ZACHODNIOPOMORSKI UNIWERSYTET
TECHNOLOGICZNY W SZCZECINIE
WYDZIAŁ BIOTECHNOLOGII I HODOWLI ZWIERZĄT

Natalia Wojtasiak

WYKORZYSTANIE ULTRASONOGRAFII W OCENIE PRZEBIEGU CIĄŻY I DIAGNOSTYCE PRENATALNEJ U KÓZ

Rozprawa doktorska wykonana
w Katedrze Biotechnologii Rozrodu Zwierząt
i Higieny Środowiska pod kierunkiem
dr hab. inż. Tomasza Stankiewicza, prof. ZUT

Szczecin 2023

Składam serdeczne podziękowania
i wyrazy uznania Panu
dr hab. inż. Tomaszowi Stankiewiczowi,
prof. ZUT za poświęcony czas,
cierpliwość i wsparcie w trudnych chwilach.
A zwłaszcza za nieocenioną pomoc udzieloną mi
w trakcie pisania niniejszej pracy.

Pragnę również podziękować Pani
dr hab. inż. Barbarze Błaszczyk,
prof. ZUT za zaangażowanie,
życzliwość i wsparcie merytoryczne.

.Chciałabym również podziękować moim wspaniałym Rodzicom,
dzięki którym miałam możliwość
realizować marzenia, kształcić się i zdobywać cenną wiedzę,
którzy stale mnie mobilizowali i
wspierali przez okres trwania studiów.

Niniejszą pracę pragnę zadedykować mojemu synowi.

Spis treści

1. WYKAZ PUBLIKACJI STANOWIĄCYCH ROZPRAWĘ DOKTORSKĄ

Praca doktorska pod tytułem „Wykorzystanie ultrasonografii w ocenie przebiegu ciąży i diagnostyce prenatalnej u kóz" została udokumentowana cyklem publikacji naukowych powiązanych tematycznie:

1. **Wojtasiak N.**, Stankiewicz T., Błaszczyk B., Udała J. (2022): Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43, (1), 97-102, IF: 2,3; Liczba punktów MEiN: 70; (Załącznik nr 1).
2. **Wojtasiak N.**, Stankiewicz T., Błaszczyk B., Szewczuk M.A. (2023): Doppler parameters in ductus venosus during the third trimester of pregnancy in goats. Animal Science and Genetics, 19, (3), 17-25; Liczba punktów MEiN: 20; (Załącznik nr 2).

Ponadto dokonano analizy aktualnego stanu wiedzy w zakresie wykorzystania ultrasonografii u ciężarnych kóz. Wyniki tej analizy przedstawiono w pracy przeglądowej:

3. **Wojtasiak N.**, Stankiewicz T., Udała J. (2020): Ultrasound examination of pregnancy in the domestic goat (Capra hircus) - a Review. Scientific Annals of the Polish Society of Animal Production, 16, (2), 65-78; Liczba punktów MEiN: 70; (Załącznik nr 3).

W roku wydania wyżej wymienionych publikacji sumaryczny Impact Factor (IF) wynosił 2,3 Według punktacji MEiN, zgodnie z rokiem opublikowania, publikacje te mają łącznie 160 punktów.

2. STRESZCZENIE

Celem naukowym rozprawy doktorskiej była ocena możliwości wykorzystania ultrasonografii w badaniu przebiegu ciąży i rozwoju zarodkowo-płodowego u kóz. Dla realizacji tego celu określono parametry morfometryczne zarodków/płodów oraz oceniono parametry dopplerowskie tętnic pępowinowych i przewodu żylnego podczas ciąży u kóz. Wykonano także analizę aktualnego stanu wiedzy w zakresie badań ultrasonograficznych ciąży u tego gatunku.

Badania wykonano na kozach rasy Burskiej utrzymywanych na fermie w Zakładzie Doświadczalnym Instytutu Zootechniki Państwowego Instytutu Badawczego w Kołbaczu. Kozy były kryte w czasie ich naturalnego sezonu rozrodczego, nie stosowano metod synchronizacji rui. Okres trwania ciąży określono na podstawie dnia pokrycia i potwierdzono retrospektywnie po porodzie.

W badaniach przeprowadzono analizę parametrów morfometrii zarodkowo-płodowej w pierwszym trymestrze ciąży wykorzystując ultrasonografię w trybie B. Natomiast wykorzystując ultrasonografię dopplerowską przeprowadzono ocenę hemodynamiki w tętnicach pępowinowych oraz w przewodzie żylnym u ciężarnych kóz w pierwszym i trzecim trymestrze. Określono również przydatność badanych parametrów w diagnostyce prenatalnej u kóz.

Uzyskane wyniki wykazały, że parametry morfometrii zarodkowo-płodowej zależą od dnia ciąży. Dlatego też parametry te powinny być uwzględniane w ultrasonograficznej ocenie rozwoju płodu u kóz, zwłaszcza w pierwszym trymestrze ciąży.

Przeprowadzone badania wskazują na możliwość wykorzystania ultrasonografii dopplerowskiej w monitorowaniu hemodynamiki tętnicy pępowinowej i przewodu żylnego podczas ciąży u kóz.

3. ABSTRACT

The scientific aim of the doctoral dissertation was to assess the possibility of using ultrasonography in the study of the course of pregnancy and embryo-foetal development in goats. To achieve this goal, the morphometric parameters of embryos/fetuses were determined and the Doppler parameters of the umbilical arteries and *ductus venosus* were assessed during pregnancy in goats. An analysis of the current state of knowledge in the field of ultrasound examination of pregnancy in this species was also carried out.

The research was carried out on Boer goats kept on a farm at the Experimental Department of the National Research Institute of Animal Production in Kołbacz. The goats were mated during their natural breeding season, estrus synchronization methods were not used. The duration of gestation was determined by the day of coverage and confirmed retrospectively after delivery.

In the study, the parameters of embryo-foetal morphometry were analyzed in the first trimester of pregnancy using B-Mode ultrasound. On the other hand, using Doppler ultrasonography, hemodynamics in the umbilical arteries and in the *ductus venosus* in pregnant goats was assessed in the first and third trimester. The usefulness of the examined parameters in prenatal diagnosis in goats was also determined.

The obtained results showed that the parameters of embryo-foetal morphometry depend on the day of pregnancy. Therefore, these parameters should be taken into account in ultrasound assessment of fetal development in goats, especially in the first trimester of pregnancy.

The conducted research indicates the possibility of using Doppler ultrasonography in monitoring the hemodynamics of the umbilical artery and *ductus venosus* during pregnancy in goats.

4. CEL NAUKOWY

Celem naukowym rozprawy doktorskiej była ocena możliwości wykorzystania ultrasonografii w badaniu przebiegu ciąży i rozwoju zarodkowo-płodowego u kóz.

Dla realizacji nadrzędnego celu naukowego wyznaczono następujące cele szczegółowe:

1. Analiza aktualnego stanu wiedzy w zakresie wykorzystania ultrasonografii w diagnostyce ciąży u kóz.
2. Ocena możliwości wykorzystania ultrasonograficznych parametrów morfometrycznych w monitorowaniu ciąży i rozwoju zarodkowo-płodowego u kóz.
3. Określenie możliwości wykorzystania techniki dopplerowskiej w ocenie hemodynamiki tętnic pępowinowych i przewodu żylnego podczas ciąży u kóz.

5. WPROWADZENIE I UZASADNIENIE PODJĘCIA BADAŃ

Aktualnie ultrasonografia w czasie rzeczywistym jest jedną z ważniejszych technik umożliwiających wykrywanie i monitorowanie ciąży u kóz. Zapewnia ona wczesną diagnozę ciąży, a w późniejszych etapach pozwala na monitorowanie rozwoju zarodka/płodu oraz szacowanie liczby oraz płci płodów (Jones i Reed, 2017).

U kóz ciąża trwa średnio 155 dni i okres ten podzielony jest na trzy trymestry. Pierwszy trymestr trwa do 49 dnia ciąży, drugi od 50 do 100 dnia ciąży, a trzeci trymestr rozpoczyna się w 101. dniu ciąży (Karadaev i in., 2018). Utrata ciąży może nastąpić w każdym okresie ciąży, ale największy odsetek obumierania zarodków występuje w pierwszym trymestrze (Samir i in., 2016).

Pierwszym parametrem w ultrasonograficznej ocenie ciąży jest obecność pęcherzyka ciążowego. Duży wpływ na czas wizualizacji i mierzoną średnicę pęcherzyka ciążowego ma rodzaj i częstotliwość sondy ultrasonograficznej (Devi i in., 2019). Wielkość pęcherzyka ciążowego można ocenić za pomocą głowicy transrektalnej już w 19 dniu po zapłodnieniu (Amer, 2010), natomiast od 25 dnia ciąży ocenę pęcherzyka ciążowego u kóz można przeprowadzić przezbrzusznie (Devi i in., 2019). Z drugiej strony inni autorzy proponują odłożenie badania do 32 dnia ciąży, aby uniknąć fałszywie dodatniego rozpoznania wynikającego z wczesnej utraty zarodka (Jones in., 2016).

Dla ultrasonograficznej oceny rozwoju płodu określane są między innymi takie parametry jak: długość ciemieniowo-siedzeniowa (ang. *crown-rump length* - CRL), średnica dwuciemieniowa (ang. *biparietal diameter* - BPD), średnica oczodołu (ang. *foetal orbit diameter* - OD) czy średnicy klatki piersiowej (ang. *chest diameter* - CD). Badania wykazały wysoką korelację między wymienionymi parametrami a wiekiem ciążowym u kóz (Karadaev i in., 2018; Yazici i in., 2018). W diagnostyce ultrasonograficznej ciąży wykorzystuje się pomiar średnicy łożyska (ang. *placentome diameter* - PD) (Enginler i in., 2021).

Większość badań koncentruje się na skuteczności wykrywania ciąży, wieku ciążowym w drugiej połowie ciąży (Airina i in., 2011; Elmetwally i in., 2016a,b; Gouda i in., 2021). Ustalenie dokładnych przyczyn utraty zarodków/płodów u kóz jest trudne ze względu na ograniczenia w dokładnym określeniu stanu wczesnej ciąży. Dlatego tak ważne są badania nad oceną ultrasonograficznych parametrów pozwalających wykryć ciążę i określić wiek ciąży w możliwie jak najszybszym okresie.

