (Figure 1). Ultrasonography gel was applied on the genital area and was flattened so that the hairs, skins, and the spaces between hairs are completely covered by the gel. Transducer was directly pressed on the scrotum transcutaneously from dorsal to ventral through the caudal area. The views used on testicles and penis were oblique, longitudinal, and transverse; then the skin and prepuce were evaluated. This direct ultrasonography method is commonly used nowadays so that the image obtained in the focus area could be seen clearly. Direct imaging method was performed on testicles and epididymis while indirect imaging method was performed on penis, prepuce, and scrotum. Indirect method was performed by placing the imaging target on focus area so that target organ is not directly in contact with the transducer. The obtained image was documented as a video and the next documented image was then analyzed to be further interpreted. The image was then interpreted based on the structures appearance such as black (anechoic), gray (hypoechoic), and white (hyperechoic).

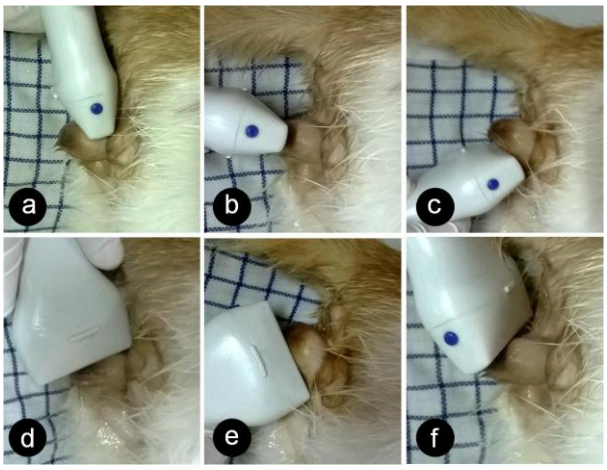

Figure 1. Technique of ultrasonography imaging of the external reproductive organ of tom. a= Transversal view at the base of dorsal scrotum, b= Transversal view at apex of caudal scrotum, c= Transversal view at the base of ventral scrotum, d= Longitudinal view at the base of dorsal scrotum, e= Longitudinal view at the base of ventral scrotum, f= Oblique view at the base of dorsal scrotum

Table 1. Specification of brightness mode (B-mode) ultrasonography console unit

No	Specification	Console	
		Stationary	Portable
1	Frequency (MHz)	10	6.5
2	Transducer	Linear	Linear
3	Gain	45/255	27/44
4	Power supply	AC	DC (Battery)

Data Analysis

The ultrasonographic image of external reproductive organs and its parts was analyzed descriptively.

RESULTS AND DISCUSSION

Reproductive Organs of Tom

The reproductive organs of tom was consist of 4 parts (Constantinescu *et al.*, 2007; Davidson and Baker, 2009b; Dyce *et al.*, 2009). The first part is primary reproductive organs such as testicles, epididymis, vas deferens, spermatic cord and tunica. The second part is accessory glands such as prostatic glands and bulbourethral (Cowper) gland. The third part is penis as the copulatory organ, and lastly urethra which is also part of the urinary system. External reproductive organs

are located outside of the body and it is visible by bare eyes. External reproductive organs in toms include: 1) scrotum, which contains testicles and epididymis, and 2) penis which is enveloped by prepuce (Davidson and Baker, 2009b; Dyce *et al.*, 2009). External reproductive organs of tom are located at the perineum, under the tail and rectum. Penile body is linked to ischiocavernous muscle and penile retractor muscle (Dyce *et al.*, 2009). Scrotal sac emerges from the perineum with penis at its ventral side (Figure 2).

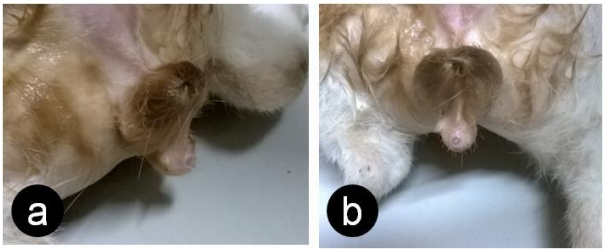

Figure 2. External reproductive organ of tom after given ultrasound gel. a= Side view, b= Back view