Oprócz diagnostyki ultrasonograficznej w trybie B w ocenie rozwoju zarodka/płodu wiele uwagi poświęca się obecnie badaniom dopplerowskim (Elmetwally i in., 2016a,b; Elmetwally i Meinecke-Tillmann, 2018; Stankiewicz i in., 2020). W ostatnich latach możliwości obrazowania hemodynamiki naczyniowej z wykorzystaniem ultradźwięków ogromnie wzrosły. Ultrasonografia dopplerowska służy między innymi do monitorowania rozwoju zarodkowo-płodowego (Kumar i in., 2015). Za pomocą tej nieinwazyjnej techniki bada się tętnice maciczno-łożyskowe (Ramírez-González i in., 2023), naczynia pępowinowe (Kumar i in., 2015) oraz naczynia płodowe takie jak aorta (Fasulkov i in., 2021) czy naczynia nerkowe płodu (Stankiewicz i in., 2023). Dostarcza ona również informacji o stanie płodu, pomaga w diagnostyce ewentualnych nieprawidłowości, które mogą mieć wpływ na przeżycie płodu i noworodka (Da Silva i in., 2018).

Jest coraz więcej dowodów, że prawidłowa budowa i funkcja naczyń pępowinowych oraz całej pępowiny warunkują prawidłowy rozwój płodu (Krzyżanowski i in., 2019). Określenie wskaźnika pulsacji (ang. *pulsatility index* - PI) i wskaźnika oporu (ang. *resistance index* - RI) naczyń pępowiny jest ważnym elementem ultrasonografii dopplerowskiej i często kluczowym podczas oceny rozwoju ciąży. Podwyższony RI tętnicy pępowinowej jest związany z opóźnionym wzrostem wewnątrzmacicznym, wadami wrodzonymi i innymi nieprawidłowościami w przebiegu ciąży (Elmetwally i in., 2016a,b).

Ogromne znaczenie w tych badaniach ma ultrasonografia wątroby płodu. Wątroba płodu jest głównym narządem krwiotwórczym podczas rozwoju płodowego, odgrywa istotną rolę w hematopoezie (Lewis i in., 2021). W życiu płodowym zajmuje również uprzywilejowaną pozycję w krążeniu płodowym, ponieważ jest pierwszym narządem, do którego krew trafia bezpośrednio z łożyska (Sørensen i in., 2011). Natleniona krew do wątroby płodu kozy trafia z żyły wrotnej i żył pępowinowych, które wewnątrz płodu tworzą wewnątrzbrzuszną żyłę pępowinową (Bejdić i in., 2021). Dopływ żylny do lewego płata wątroby pochodzi z bogatej w składniki odżywcze krwi z wewnątrzbrzusznej żyły pępowinowej, w przypadku prawego płata wątroby połowa krwi pochodzi z wewnątrzbrzusznej żyły pępowinowej, a druga połowa z ubogiej w składniki odżywcze krwi z żyły wrotnej (Hauge i in., 2004; Sørensen i in., 2011). Ten podział przepływu krwi między żyłą wrotną a wewnątrzbrzuszną żyłą pępowinową odpowiada za dychotomię czynnościową, która może być modyfikowana przez wpływy hemodynamiczne (Kiserud, 2003; Hauge i in., 2004).

Wątroba płodu z jej układem żylnym to główne obszary zainteresowania w badaniu krążenia płodowego. W szczególności przepływ krwi przez przewód żylny (*ductus venosus*)

oraz jego połączenie z wewnątrzbrzuszną żyłą pępowinową. Przewód żylny płodu kóz jest zakrzywionym naczyniem w kształcie trąbki, położonym w centralnej części wątroby, powyżej *porta hepatis* (Bejdić i in., 2021). Odgrywa on kluczową rolę w dystrybucji wysoko natlenionej krwi żylnej pępowinowej, której część omija miąższ wątroby przechodząc przez przewód żylny, żyłę główną ogonową, otwór owalny, lewy przedsionek serca i ostatecznie dociera do serca i mózgu (Seravalli i in., 2016).

Dlatego badania te mają duże znaczenie poznawcze na temat życia wewnątrzmacicznego i mogą być użytecznym wskaźnikiem w ocenie prawidłowego rozwoju płodu.

6. METODYKA BADAŃ, UZYSKANE WYNIKI I ICH OMÓWIENIE

Badania wykonano na kozach rasy Burskiej utrzymywanych na ekologicznej fermie kóz w Zakładzie Doświadczalnym Instytutu Zootechniki Państwowego Instytutu Badawczego w Kołbaczu (Kołbacz, Polska: szerokość geograficzna 53’30’’N). Kozy utrzymywane były w systemie pastwiskowo-alkierzowym. Żywienie odbywało się zgodnie z normami przyjętymi dla tego gatunku, w oparciu o zielonkę pastwiskową i inne pasze objętościowe oraz treściwe, w zależności od pory roku. Zwierzęta miały stały dostęp do wody i lizawek solnych. Kozy były kryte w czasie ich naturalnego sezonu rozrodczego w sposób naturalny (krycie z ręki), nie stosowano metod synchronizacji rui. Okres trwania ciąży określono na podstawie dnia pokrycia, a skuteczność krycia potwierdzono ultrasonografią transrektalną (aparat ultrasonograficzny EDAN U50, sonda linearna, 9.4 MHz, model V742UB) w okresie od 20 do 30 dnia po kryciu. Data poczęcia została potwierdzona retrospektywnie przy założeniu, że ciąża trwała 148 dni (Stankiewicz i in., 2020; Wojtasiak i in., 2023).

6.1. Realizacja celu pierwszego (cel szczegółowy I)

Dla realizacji tego celu przeprowadzono szczegółową analizę dostępnych danych z badań ultrasonograficznych na ciężarnych kozach. Na podstawie wyników badań opublikowanych w artykułach naukowych zebrano i usystematyzowano dane dotyczące ultrasonograficznych parametrów biometrycznych zarodków-płodów wykonywanych podczas badania ciężarnych kóz. Opisano przygotowanie ciężarnej kozy do badania oraz stosowane techniki badań ultrasonograficznych. W artykule dokonano szczegółowego opisu pomiarów fetometrycznych stosowanych w badaniu ultrasonograficznym ciąży u kóz. W tabelach przedstawiono średnie wartości parametrów biometrycznych, takich jak: długość ciemieniowo-siedzeniowa, średnica dwuciemieniowa płodu, średnica oczodołu, długość potyliczno-nosowa, średnica brzucha i klatki piersiowej, długość poprzeczna i podłużna osi serca, średnica pępowiny i aorty płodu, a także zewnętrzna i wewnętrzna średnica łożyszczy dla poszczególnych ras kóz w różnych etapach ciąży. Wskazano, że wdrożenie tej techniki USG w praktyce pozwala na wykonywanie badań w cyklicznych powtórzeniach na tych samych zarodkach/płodach. Ma to kluczowe znaczenie dla monitorowania wzrostu i rozwoju struktur zarodkowo-płodowych i pozapłodowych na każdym etapie ciąży.

Wyniki tej pracy opublikowano w artykule pt. „*Ultrasound examination of pregnancy in the domestic goat (Capra hircus) - a Review*”.

6.2. Realizacja celu drugiego (cel szczegółowy II)

Dla realizacji tego celu badania przeprowadzono na 14 kozach. U wszystkich badanych samic stwierdzono ciąże mnogie, które diagnozowano podczas ultrasonograficznego badania transrektalnego i potwierdzano podczas porodu. Ujednolicono też wiek kóz – badania przeprowadzono u wieloródek w wieku od 5 do 6 lat.

Dla wykonania pomiarów średnicy pęcherzyka ciążowego, długości ciemieniowo-siedzeniowej (CRL), wymiaru dwuciemieniowego głowy płodu (BPD), długości potyliczno-nosowej (ONL), średnicy klatki piersiowej (TD) i łożyszczy (PD) badanie rozpoczęto od 30 dnia i kontynuowano do 45 dnia ciąży w 5 dniowych odstępach. Badanie ultrasonograficzne wykonywano za pomocą aparatu ultrasonograficznego (EDAN U50) wyposażonego w głowicę linearną o częstotliwości do 9.4 MHz (Model, V742UP) i głowicę sektorową o częstotliwości do 5 MHz (Model, C352UB). Badanie ultrasonograficzne wykonywano przezbrzusznie. Badanie ultrasonograficzne wykonano u ciężarnych kóz, które nie były wcześniej poddane działaniu środków usypiających.

Każdorazowo u każdej kozy badano 10 największych łożyszcz (5 leżących w pobliżu płodu i 5 leżących w rogach macicy). Pomiar wykonywano między dwiema najbardziej dystalnymi częściami łożyszcza przy użyciu ultrasonografii w trybie B. Uzyskane wyniki następnie uśredniano.

Przeprowadzone badania wykazały, że wszystkie badane parametry były istotnie skorelowane z dniem ciąży. Zarówno średnica pęcherzyka ciążowego, łożyszczy i parametry biometryczne zarodków/płodów wzrastały wraz z zaawansowaniem ciąży.

Wyniki tych badań opublikowano w artykule pt. *„Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats”*.

6.3. Realizacja celu trzeciego (cel szczegółowy III)

Dla realizacji tego celu badania przeprowadzono na 14 kozach rasy Burskiej. U wszystkich badanych samic stwierdzono ciąże mnogie potwierdzone w badaniu transrektalnym. Badania przeprowadzono u wieloródek w wieku od 5 do 6 lat.

Dla wykonania pomiarów przepływu krwi w tętnicach pępowinowych badanie rozpoczęto w 30 dniu ciąży i kontynuowano do 45 dnia ciąży w 5 dniowych odstępach. Natomiast dla wykonania pomiaru przepływu krwi w przewodzie żylnym badanie rozpoczęto od 100 dnia ciąży i kontynuowano przez cały trzeci trymestr ciąży w odstępach kilkudniowych. Badanie ultrasonograficzne wykonywano za pomocą aparatu ultrasonograficznego (EDAN U50) wyposażonego w głowicę linearną o częstotliwości do 9.4 MHz (Model, V742UP) i głowicę sektorową o częstotliwości do 5 MHz (Model C352UB). Badanie ultrasonograficzne wykonywano przezbrzusznie u kóz, które nie były wcześniej poddane działaniu środków uspokajających.