Technique of Ultrasonography Imaging

Ultrasonography imaging works by utilizing of high-frequency sound wave, which is divided into 3 zones namely Fresnel zone, focal zone, and Fraunhofer zone (Matton and Nyland, 2014; Noviana *et al.*, 2012). Fresnel zone is where complex diffractions patterns occur, located nearest to the transducer as the sound source. Focal zone is where the sound wave is most focused, producing an image with the best resolution. Fraunhofer zone is the part where sound wave diverges and the resolution of the image gradually decreases (Figure 3a). Ideally, the target organ of ultrasonography is directly positioned in the focus zone by placing the transducer in the imaging area, especially for large organ or dense tissues. Focal zone position can be set to be close to the sound source in the transducer. However, visualizing small organ with direct technique would be difficult because of its small size. Organs sized < 10 mm will be automatically visualized in Fresnel zone, hence the image produced will not have any diagnostic value. Ultrasonography on small organs < 10 mm was done using indirect approach in this study (Figure 3b) to position the target organ in the focus zone. Indirect ultrasonography imaging technique on penis, for example, was done by placing the transducer at dorsal surface of the scrotum to visualize penis which is located at its ventral side (Figure 1a, Figure 1d, and Figure 1f).

Ultrasonography imaging of cat external reproductive organs (testicle, epididymis, scrotum, prepuce, and penis) in this study were done using indirect technique by utilizing console unit and transducer with different frequencies, 10 MHz and 6.5 MHz. The internal structure of organ tissues can be visualized by using ultrasonography imaging. Image produced shows different results as sonogram with difference of echogenicity, especially in detailed basic tissue structure such as testicle, epididymis, scrotum, prepuce, and penis.

Sonogram of Prepuce

Figure 4 shows the sonogram of prepuce (p) which appears hypoechoic compared to the more hyperechoic scrotum (s). Prepuce lumen (lp) appeared as anechoic in contrast to the more hypoechoic prepuce (p), glans penis (gp), and body of penis (bp). Sonogram at 10 MHz transducer could delineate and distinguish prepuce more clearly compared to 6.5 MHz transducer. Scrotal skin (k) located near the transducer could not be imaged clearly while the components of the parts inside the focus area could be delineated clearly.

Prepuce is a skin that acts as cover of glans penis (Dyce *et al.*, 2009). The thickness of prepuce is < 2 mm, hence indirect imaging is the technique of choice. In ultrasonography, prepuce appeared hypoechoic compared to the hyperechoic scrotum because prepuce is only composed of tubular sheets of skin. This skin consists of internal and external lamina. Prepuce lumen appeared as an anechoic structure between the hypoechoic prepuce skin tissues, glans penis, and penile body because skin tends to repel sound waves (Figure 4 and Figure 5). Sonogram with higher frequency transducer can differentiate the structures that made up prepuce tissues clearly.

Sonogram of Penis

Figure 5 shows sonogram of penis that consisted of glans penis (gp), body of penis (bp), and urethra (u).

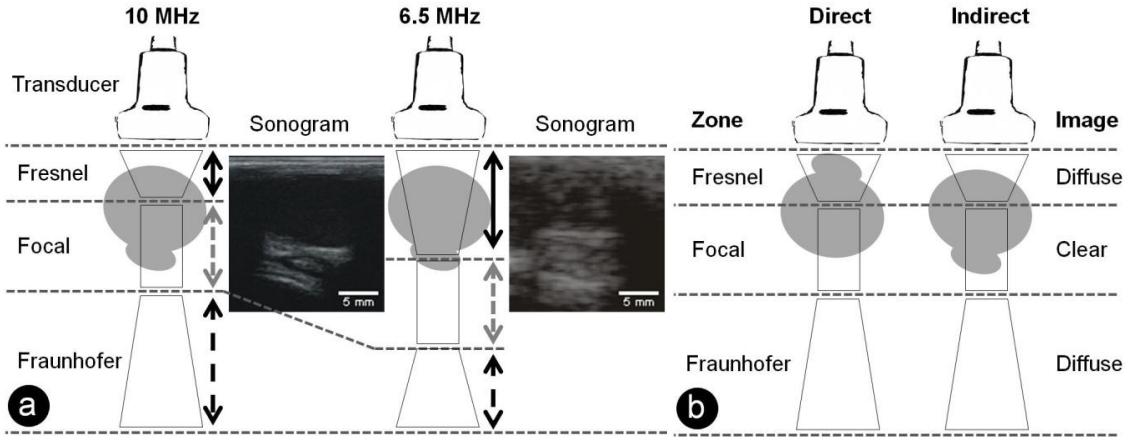

Figure 3. Classification zone of ultrasonography imaging. a= Transducers with different frequencies, b= Different imaging method