Dla zbadania parametrów dopplerowskich tętnicy pępowinowej każdorazowo u każdej kozy badano wolną część pępowiny w pobliżu wstawki brzusznej, której skanowanie przeprowadzano przy użyciu ultrasonografii w trybie B. W celu identyfikacji naczyń tętniczych w pępowinie wykorzystano kolorowego Dopplera. Po zlokalizowaniu naczyń krwionośnych dokonano pomiarów przepływu krwi w tętnicach pępowinowych wykorzystując pulsacyjną ultrasonografię dopplerowską. Określono następujące parametry dopplerowskie: szczytową prędkość skurczową (PSV), końcową prędkość rozkurczową (EDV), indeks oporności (RI) i indeks pulsacji (PI).

Przeprowadzone badania wykazały, że parametry dopplerowskie takie jak PSV i PI w tętnicach pępowinowych były istotnie skorelowane z dniem ciąży. W tętnicach pępowinowych prędkość przepływu krwi wzrastała, a indeks pulsacji malał wraz z zaawansowaniem ciąży. Wartość indeksu oporności (RI) pozostawał taki sam przez cały okres badania. W tętnicach pępowinowych nie odnotowano końcowej prędkości rozkurczowej (EDV) u żadnej kozy w okresie badania.

W tętnicach pępowinowych stosunek PSV/EDV nie został określony ze względu na brak pomiaru końcowej prędkości rozkurczowej w badanym okresie ciąży. Według Stankiewicza i in. (2020) stwierdza się istotne obniżenie wartości PSV/EDV od 70. dnia ciąży. Elmetwally i in. (2016a,b) podają, że stosunek PSV/EDV, który jest jednym z najważniejszych parametrów dopplerowskich, wskazuje na zmiany końcowej prędkości rozkurczowej, zwłaszcza w drugim i trzecim trymestrze ciąży.

Dla zbadania parametrów dopplerowskich w przewodzie żylnym każdorazowo u każdej kozy po uwidocznieniu płodu w trybie B przeprowadzono ocenę unaczynienia z wykorzystaniem kolorowego i pulsacyjnego Dopplera. Przewód żylny uwidoczniono w strzałkowym lub skośnym przekroju brzucha płodu. Przewód żylny badano w przesmyku, w pobliżu jego odejścia od żyły pępowinowej. Naczynie to znajdowało się w centralnej części wątroby i biegło w kierunku dogłowowym, z rosnącym nachyleniem w tej samej płaszczyźnie strzałkowej, co pierwotny kierunek żyły pępowinowej. Po zlokalizowaniu naczyń krwionośnych dokonano pomiaru przepływu krwi w przewodzie żylnym wykorzystując pulsacyjną ultrasonografię dopplerowską. Określono następujące parametry dopplerowskie: szczytową prędkość skurczową (PSV), końcową prędkość rozkurczową (EDV), stosunek PSV/EDV, indeks oporności (RI) i indeks pulsacji (PI).

Przeprowadzone badania wykazały, że widmo fali przepływu krwi w przewodzie żylnym ma charakter pulsacyjny z widocznymi dwiema fazami przyspieszenia (first and second peak). Uważa się, że przebieg i kształt fali widma dopplerowskiego w przewodzie żylnym mogą tłumaczyć występujące zmiany ciśnienia i objętości przedsionków w skurczowej i rozkurczowej fazie cyklu serca (Seamundsson i in., 2011; Seravalli i in., 2016). W pracy zanotowano też, że wszystkie parametry dopplerowskie w przewodzie żylnym były istotnie skorelowane z dniem ciąży.

Przeprowadzone badania wykazały, że szczytowa prędkość skurczowa znacznie wzrosła w ostatnich dniach trzeciego trymestru ciąży, natomiast wartość EDV uległa znaczącemu obniżeniu w przewodzie żylnym. W pracy po raz pierwszy określono wartości second peak systolic dla piku rozkurczowego. Można sądzić, że występowanie drugiej fazy przyśpieszenia związane jest ze wzrostem przepływu krwi przez serce (Seravalli i in., 2016).

W niniejszej pracy wykazano także, że średnie wartości RI w przewodzie żylnym pod koniec trzeciego trymestru, podobnie jak w przypadku PI, były znacznie wyższe niż w początkowym okresie trzeciego trymestru. Być może krew pępowinowa aby dostać się do lewego przedsionka musi zostać obciążona energią kinetyczną oraz charakteryzować się wysokim ciśnieniem w celu pokonania oporu przewodu żylnego i dostania się do otworu owalnego (Kiserud i Kessler, 2023). Może to tłumaczyć wysokie wartości takich parametrów dopplerowskich jak prędkość przepływu, wskaźnik pulsacji i wskaźnik oporu.

Przeprowadzone badania wskazują na możliwość wykorzystania ultrasonografii dopplerowskiej, zarówno w monitorowaniu hemodynamiki tętnic pępowinowych, jak i hemodynamiki przewodu żylnego podczas ciąży u kóz.

Wyniki tych badań zostały opublikowane w artykule pt. „*Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats*" oraz w artykule pt. „*Doppler parameters in ductus venosus during the third trimester of pregnancy in goats*".

7. WNIOSKI

1. Wykazane w niniejszej pracy zmiany w morfometrii zarodków/płodów i łożyszczy zależą od wieku ciążowego, co może być pomocne w diagnostyce wczesnej ciąży i monitorowania dalszych jej etapów.
2. Przeprowadzone badania wskazują na możliwość wykorzystania ultrasonografii dopplerowskiej w ocenie hemodynamiki tętnic pępowinowych i przewodu żylnego podczas ciąży u kóz.
3. Analiza dostępnych danych piśmiennictwa naukowego oraz przeprowadzone badania wykazały, że ultrasonografia jest skuteczną metodą diagnostyczną w monitorowaniu przebiegu ciąży i rozwoju zarodkowo-płodowego u kóz.

8. PIŚMIENNICTWO

1. Airina RI, Nizam AM, Abdullah RB, Khadijah WW. (2011). Using fetal-heart size measured from ultrasound scanner images to estimate age of gestation in goat. Journal of Animal and Veterinary Advances, 10: 2528-2540.
2. Amer HA. (2010). Ultrasonographic assessment of early pregnancy diagnosis, fetometry and sex determination in goats. Animal Reproduction Science, 117: 226–31. https://doi.org/10.1016/j.anireprosci.2009.05.015
3. Bejdić P., Ćutuk A., Alić A., Čengić B., Avdić R., Tandir F., Hadžiomerović N., Mrvić V. (2021). Comparative Anatomical Studies on Ductus Venosus in Fetuses of Domestic Ruminants. Macedonian Veterinary Review, 44: 29-36. https://doi.org/10.2478/macvetrev-2020-0034
4. Devi NB, Bhuyan D and Das A. (2019). Application of ultrasonic doppler technique for pregnancy diagnosis in local Assam goat. The Pharma Innovation Journal, 8:1088-91.
5. Da Silva PDA, Uscategui RAR, Santos VJC, Taira AR, Mariano RSG, Rodrigues MGK, Simões APR, Maronezi MC, Avante ML, Vicente WRR, Feliciano MAR. (2018). Qualitative and quantitative ultrasound attributes of maternal-foetal structures in pregnant ewes. Reproduction in Domestic Animals, 53: 725-732. https://doi.org/10.1111/rda.13163
6. Elmetwally MA and Meinecke-Tillmann S. (2018). Simultaneous umbilical blood flow during normal pregnancy in sheep and goat foetuses using non-invasive colour Doppler ultrasound. Animal Reproduction, 15: 148. doi: 10.21451/1984-3143-AR2017-976
7. Elmetwally MA, Rohn K and Meinecke-Tillmann S. (2016a). Doppler sonography is a useful method to assess the effects of maternal anxiety on intrauterine fetal growth in pregnant sheep and goats. Quality in Primary Care, 24: 137-45.
8. Elmetwally MA, Rohn K and Meinecke-Tillmann S. (2016b). Non-invasive color Doppler sonography of uterine blood flow throughout pregnancy in sheep and goats. Theriogenology, 85: 1070-79. https://doi.org/10.1016/j.theriogenology.2015.11.018
9. Enginler SÖ, Dal GE, Cetin AC, Sabuncu A., Baykal K. (2021). Can gestational age be determined by placentome diameter, placentome blood flow pixel area and progesterone concentration during pregnancy in Kivircik ewes. Kafkas Universitesi Veteriner Fakultesi Dergisi, 27: 6. DOI : 10.9775/kvfd.2021.26292
10. Fasulkov IR, Karadaev M., Vasilev N., Hristov K., Fedev I. (2021). Doppler ultrasound measurements of the blood flow velocity in the fetal heart and aorta in Bulgarian White

milk goats. Veterinary Medicine and Science, 7: 1297-1302. https://doi.org/10.1002/vms3.463