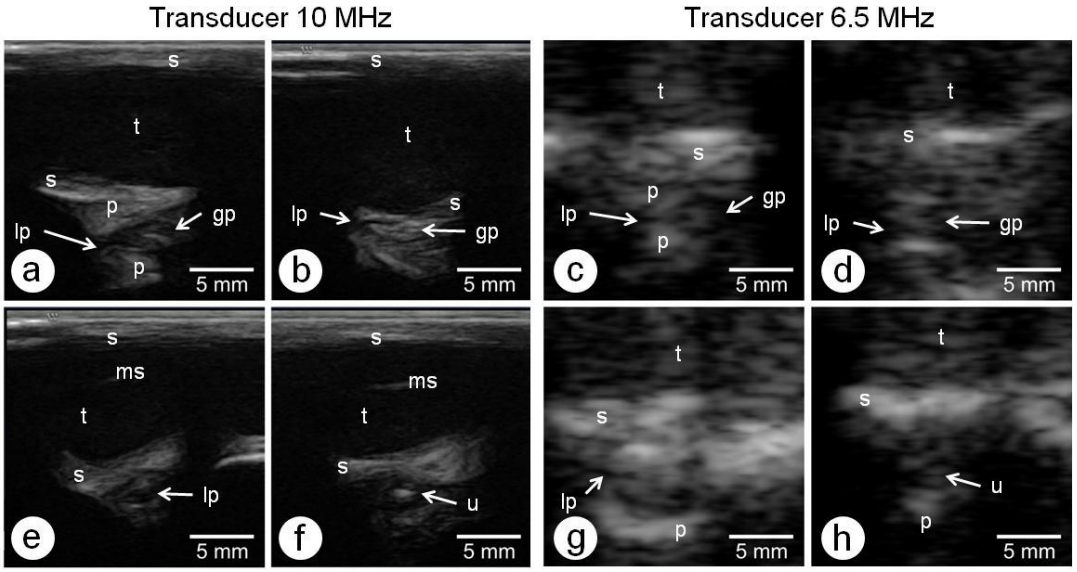

Figure 4. Sonogram of tom's prepuce and glans penis using 10 MHz transducer (a,b,e,f) and 6.5 MHz transducer (c,d,g,h). a-d= Longitudinal view, e-h= Transversal view, t= Testicle, ms= Mediastinum, s= Scrotum, p= Prepuce, lp= Prepuce lumen, gp= Glans penis, u= Urethra

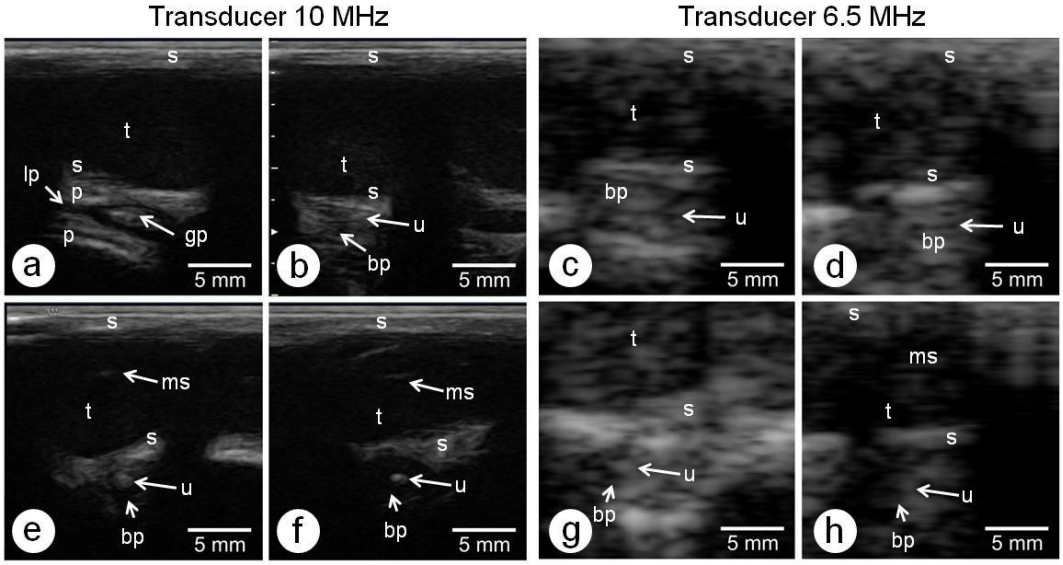

Figure 5. Sonogram of tom's penis using 10 MHz transducer (a,b,e,f) and 6.5 MHz transducer (c,d,g,h). a-d= Longitudinal view, e-h= transversal view, t= Testicle, ms= Mediastinum, s= Scrotum, lp= Prepuce lumen, bp= Penile body, gp= Glans penis, u= Urethra

Glans penis (gp), and body of penis (bp) appeared hypoechoic. Urethra (u) appeared hyperechoic while its lumen appeared anechoic. The components of penis could be seen more clearly in the image from sonogram with 10 MHz transducer compared to 6.5 MHz transducer. Scrotal skin (k) located near the transducer could not be imaged clearly while the components of the parts inside the focus area could be delineated clearly.

Penis is a copulatory organ that located at the ventral of scrotum and consists of body of penis, glans penis, and urethra (Dyce *et al.,* 2009). Toms have cone-shaped glans penis; the tip points caudally and has penile spikes (Johnston *et al.,* 2001). The spike numbers about 120-150 pieces (Davidson and Baker, 2009) and act as vaginal simulator or ovulation inductor for queen during copulation (Dyce *et al.,* 2009). For a very small penis or organ (< 5 mm), indirect ultrasonography is the technique of choice. Glans penis and body of penis appear hypoechoic because of its horn epithelium covered connective tissues that reflect of sound wave (Figure 4 and Figure 5). Short os (bone) of penis at the tip of glans penis appears distinctly on sonogram. Urethra appears hyperechoic with anechoic urethra lumen inside because it is filled with fluids. Sonogram equipped with higher frequency transducer can clearly differentiate all basic structure of an organ.

Sonogram of Epididymis

Figure 6 shows sonogram of epididymis as more hypoechoic structure compared to testicle (t). Caput epididymis (cpe) appeared more hyperechoic compared to cauda epididymis (cde). The boundary between epididymis and testicular body (t) could be seen more clearly in the image from 10 MHz transducer compared

to the image from 6.5 MHz transducer. Scrotal's skin (k) located near the transducer could not be imaged clearly while the components of the parts inside the focus area could be delineated clearly.

Epididymis is elongated ducts that located at the medial part of testicles and it has a function as a spermatozoa maturation site (Dyce *et al.*, 2009; Hebert *et al.*, 2012). Epididymis can be identified around testicles as a homogenous structure and appears hypoechoic compared to the testicles (Mantis, 2008). Ultrasonography of epididymis can be performed using either direct or indirect technique. In ultrasonography, caput epididymis appears as an anechoic structure. The size of duct is smaller in caput epididymis compared to cauda epididymis (Figure 6). In lower frequencies (6.5 MHz), it appears as a combination of hypoechoic and anechoic structure. At caput epididymis, anechoic appearance is more dominant compared to cauda epididymis. Scrotal skin appeared as hypoechoic to hyperechoic line.

Sonogram of Testicle

Figure 7 shows sonogram of testicle, the main organ that produces spermatozoa and the main reproductive organ in male animals. At the dorsolateral of testicle (t) there is cauda epididymis which is connected to spermatic duct. Testicles appeared anechoic at 10 MHz transducer while they appeared hypoechoic at 6.5 MHz transducer. The mediastinum (ms) in the middle of testicle (t) appeared hyperechoic. The boundary between scrotum (s) and testicular body (t) could be seen more clearly using 10 MHz transducer compared to 6.5 MHz transducer. Scrotal skin (k) located near the transducer could not be imaged clearly while the components of the parts inside the focus area could be delineated clearly.