11. Gouda A., Aggag M., Kandiel MM. (2021). Ultrasound-guided fetometry in Egyptian sheep and she-goats. Benha Veterinary Medical Journal, 40: 114-18. 10.21608/BVMJ.2021.58131.1330
12. Hauge G., Kiserud T., Godfrey K., Crozier S., Hanson M. (2004). Portal and umbilical venous blood supply to the liver in the human fetus near term. Ultrasound in obstetrics & gynecology, 24: 599-605. https://doi.org/10.1002/uog.1744
13. Jones AK, Reed SA. (2017). Benefits of ultrasound scanning during gestation in the small ruminant. Small Ruminant Research, 149: 163–171. https://doi.org/10.1016/j.smallrumres.2017.02.008
14. Jones, AK, Gately RE, McFadden KK, Zinn SA, Govoni KE, Reed SA. (2016). Transabdominal ultrasound for detection of pregnancy, fetal and placental landmarks, and fetal age before day 45 of gestation in the sheep. Theriogenology, 85: 939-945. https://doi.org/10.1016/j.theriogenology.2015.11.002
15. Karadaev M., Fasulkov I., Stanimir Y., Atanasova S., Vasilev N. (2018). Determination of gestational age through ultasound measurements of some uterine and foetal parameters in Bulgarian local goats. Reproduction in Domestic Animals, 53: 1456–1465. https://doi.org/10.1111/rda.13305
16. Kiserud T. (2003): Fetal venous circulation. Fetal and Maternal Medicine Review, 14: 57-95. doi:10.1017/S0965539503001037
17. Kiserud T., Kessler J. (2023). The ductus venosus. Doppler Ultrasound in Obstetrics and Gynecology, 449-473. https://doi.org/10.1007/978-3-031-06189-9_28
18. Krzyżanowski A., Kwiatek M., Gęca T., Stupak A., Kwaśniewska A. (2019). Modern ultrasonography of the umbilical cord: prenatal diagnosis of umbilical cord abnormalities and assessement of fetal wellbeing. Medical Science Monitor: international medical journal of experimental and clinical research, 25: 3170-3180. doi: 10.12659/MSM.913762
19. Kumar K., Chandolia RK, Kumar S., Jangir T., Luthra RA, Kumari S., Kumar S. (2015). Doppler sonography for evaluation of hemodynamic characteristics of fetal umbilicus in Beetal goats. Veterinary World, 8: 412. doi: 10.14202/vetworld.2015.412-416
20. Lewis K., Yoshimoto M., Takebe T. (2021). Fetal liver hematopoiesis: from development to delivery. Stem Cell Research & Therapy, 12: 1-8. https://doi.org/10.1186/s13287-021-02189-w

21. Ramírez-González D., Poto Á., Peinado B., Almela L., Navarro-Serna S., Ruiz S. (2023). Ultrasonography of Pregnancy in Murciano-Granadina Goat Breed: Fetal Growth Indices and Umbilical Artery Doppler Parameters. Animals, 13: 618. https://doi.org/10.3390/ani13040618
22. Saemundsson Y., Johansson C., Wenling S., Gudmundsson S. (2011). Hepatic venous Doppler in the evaluation of fetal extrasystoles. Ultrasound in Obstetrics & Gynecology, 37: 179-183. https://doi.org/10.1002/uog.7684
23. Samir H., Karen A., Ashmawy T., Abo-Ahmed M., El-Sayed M., Watanabe G. (2016). Monitoring of embryonic and fetal losses in different breeds of goats using real-time B-mode ultrasonography. Theriogenology, 85: 207–215. https://doi.org/10.1016/j.theriogenology.2015.09.039
24. Seravalli V., Miller JL, Block-Abraham D., Baschat AA. (2016). Ductus venosus Doppler in the assessment of fetal cardiovascular health: an updated practical approach. Acta Obstetricia et Gynecologica Scandinavica, 95: 635-644. https://doi.org/10.1111/aogs.12893
25. Sørensen A., Holm D., Pedersen M., Tietze A., Stausbøl-Grøn B., Duus L., Uldbjerg N. (2011). Left–right difference in fetal liver oxygenation during hypoxia estimated by BOLD MRI in a fetal sheep model. Ultrasound in obstetrics & gynecology, 38: 665-672. https://doi.org/10.1002/uog.9044
26. Stankiewicz T., Błaszczyk B., Chundekkad P. (2023). Ultrasound evaluation of biometric and Doppler parameters of kidneys in sheep in the prenatal and postnatal periods. Small Ruminant Research, 219. https://doi.org/10.1016/j.smallrumres.2022.106891
27. Stankiewicz T., Błaszczyk B., Udała J., Chundekkad P. (2020). Morphometric measurements of the umbilical cord and placentomes and Doppler parameters of the umbilical artery through ultrasonographic analysis in pregnant sheep. Small Ruminant Research, 184: 106043. https://doi.org/10.1016/j.smallrumres.2019.106043
28. Wojtasiak N., Stankiewicz T., Błaszczyk B., Udała J. (2023). Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43: 97-102. DOI: 10.29261/pakvetj/2022.067
29. Yazici E., Ozenc E., Celik HA, Ucar M. (2018). Ultrasonographic foetometry and maternal serum progesterone concentrations during pregnancy in Turkish Saanen goats. Animal reproduction science, 197: 93-105. https://doi.org/10.1016/j.anireprosci.2018.08.017

9. WYKAZ ZAŁĄCZNIKÓW

Załącznik 1: publikacja

Wojtasiak N., Stankiewicz T., Błaszczyk B., Udała J. (2022): Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43, (1), 97-102.

Załącznik 2: publikacja

Wojtasiak N., Stankiewicz T., Błaszczyk B., Szewczuk M.A. (2023): Doppler parameters in ductus venosus during the third trimester of pregnancy in goats. Animal Science and Genetics, 19, (3), 17-25.

Załącznik 3: publikacja

Wojtasiak N., Stankiewicz T., Udała J. (2020): Ultrasound examination of pregnancy in the domestic goat (Capra hircus) - a Review. Scientific Annals of the Polish Society of Animal Production, 16, (2), 65-78.

Załącznik 4: Oświadczenia współautorów o procentowym udziale w przygotowaniu publikacji.

9.1. Załącznik 1.

Wojtasiak N., Stankiewicz T., Błaszczyk B., Udała J. (2022): Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43, (1), 97-102.

Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
DOI: 10.29261/pakvetj/2022.067

RESEARCH ARTICLE

Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats

Natalia Wojtasiak*, Tomasz Stankiewicz, Barbara Błaszczyk and Jan Udała

West Pomeranian University of Technology in Szczecin, Faculty of Biotechnology and Animal Husbandry, Department of Animal Reproduction Biotechnology and Environmental Hygiene, 29 Klemensa Janickiego Street, 71-270 Szczecin, Poland
*Corresponding author: wojtasiak1natalia@gmail.com

ARTICLE HISTORY (22-265)

Received:	August 05, 2022
Revised:	August 22, 2022
Accepted:	August 31, 2022
Published online:	September 20, 2022

Key words:
Embryo-fetal fetometry,
Umbilical cord
Ultrasonography
Doppler
Pregnancy
Goats

A B S T R A C T

The aim of this study was to determine the biometric parameters of the embryo/fetus and to assess the haemodynamics in the umbilical artery during the first trimester of pregnancy in 14 Boer goats. All the goats under study had twin pregnancy. Gestational vesicle diameter (GSD), crown-rump length (CRL), biparietal diameter (BPD), occipital-nasal length (ONL), trunk diameter (TD) and the placentome diameter (PD) were measured from the 30th to the 45th day of pregnancy at 5-day intervals, using a B-mode ultrasound scanner. Measurements of blood flow parameters in the umbilical artery were made using a colour Doppler combined with a pulse wave Doppler technique. During the study period, there was a significant ($P<0.05$) increase in the diameter of the gestational sac and most of the biometric parameters of the embryo/fetus. The average CRL on day 45 was almost 10 times higher than that on day 30 of pregnancy. The peak systolic velocity in the arterial vessels on day 45 of pregnancy was higher ($P<0.05$) than that from 30 to 40 days of pregnancy. On the other hand, mean pulsation index (PI) at day 45 of pregnancy was lower ($P<0.05$) than that on day 30 and day 40 of pregnancy. No end-diastolic velocity was diagnosed in any goat during the study period. All parameters showed positive ($P<0.01$) correlations with the day of pregnancy, except the PI in the arterial vessels, which showed negative ($P<0.01$) correlation with the day of pregnancy. In conclusion, changes in the parameters of embryo-fetal morphometry, placentomes and blood flow parameters of the umbilical artery may be helpful in assessing the development of the embryo and the fetus during the first trimester of pregnancy in goats.

To Cite This Article: Wojtasiak N, Stankiewicz T, Błaszczyk B and Udała J, 2023. Ultrasound parameters of embryo-fetal morphometry and doppler indices in the umbilical artery during the first trimester of pregnancy in goats. Pak Vet J, 43(1): 97-102. http://dx.doi.org/10.29261/pakvetj/2022.067

INTRODUCTION

Ultrasonography is a useful diagnostic imaging technique used in the reproductive management of small and large animals, as well as in humans. This technique is mainly used for the detection of early pregnancy, monitoring of embryo/foetal development and the assessment of the number and gender of fetuses (Jones and Reed, 2017). Previous studies in goats indicate that the presence of a gestational vesicle (GS) in the lumen of the uterus is a confirmation of developing embryo (Muhammad and Aziz, 2021). In goats, pregnancy lasts for about 155 days on the average and this period is divided into three trimesters. The first trimester lasts until the 49th day of pregnancy, the second from the 50th to the 100th day of pregnancy, and the third trimester begins on the 101st day of pregnancy (Karadaev *et al.*, 2018). Previous studies also showed that from the 35th day of goat's pregnancy, organs such as forestomach are visible, which are indicative of an early stage of fetal development (Garcia *et al.*, 2014).

Loss of pregnancy can occur at any time, with the highest percentage of losses occurring in the early and late stages of embryonic development and in the early stages of fetal development, including the first trimester of pregnancy (Samir *et al.*, 2016). Determining the exact causes of fetal loss or embryonic mortality in ruminants is difficult due to limitations in accurately determining the state of early pregnancy. However, studies emphasize the importance of ultrasonography in monitoring the loss of embryos and fetuses (Jones *et al.*, 2016). These studies indicate a high efficiency of detecting pregnancy loss in

ruminants, amounting to nearly 100%, which results in a small number of false negative diagnoses (Samir *et al.*, 2016; Jones *et al.,* 2016). For the ultrasonographic evaluation of the development of the fetus from the 30th day of pregnancy, parameters such as crown-rump length (CRL), biparietal diameter (BPD), orbital diameter (OD) and chest diameter (CD) are used (Karadaev *et al.,* 2018; Yazici *et al.,* 2018). Placentome diameter (PD) is also used in the ultrasound diagnosis of pregnancy (Enginler *et al.,* 2021). In addition to ultrasound diagnostics in the assessment of embryo/fetal development through B mode, much attention is paid to Doppler examinations, enabling the determination of the haemodynamics of the umbilical artery (Elmetwally *et al.,* 2016a,b; Elmetwally and Meinecke-Tillmann, 2018; Stankiewicz *et al.,* 2020).

The umbilical cord is the only link between the mother and the fetus through which respiratory gases, nutrients and metabolites can be transported. The correct structure and function of the umbilical vessels and the entire umbilical cord determine the proper development of the fetus. It is extremely important to confirm the correct number of umbilical vessels and their function in the abdominal cavity (Krzyżanowski *et al.,* 2019). Determining the pulsation index (PI) and resistance index (RI) of umbilical cord vessels is an important element of Doppler ultrasound examination and often a key element in the assessment of pregnancy development. An increased umbilical artery RI is associated with intrauterine growth retardation, congenital anomalies and other abnormalities in the course of pregnancy (Elmetwally *et al.,* 2016a,b). No lesions have been found in studies of the umbilical artery in pregnant goats in single and multiple pregnancies so far, such as end-diastolic flow (EDV) or a significant difference in PI and RI in single and multiple pregnancies (Serin *et al.*, 2010).

Most studies focus on the effectiveness of pregnancy detection, gestational age and the assessment of Doppler parameters in the second half of pregnancy (Airina *et al*., 2011; Elmetwally *et al*., 2016a,b; Gouda *et al*., 2021). The studies on the haemodynamics of umbilical vessels in the early stages of pregnancy in goats are limited. Therefore, aim of the present study was to determine the biometric parameters of the embryo, gestational vesicle and placenta, as well as to assess haemodynamics in the umbilical artery, during the first trimester of pregnancy in goats.

MATERIALS AND METHODS

Experimental animals and management: The present study was carried out on 14 Boer goats from September to December, 2021. These goats were multiparous (5-6 years old), with a balanced body weight (80 kg) and were kept on an ecological goat farm in Kołbacz (Poland: latitude 53'30 "N). They were kept on the pastures, as well as under indoor system with uniform feeding. In the grazing season, from May to October, the goats grazed on the meadow and received oats, hay and straw in the farm. In winter, from November to April, the goats stayed inside the pen, where they were fed with oats, straw and hay. The animals had constant access to water and salt licks.

The experimental goats were mated in a natural way during the natural breeding season. The stage of pregnancy was determined on the basis of the day of mating and the mating efficiency was confirmed by transrectal ultrasound (EDAN U50 ultrasound scanner, 4 MHz linear probe model V742UB) during the period from 20th to 30th day after mating. The date of conception was confirmed retrospectively on the assumption that the pregnancy lasted 148 days (Đuričić *et al.*, 2012). All the goats under study had twin pregnancy.

Ultrasound examination: The ultrasound examination was performed trans-abdominally from the 30th to the 45th day of pregnancy at 5-day intervals, using an ultrasound scanner (EDAN U50 ultrasound scanner) equipped with a sector probe with a frequency of up to 5 MHz (Model, C352UB). Before the examination, the inguinal area of the abdomen was clipped and a transmission gel was applied. The gestational vesicle diameter (GSD), crown-rump length (CRL), biparietal diameter (BPD), occipital-nasal length (ONL), trunk diameter (TD) and placentome diameter (PD) were recorded using B-mode ultrasound. The 10 largest placentomes were measured in each goat, 5 lying near the fetus and 5 lying in the uterine horns (Lekatz *et al.,* 2013) and the obtained results were then averaged. The diameter was measured at the highest position of ultrasound imaging (Stankiewicz *et al.,* 2020). In this study, the outer diameter of placentomes was measured, i.e. between the two most distal parts of the bearing (Lee *et al.,* 2005; Nwaogu *et al.,* 2010).

Measurements of blood flow parameters in the umbilical artery were made using a colour Doppler combined with a pulse wave Doppler technique. After locating the umbilical artery, blood flow measurements were taken and the following Doppler parameters were determined: peak systolic velocity (PSV), end diastolic velocity (EDV), resistance index (RI), and pulsation index (PI). The flow angle during the test was as close as possible to zero degree. The Doppler ultrasound examination of each goat took approximately 20-30 minutes. Doppler measurements were made on at least 5 continuous, regular waves of the Doppler spectrum. The ultrasound images and all relevant measurements were recorded automatically by the ultrasound scanner and saved. Measurements were not recorded during maternal and fetal movements. If any symptoms of anxiety appeared, the ultrasound examination was interrupted and repeated at a later date. No pharmacological interference was applied to the test animals, and proper welfare was ensured during the study.

Statistical analysis: Morphometric parameters are presented as mean ± standard deviation (SD), while Doppler parameters as mean±standard error of the mean (SEM). In the initial analysis, no significant differences were found between the two twin fetuses. Therefore, the results for both fetuses were averaged. The differences among means of four periods of pregnancy were analysed using the analysis of variance and the post-hoc test. Duncan's multiple range test was used to verify the significance of differences among means. The correlations between the tested parameters and days of pregnancy were calculated using the Pearson's correlation coefficient (r). Statistical analysis was performed using STATISTICA version 13.1, Stat Soft, Poland.

Fig. 1: Representative ultrasonograph showing the measurement of the crown-rump length of a goat on day 45 of pregnancy (CRL1).

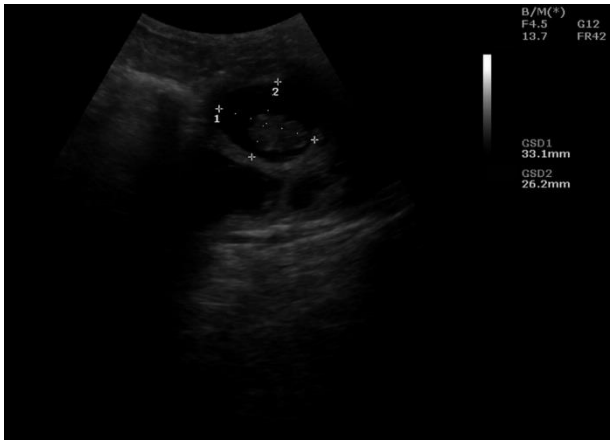

Fig. 2: Representative ultrasonograph showing the measurement of the gestational sac diameter of a goat on day 40 of pregnancy (GSD1, GSD2).

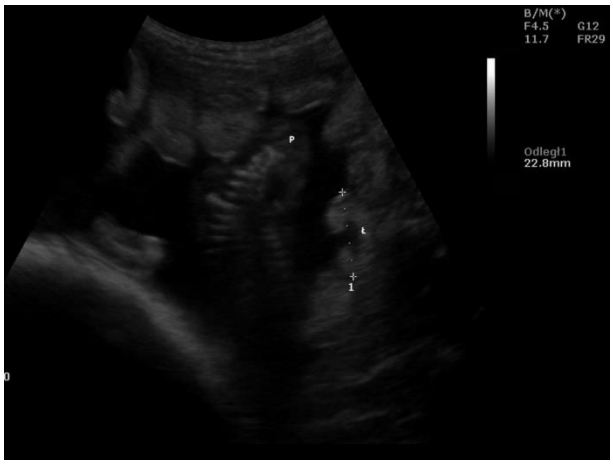

Fig. 3: Representative ultrasonograph showing the position of the fetus (P) and the assessment of placenta diameter (L) on day 45 of pregnancy in a goat.

RESULTS

Table 1 shows the mean values of the biometric parameters of the embryo/fetus, gestational sac and placentomes during the period from 30 to 45 days of pregnancy. In the analysed period, there was a significant increase in the diameter of the gestational sac and most of the examined biometric parameters of the embryo/fetus. The average CRL on day 45 was almost 10 times greater than on day 30 of pregnancy. There was also an increase in GSD and placentome diameter, but the differences in GSD between days 35 and 40 of gestation and placentome diameter between days 40 and 45 were statistically non-significant. Representative ultrasound images of the gestational vesicle, placenta and embryo are presented in Fig. 1-3.

Table 1: Biometric parameters of the embryo/fetus, embryonic vesicle and placentomes during the first trimester of pregnancy in goats (n=14)

Parameters		Days of pregnancy			
		30	35	40	45
GSD (mm)	mean±SD	21.44±1.24[a]	29.92±1.18[b]	31.47±1.58[b]	39.13±2.98[c]
	range	(19.90-24.00)	(27.20-32.10)	(29.10-34.01)	(34.90-43.60)
PD (mm)	mean±SD	8.42±0.76[a]	9.73±0.64[b]	11.07±0.81[c]	11.71±1.78[c]
	range	(7.65-10.01)	(8.95-11.00)	(9.58-12.20)	(8.49-14.01)
CRL (mm)	mean±SD	3.30±0.51[a]	17.83±2.66[b]	21.33±2.34[c]	30.94±1.61[d]
	range	(2.39-4.21)	(14.00-23.80)	(15.90-25.10)	(28.30-33.10)
BPD (mm)	mean±SD	6.13±0.13[a]	7.19±0.34[b]	8.05±0.49[c]	11.80±3.17[d]
	range	(5.92-6.30)	(6.85-7.91)	(7.20-8.51)	(8.60-16.20)
ONL (mm)	mean±SD	6.27±0.31[a]	7.21±0.69[b]	10.59±0.41[c]	18.24±1.66[d]
	range	(5.54-6.58)	(6.32-8.90)	(10.11-11.25)	(16.30-22.00)
TD (mm)	mean±SD	6.82±0.44[a]	10.05±0.61[b]	11.16±0.45[c]	14.38±2.16[d]
	range	(5.99-7.59)	(9.20-11.30)	(10.50-11.90)	(11.20-17.30)

GSD –gestational sac diameter; PD – placentome diameter; CRL –crown-rump length; BPD –biparietal diameter; ONL - occipital-nasal length; TD –trunk diameter: Mean values within a row marked with different alphabets differ significantly (P<0.05).

Table 2: Mean (±SEM) values of Doppler parameters in the umbilical artery during the first trimester of pregnancy in goats (n=14)

Parameters	Days of pregnancy			
	30	35	40	45
PSV (cm/s)	21.30±1.02[a]	22.44±0.71[a]	21.51±0.34[a]	26.65±0.93[b]
EDV (cm/s)	NO	NO	NO	NO
RI	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00
PI	2.98±0.09[a]	2.83±0.10[a]	2.75±0.11[a]	2.25±0.09[b]

PSV – peak systolic velocity; EDV – end-diastolic velocity; RI – resistance index; PI – pulsatility index NO – incalculable value: Values with different letters in a row are significantly different from one another (P<0.05).