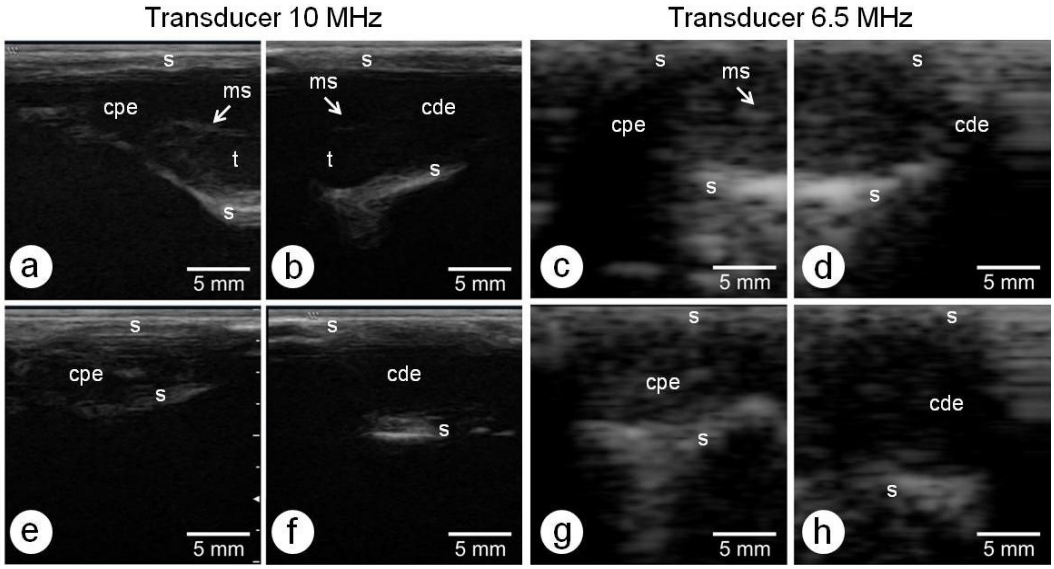

Figure 6. Sonogram of tom's epididymis using 10 MHz transducer (a,b,e,f) and 6.5 MHz transducer (c,d,g,h). a-d= Longitudinal view, e-h= Transversal view, s= Scrotum, ms= Mediastinum, cpe= Caput epididymis, cde= Cauda epididymis

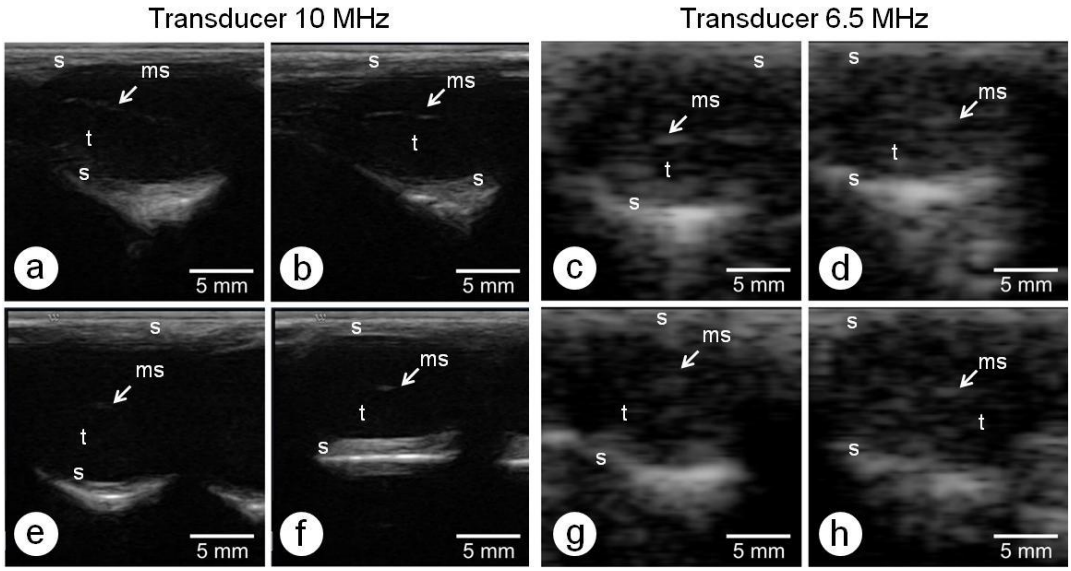

Figure 7. Sonogram of tom's testicle using 10 MHz transducer (a,b,e,f) and 6.5 MHz transducer (c,d,g,h). a-b= Longitudinal view, c-d= Transversal view, t= Testicle, ms= Mediastinum, s= Scrotum