Table 3: Pearson correlation coefficients (r) between the day of pregnancy and the biometric parameters of the embryo, placentomes diameter, gestational sac, and Doppler parameters in the umbilical artery in goats during the first trimester of pregnancy (n = 14)

Parameters	Correlation coefficients
Gestational sac diameter (GSD)	0.93**
Placentome diameter (PD)	0.75**
Crown-rump length (CRL)	0.95**
Biparietal diameter (BPD)	0.75**
Occipital nasal length (ONL)	0.92**
Trunk diameter (TD)	0.90**
Peak systolic velocity (PSV) in arterial vessels	0.51**
Pulsation index (PI) of arterial vessels	-0.60**

** = Significant at P<0.01.

Table 2 shows the mean values of Doppler blood flow parameters in the umbilical artery in the first trimester of pregnancy in goats. PSV from the 30th to the 40th day of pregnancy was similar, while on the 45th day the value of this parameter was significantly higher (P<0.05). On the other hand, the mean value of pulsatility index (PI) at the end of the study period (day 45 of pregnancy) was significantly lower (P<0.05) than in the period from the day 30 to the day 40 of pregnancy. Resistance index (RI) remained the same throughout the study period. No end-diastolic velocity (EDV) could be detected in any goat during the study period (Fig. 4).

Pearson's correlation coefficients between the day of pregnancy and the biometric parameters of the embryo, the diameter of the placentomes and gestational sac, and

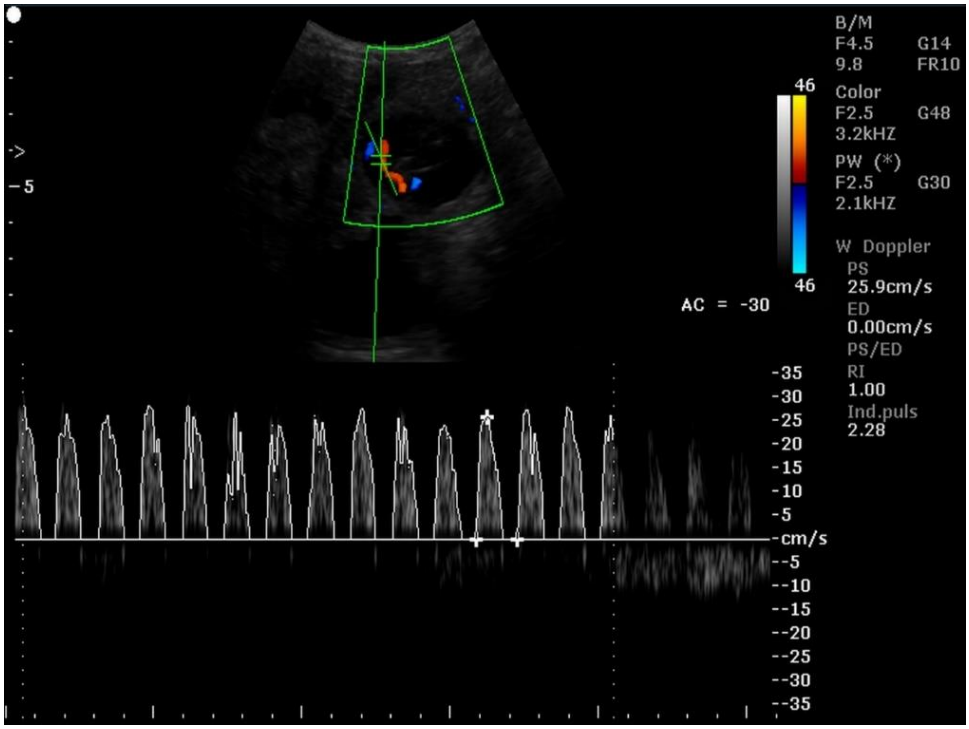

Fig. 4: Ultrasound image obtained using the pulse Doppler method showing the hemodynamic measurement of the umbilical artery on day 45 of pregnancy in a goat. PS - peak systolic velocity; ED - end-diastolic velocity; RI - resistance index; Ind. Puls - pulsation index.

Doppler parameters in the umbilical artery in goats during the first trimester of pregnancy are shown in Table 3. All parameters exhibited significant positive correlations with the day of pregnancy, except the pulsation index (PI) in the arterial vessels, which showed significant negative correlation with the day of pregnancy ($P<0.01$).

DISCUSSION

During an ultrasound examination, the pregnancy vesicle is visible as an anechoic spherical area with a hyperechoic embryo inside. However, type and frequency of the ultrasound head used in the examination can affect the visualization time and the measured diameter of the gestational sac (Devi *et al.*, 2019). The size of the gestational sac can be assessed using a transrectal transducer as early as 19 days after conception (Amer, 2010), while a more accurate method of assessing the gestational vesicle in goats is the transabdominal examination from the day 25 of pregnancy (Devi *et al.*, 2019). On the other hand, other authors suggest delaying the test until the 32nd day of pregnancy to avoid a false positive diagnosis resulting from early embryo loss (Jones *et al.*, 2016). In this experiment, a transabdominal transducer was used and the GSD diameter was measured from the 30th day of pregnancy and it was found that it was strongly correlated with the day of pregnancy. These results differ from those reported by Karadaev *et al.* (2016), who performed the first measurement on day 21 of pregnancy using a transrectal probe and diameter of the gestational sac was recorded as 10.3±2.7mm.

Crown-rump length is a measurement of the length of the embryo and fetus from the top of the head to the bottom of the buttock and is useful for determining growth rate and gestational age. CRL measurement in goats can be performed from days 21 to 49 of pregnancy (Karadaev *et al.*, 2018). However, some authors have shown the possibility of determining CLR of a goat fetus as early as on the 19th day of pregnancy and in the later stages of fetal development, reaching even the day 75 of pregnancy (Kuru *et al.*, 2018). In this study, the CRL was recorded from day 30 to day 45 of pregnancy and was strongly correlated with the day of pregnancy (R=0.95). Similarly, Gouda *et al.* (2021) and Kandiel *et al.* (2015) showed a high correlation between CRL and gestational age in sheep and goats, respectively. This indicates the reliability of this parameter in determining the gestational age in the first trimester of pregnancy. Interestingly, there was a 10-fold increase in CRL between 30 and 45 days of pregnancy, which was not observed by Karadaev *et al.* (2016). The differences are probably due to the individual breed characteristics of goats, type and frequency of the ultrasound probe used in the examination.

During the first trimester of pregnancy, internal organs such as the stomach, heart and lungs develop intensively. Therefore, during ultrasound examination of pregnancy in goats, the trunk diameter (TD) of the fetus is also analysed. First TD measurement can be performed from day 28 of pregnancy (Karadaev *et al.*, 2016). In this study, the abdominal or trunk diameter was defined as the diameter at the height of the stomach and liver or at the entrance of the umbilical cord to the fetus (Kandiel *et al.*, 2015) and measured from the day 30 of pregnancy. A significant increase in the TD was recorded from day 30 to day 45, which on the 45th day after insemination was 14.38±2.16 mm. However, Kandiel *et al.* (2015) reported the mean abdominal diameter of 16.32 mm in Shiba goats on day 42 of pregnancy. The differences seem to be due to the individual breed characteristics. The present study also showed a higher correlation between the day of pregnancy and TD (r=0.90) than in the case of BPD (r=0.75). Lee *et al.* (2005) also showed higher correlation between TD and

gestational age than BPD and CRL in Korean black goats. This indicates the reliability of TD in determining the gestational age in goats.

In the present study, BDP and ONL were recorded from the day 30 of pregnancy. On the other hand, Yazici *et al.* (2018) indicated the possibility of measuring the parameters of the fetal head only from the 37th day after conception. Similar observations to ours were made by Karadaev *et al.* (2016) and Nwaogu *et al.* (2010). In the present study, it was shown that ONL had higher correlation with pregnancy day (r=0.92) than BPD (r=0.75). However, Nwaogu *et al.* (2010) and Yazici *et al.* (2018) recorded relatively higher correlation between BPD (r=0.98; r=0.99) and ONL (r=0.97; r=0.99) with gestational age. These differences probably result from the characteristics of the studied breeds of goats.

In goats, the placentomes are perceived as concave structures with round shape. On the ultrasound image, depending on the imaging plane, they appear as gray image in the shape of the letter C or O (Rasheed, 2016). The previous studies show that during the first trimester of pregnancy, the mean diameter of the placentomes increases rapidly. Karadaev *et al.* (2018) indicated the mean diameter of placentomes as 9.00±1.4 mm on the 42nd day of pregnancy. These authors also suggested that the measurement of placentome diameter may be done from the day 42 of pregnancy, when they take a species-specific shape. In the present study, each placentome differed in size regardless of the age of the fetus. Similar observations were made by Karadaev *et al.* (2016; 2018). The increase in the size of the placentomes in the first half of pregnancy is mainly related to the expansion of the fetal villi and the associated network of capillaries. Rasheed (2016) showed wide differences in placentome size, depending on whether they are closer to the fetus or in the corners of the uterus. In the present study, the placentome diameter showed lower correlation with the day of pregnancy than most of the other parameters. Similar observations have been reported by Karen *et al.* (2009). According to Nwaogu *et al.* (2010), placentome diameter is the least reliable parameter in the assessment of gestational age in goats.

The changes in the size of the embryo-fetal parameters as well as the placentomes in goats in the first trimester of pregnancy reported in this study can also affect the changes in the haemodynamics of the umbilical artery. In the present study, the peak systolic velocity increased significantly in the last period (day 45) of the first trimester of pregnancy, while the end-diastolic velocity of blood flow in the umbilical artery was not recorded in any goat. Similar observations were made by Elmetwally and Meinecke-Tillmann (2018) in goats and sheep. In our view, the end-diastolic velocity of blood flow in the umbilical artery is not detected between weeks 4 and 12 of pregnancy. It should be noted that the PSV/EDV ratio was not determined due to the lack of measurement of the end-diastolic velocity during the first trimester of pregnancy. According to Stankiewicz *et al.* (2020), a significant decrease in the PSV/EDV ratio is found from the day 70 of pregnancy. Elmetwally *et al.* (2016a,b) have reported that the PSV/EDV ratio, which is one of the most important Doppler parameters, indicates about changes in end-diastolic velocity, especially in the second and third trimesters of pregnancy.

In the current study, the resistance index (RI) remained unchanged from day 30 to day 45 of pregnancy. Elmetwally *et al.*(2016a,b) indicated that the RI through week 8 of pregnancy remained unchanged at 1.00±0.00 and then dropped sharply to week 19. According to Serin *et al.* (2010), the RI increases from day 40 to day 55 of pregnancy, followed by a decrease.

Previous studies have shown that PI increases from day 39 to day 60 of pregnancy and then decreases to day 90 of pregnancy (Kumar *et al.*, 2015; Troisi *et al.* 2018). In the present study, the PI value did not change from day 30 to day 40 of pregnancy but decreased significantly at day 45. At the same time, the study also showed that the pulsation index was negatively correlated with the day of pregnancy. On the other hand, PSV showed a significant positive correlation with the day of pregnancy. Therefore, it can be assumed that along with the development of the fetus, the value of the umbilical artery blood flow pulsation index decreases in favour of an increased peak value of the systolic velocity in the first trimester of pregnancy. It is probably related to the intensive development of the fetus and the increased demand for nutrients.

Conclusions: The results of the current study showed that changes in the parameters of embryo-fetal morphometry, placentomes and blood flow parameters of the umbilical artery may be helpful in the assessment of fetal development during the first trimester of pregnancy in goats. Further research may be performed, especially with regard to any feto-placental abnormalities during pregnancy in this species.

Authors contribution: NW, TS, BB and JU conceived the idea and designed the study. NW, TS, BB and JU executed the experiment. BB, JU, NW and TS analyzed the data. All authors interpreted the data, reviewed the manuscript for important intellectual contents, and approved the final version.

REFERENCES

Airina RI, Nizam AM, Abdullah RB, *et al.*, 2011. Using fetal-heart size measured from ultrasound scanner images to estimate age of gestation in goat. J Anim Vet Adv 10:2528-40.

Amer HA, 2010. Ultrasonographic assessment of early pregnancy diagnosis, fetometry and sex determination in goats. Anim Reprod Sci 117:226–31.

Devi NB, Bhuyan D and Das A, 2019. Application of ultrasonic doppler technique for pregnancy diagnosis in local Assam goat. J Pharm Innov 8:1088-91.

Đuričić D, Grizelj J, Dobranić T, *et al.*, 2012. Reproductive performance of Boer goats in a moderate climate zone. Vet Arhiv 82:351-58.

Elmetwally MA and Meinecke-Tillmann S, 2018. Simultaneous umbilical blood flow during normal pregnancy in sheep and goat foetuses using non-invasive colour Doppler ultrasound. Anim Reprod 15:148.

Elmetwally MA, Rohn K and Meinecke-Tillmann S, 2016a. Doppler sonography is a useful method to assess the effects of maternal anxiety on intrauterine fetal growth in pregnant sheep and goats. Quality in Primary Care, 24:137-45.

Elmetwally MA, Rohn K and Meinecke-Tillmann S, 2016b. Non-invasive color Doppler sonography of uterine blood flow throughout pregnancy in sheep and goats. Theriogenology 85:1070-79.

Enginler SÖ, Dal GE, Çetin AC, *et al.*, 2021. Can gestational age be determined by placentome diameter, placentome blood flow pixel area and progesterone concentration during pregnancy in Kivircik ewes. Kafkas Univ Vet Fak Derg 27:6.

Garcia A, Rodriguez P, Masot J, *et al.*, 2014. Histomorphometric study of the goat stomach during prenatal development. Anim Sci J 85:951-62.

Gouda A, Aggag M and Kandiel MM, 2021. Ultrasound-guided fetometry in Egyptian sheep and she-goats. Benha Vet Med J 40:114-18.

Jones AK and Reed SA, 2017. Benefits of ultrasound scanning during gestation in the small ruminant. Small Rumin Res 149:163-71.

Jones AK, Gately RE, McFadden KK, *et al.*, 2016. Transabdominal ultrasound for detection of pregnancy, fetal and placental landmarks, and fetal age before day 45 of gestation in the sheep. Theriogenology 85: 939-45.

Kandiel MM, Watanabe G and Taya K, 2015. Ultrasonographic assessment of fetal growth in miniature "Shiba" goats (*Capra hircus*). Anim Reprod Sci 162:1-10.

Karadaev M, Fasulkov I, Vassilev N, *et al.*, 2016. Ultrasound monitoring of the first trimester of pregnancy in local goats through visualisation and measurements of some biometric parameters. Bulg J Vet Med 19:209-17.

Karadaev M, Fasulkov I, Yotov S, *et al.*, 2018. Determination of the gestational age through ultrasound measurements of some uterine and foetal parameters in Bulgarian local goats. Reprod Domest Anim 53:1456-65.

Karen AM, Fattouh ESM and Abu-Zeid SS, 2009. Estimation of gestational age in Egyptian native goats by ultrasound fetometry. Anim Reprod Sci 114:167–74.

Krzyżanowski A, Kwiatek M, Gęca T, *et al.*, 2019. Modern ultrasonography of the umbilical cord: Prenatal diagnosis of umbilical cord abnormalities and assessement of fetal wellbeing. Med Sci Monit 25:3170-80.

Kumar K, Chandolia RK, Kumar S, *et al.*, 2015. Doppler sonography for evaluation of hemodynamic characteristics of fetal umbilicus in Beetal goats. Vet World 8:412-16.

Kuru M, Oral H and Kulaksiz R, 2018. Determination of gestational age by measuring defined embryonic and foetal indices with ultrasonography in Abaza and Gurcu goats. Acta Vet Brno 87:357–62.

Lee Y, Lee JO, Cho HJ, *et al.*, 2005. Ultrasonic measurements of fetal parameters for estimation of gestational age in Korean black goats. J Vet Med Sci 67:497–502.

Lekatz LA, Luther JS, Caton JS, *et al.*, 2013. Impacts of maternal nutritional plane on umbilical artery hemodynamics, fetal and placentome growth in sheep. Anim Reprod 10:99-105.

Muhammad RS and Aziz DM, 2021. Determination of gestational age in Shami goats based on transabdominal ultrasonographic measurements of placentomes and uterine diameter. Al-Anbar J Vet Sci 14:30-41.

Nwaogu IC, Anya KO, Agada PC, *et al.*, 2010. Estimation of foetal age using ultrasonic measurements of different foetal parameters in red Sokoto goats (*Capra hircus*). Vet Arhiv 80:225-33.

Rasheed YM, 2016. Ultrasonic estimation of gestation age in goats via placentome diameter. Iraqi J Vet Sci 40:100–106.

Samir H, Karen A, Ashmawy T, *et al.*, 2016. Monitoring of embryonic and fetal losses in different breeds of goats using real-time B-mode ultrasonography. Theriogenology 85:207-15.

Serin G, Gökdal Ö, Tarımcılar T, *et al.*, 2010. Umbilical artery Doppler sonography in Saanen goat fetuses during singleton and multiple pregnancies. Theriogenology 74:1082-87.

Stankiewicz T, Błaszczyk B, Udała J, *et al.*, 2020. Morphometric measurements of the umbilical cord and placentomes and Doppler parameters of the umbilical artery through ultrasonographic analysis in pregnant sheep. Small Rumin Res 184:106043; https://doi.org/10.1016/j.smallrumres.2019.106043

Troisi A, Cardinali L, Orlandi R, *et al.*, 2018. Doppler evaluation of umbilical artery during normal gestation in sheep. Reprod Domest Anim 53:1517-22.

Yazici E, Ozenc E, Celik HA, *et al.*, 2018. Ultrasonographic foetometry and maternal serum progesterone concentrations during pregnancy in Turkish Saanen goats. Anim Reprod Sci 197: 93–105.

9.2. Załącznik 2.

Wojtasiak N., Stankiewicz T., Błaszczyk B., Szewczuk M.A. (2023): Doppler parameters in ductus venosus during the third trimester of pregnancy in goats. Animal Science and Genetics, 19, (3), 17-25.

ANIMAL SCIENCE AND GENETICS
Published by the Polish Society of Animal Production
vol. 19 (2023), no 3, 17-25
DOI: 10.5604/01.3001.0053.8570
Research Article. ***Open Access***

Doppler parameters in the ductus venosus during the third trimester of pregnancy in goats

Natalia Wojtasiak[1#], Tomasz Stankiewicz[1], Barbara Błaszczyk[1],

Małgorzata A. Szewczuk[2]

[1] West Pomeranian University of Technology in Szczecin, Faculty of Biotechnology and Animal Husbandry, Department of Animal Reproduction Biotechnology and Environmental Hygiene, 29 Klemensa Janickiego, 71-270 Szczecin, Poland
[2] West Pomeranian University of Technology in Szczecin, Faculty of Biotechnology and Animal Husbandry, Department of Monogastric Animal Sciences, 29 Klemensa Janickiego, 71-270 Szczecin, Poland

SUMMARY

The aim of the study was to assess the haemodynamics in the ductus venosus in the third trimester of pregnancy in goats. Blood flow parameters in the ductus venosus were measured using colour Doppler combined with the pulsed-wave Doppler technique. The wave spectrum of blood flow in the ductus venosus during the study period had a specific pulsating character with two visible acceleration phases. The study showed an increase in peak systolic velocity, end-diastolic velocity, resistance index, and pulsatility index in the last period of the third trimester of pregnancy. All Doppler parameters in the ductus venosus were correlated with the day of gestation. Blood flow parameters and the Doppler spectrum were shown to change during the third trimester of pregnancy in goats. The results may be useful for future study of the ductus venosus in foetal goats.

KEY WORDS: ductus venosus, ultrasonography, Doppler, pregnancy, goat

[#]Corresponding author e-mail: wojtasiak1natalia@gmail.com
Received: 8.07.2023 Received in revised form:19.07.2023
Accepted: 20.07.2023 Published online: 21.07.2023

INTRODUCTION

In recent years, the possibilities for ultrasound imaging of vascular haemodynamics have increased enormously. One of the uses of Doppler ultrasonography is to monitor embryo-foetal development (Kumar *et al*., 2015). The uteroplacental arteries (Ramírez-González *et al*., 2023), umbilical vessels (Serin *et al.*, 2010; Kumar *et al*., 2015; Wojtasiak *et al*., 2023), and foetal vessels, such as the aorta (Fasulkov *et al*., 2021) and foetal renal vessels (Stankiewicz *et al*., 2023), are examined using this non-invasive technique. It also provides information about the condition of the foetus and helps in the diagnosis of any abnormalities that may affect the survival of the foetus and newborn (Da Silva *et al*., 2018).