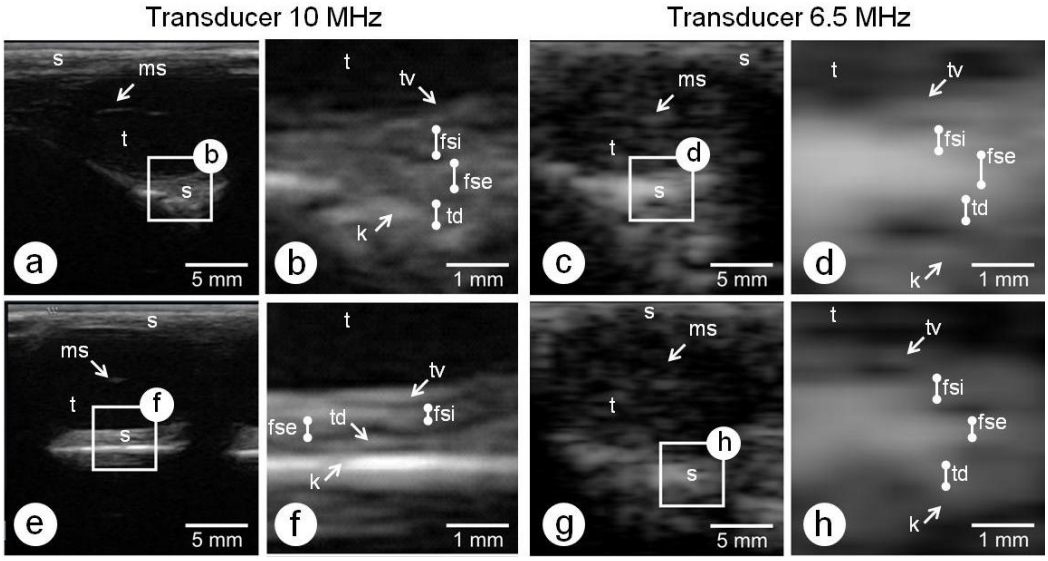

Figure 8. Sonogram of tom's scrotum using 10 MHz transducer (a,b,e,f) and 6.5 MHz transducer (c,d,g,h). a,c= Longitudinal view, e,g= Transversal view, b,d,f,h= Magnified image from boxes in figures a,c,e,g. t= Testicle, ms= Mediastinum, s= Scrotum, k= Skin, tv= Tunica vaginalis, fsi= Fascia spermatica interna, fse= Fascia spermatica externa, td= Tunica Dartos (muscle)

Testicle is an oval-shaped organ that produces spermatozoa, located inside scrotum (Davidson and Baker, 2009b; Dyce *et al.*, 2009). Testicle is protected by scrotum and it consists of right and left testicles that separated by median septum. In ultrasonography, testicles have similar echogenicity to spleen (Davidson and Baker, 2009b). Direct and indirect techniques could be used for testicle ultrasonography since it is >10 mm in size. Mediastinum in the middle part of testicular body appears more hyperechoic. At its lateral aspect, the boundary between scrotal connective tissue and testicle could be seen clearly in ultrasonography (Figure 6-8). Tunica vaginalis is a connective tissue adjacent to testicle body. This layer appeared as a hyperechoic structure under testicles as a connective tissue over the internal spermatic fascia. In ultrasonography, blood vessels would appear as anechoic structures.

Sonogram of Scrotum

Figure 8 shows sonogram of scrotum (s) as the skin (sac) that envelops testicle (t). The profundal of tunica Dartos (td) appeared more hypoechoic compared to the skin (k). Scrotal (s) parts such as skin (k), tunica vaginalis (tv), fascia spermatica interna (fsi), fascia spermatica externa (fse), and tunica Dartos (td) appeared more hypoechoic compared to the more hyperechoic skin (k). In the ultrasonographic image, skin (k) appeared hyperechoic among to the other scrotal parts. The boundary between scrotum and testicle could be seen more clearly using 10 MHz transducer compared to 6.5 MHz transducer. Scrotal skin (k) located near to the transducer could not be imaged clearly, while the components of the parts inside the focal zone could be delineated clearly.

Scrotum is a skin that cover (sac) for right and left testicle, located under the anus, and the superior part of prepuce and penis (Davidson and Baker, 2009b). Scrotal wall is formed by tunica vaginalis which consists of visceral and parietal layers, tunica albuginea, and tunica Dartos (Hebert *et al.*, 2012). Scrotum is located ventral to anus (Dyce *et al.*, 2009) with a width of < 2 mm. Indirect scrotum ultrasonography will show scrotum as a hyperechoic structure (Figure 6-8). Indirect approach could visualize each layer of scrotum, especially at 10 MHz frequency. Transducer with 10 MHz frequency can show the borders between tunica vaginalis, internal spermatic fascia, and external spermatic fascia that are adjacent to scrotum. Whereas in 6.5 MHz transducer, the border of tunica vaginalis was appear in diffuse image.

CONCLUSION

Ultrasonography of external reproductive organ of tom could be done easily. Ultrasonography of external reproductive organ is best performed using 10 MHz transducer rather than 6.5 MHz transducer because it provided clearer image. Imaging technique for external reproductive organ of tom sized less than 10 mm could be done by indirect method, transcutaneously.

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