Ultrasonography of the liver is of great importance in these studies. The liver is one of the most important organs with active roles in various metabolic functions, such as the metabolism of proteins, lipids, carbohydrates, and vitamins A and B; synthesis of fibrinogen, globulins, clotting factors, and albumins; bile secretion; glycogen and fat storage; and excretion of urea and uric acid. It is also involved in other functions, such as detoxification of various metabolic waste products and drugs (Mahadeep *et al*., 2013). The foetal liver is the main haematopoietic organ during foetal development, with an important role supporting haematopoietic homeostasis (Lewis *et al*., 2021). It also occupies a privileged position in foetal circulation, as the first organ to which blood flows directly from the placenta (Sørensen *et al*., 2011). Oxygenated blood enters the goat foetal liver from the portal vein and umbilical veins, which form the intra-abdominal umbilical vein inside the foetus (Bejdić *et al*., 2021). The left lobe of the liver is supplied with nutrient-rich blood from the intra-abdominal umbilical vein, while half of the blood for the right lobe of the liver comes from the intra-abdominal umbilical vein and the other half from the nutrient-poor blood of the portal vein (Haugen *et al*., 2004; Sørensen *et al*., 2011). This division of blood flow between the portal vein and the intra-abdominal umbilical vein accounts for a functional dichotomy that can be modified by haemodynamic influences (Kiserud, 2003; Haugen *et al*., 2004). The foetal liver with its venous system are the main areas of interest in foetal circulation – particularly the blood flow through the ductus venosus and its connection with the intra-abdominal umbilical vein. The ductus venosus in postnatal life is known as the left branch of the portal vein, while in foetal development it forms the transverse sinus and originates in the intra-abdominal section of the umbilical vein (Mavrides *et al*., 2001; Bejdić *et al*., 2021). The ductus venosus of a ruminant foetus is a curved, trumpet-shaped vessel located in the central part of the liver, above the porta hepatis (Bejdić *et al*., 2021). It plays a key role in the distribution of highly oxygenated umbilical venous blood, some of which bypasses the liver parenchyma passing through the ductus venosus, caudal vena cava, foramen ovale, and left atrium and finally reaches the heart and brain (Seravalli *et al*., 2016). The waveform of the Doppler spectrum in the ductus venous is associated with changes in pressure and volume in the atria and is therefore important in monitoring any foetal condition that may affect future cardiac function (Seravalli *et al*., 2016). Doppler examination of foetal venous circulation provides valuable diagnostic information. It is also of great importance for arterial circulation. Quantitative assessment of venous flow provides information on foetal physiology, in terms of both the umbilical circulation and details of distribution in the liver and heart (Kiserud, 2003).

Doppler ultrasonography is increasingly used to assess blood flow through the ductus venosus when growth retardation, oedema, or congenital heart defects are suspected in the foetus (Seravalli *et al*., 2016). Some studies have shown an association between foetal chromosomal/cardiac abnormalities and abnormal ductus venous blood flow during the first trimester of pregnancy (Marvides *et al*., 2001). With these considerations in mind, data from ultrasound examination of the ductus venosus during the third trimester of pregnancy in goats can be regarded as important. The main objective of the study was to evaluate Doppler parameters in the ductus venosus in the third trimester of pregnancy in goats. It was hypothesized that the blood flow in the ductus venosus may change during the third trimester of pregnancy in goats.

MATERIALS AND METHOD

Experimental animals and management

The study was carried out on 14 Boer goats kept on a farm at the Experimental Department of the National Research Institute of Animal Production in Kołbacz (Poland: latitude 53'30"N). The goats were kept in an indoor system with pasture. Feeding was in accordance with the standards adopted for this species, based on green pasture and other roughage and concentrate, depending on the season. The goats had constant access to water and salt licks. The goats were healthy, multiparous, 5 to 6 years old, with similar body weight (75–80 kg). All goats were mated in September with the same buck. Gestation length was determined based on the day of mating. The effectiveness of mating was determined by transrectal ultrasound. After delivery, the date of conception was confirmed retrospectively, assuming that the pregnancy lasted 148 days (Stankiewicz *et al*., 2020; Wojtasiak *et al*., 2023). All goats were pregnant with twins. Pregnancy and delivery were uneventful, and all kids were born healthy.

Ultrasound examination

The haemodynamics in the ductus venosus were assessed in pregnant goats that had not previously been sedated (Stankiewicz *et al*., 2020; Wojtasiak *et al*., 2023). The examination was conducted in the third trimester of pregnancy, divided into three periods: 1 (100–110 days), 2 (120–130 days) and 3 (140–148 days). The examination was always performed by the same experienced and trained operator, in a quiet and dimly lit room. Transabdominal ultrasound examination was performed using an ultrasound scanner (EDAN U50 USG scanner) equipped with a sector probe with a frequency up to 5 MHz (model C352UB) and a linear probe with a frequency up to 9.4 MHz (model V742UB). Prior to the examination, the inguinal and caudal abdominal regions areas were shaved, the skin was cleaned with soap and water, and a sufficient amount of transmission gel was applied. The goats were kept in a standing position during the examination. After visualization of the foetus in B-Mode, vascularity was assessed using colour and pulsed Doppler. The ductus venosus was visualized in a sagittal or oblique transection of the foetal abdomen and examined at the isthmus, near its origin in the umbilical vein. This vessel was located in the central part of the liver and ran cephalad with increasing inclination in the same sagittal plane as the original direction of the umbilical vein. The following Doppler parameters were determined: peak systolic velocity (PSV), end diastolic velocity (EDV), PSV/EDV ratio, resistive Index [RI = (PSV − EDV) / PSV],

and pulsatility index [PI = (PSV −EDV) / mean velocity]. The flow angle during the test was kept as close as possible to a < 20 degrees, making appropriate adjustments to the angle when necessary. This procedure was performed twice, and the average for each vessel was calculated. Measurements were not recorded during maternal and foetal movements. In the case of signs of anxiety or changes in the maternal respiration rate, the examination was postponed until the body was seen to be in a stable position.

Statistical analysis

Statistical analysis of the results was performed. The results are presented as means ± SEM. The Shapiro–Wilk test was used to assess the normality of the data distribution. One-way repeated measures ANOVA was performed, where the grouping variable was gestational days, and the dependent variable was the Doppler parameter. Differences between the means for individual groups were analysed using a post hoc test. The Tukey test was used to verify the significance of differences at $P < 0.05$. The correlations between parameters and the day of pregnancy were calculated with the Pearson rank correlation coefficient (r). Statistical analyses were conducted using STATISTICA version 13.3, Stat Soft, Poland.

The results were analysed according to the following statistical model:

$$Y_{ijk} = \mu + \tau_i + \pi_j + \varepsilon_{ijk}$$

where:

Y_{ijk} – analysed trait
μ – overall mean
τ_i – fixed effect of i^{th} period of the third trimester of pregnancy *(i=1,..., 3)*
π_j – fixed effect of j^{th} Doppler parameter *(j = 1, 2, ..., 5)*
ε_{ijk} – error associated with each record (all error terms were assumed to be random, normally distributed and independent, with expectation equal to zero).

RESULTS

The waveform of the Doppler spectrum in the ductus venosus was pulsating. Two phases of acceleration (first and second peak) were visible in the Doppler spectrum. A representative image of blood flow in the ductus venosus is shown in Figure 1.

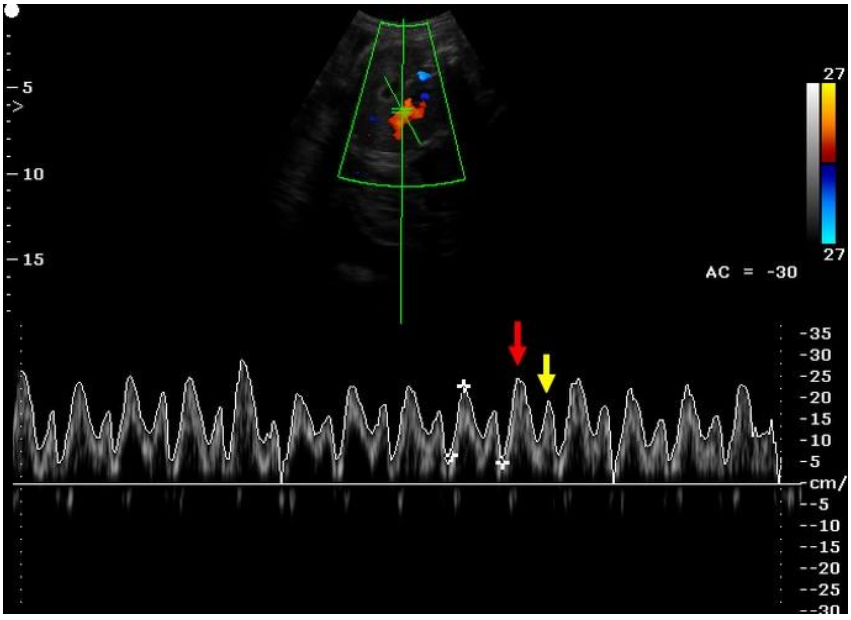

Figure 1. Ultrasound images showing Doppler blood flow velocity patterns from the ductus venosus in goats at 130 days of gestation. Red arrow – first peak systolic velocity; yellow arrow – second peak systolic velocity.

Table 1 shows the mean values of Doppler parameters in the ductus venosus in the third trimester of pregnancy in goats. Peak systolic velocities, PSV/EDV, and the resistive and pulsatility indexes were significantly higher in the final days of pregnancy than in earlier periods. However, the EDV value at the end of pregnancy was lower. All Doppler parameters examined in the ductus venosus were significantly correlated with the day of gestation.

Table 1.
Mean (± SEM) value of Doppler parameters in the ductus venosus in goat foetuses during the third trimester of pregnancy in goats (n = 14)

Parameters	Days of gestation		
	100-110	120-130	140-148
PSV I (cm/s)	22.42 ± 1.79[a]	29.96 ± 1.34[b]	38.43 ± 1.48[c]
PSV II (cm/s)	16.06 ± 1.79[a]	19.43 ± 1.55[a]	24.35 ± 0.85[b]
EDV (cm/s)	9.92 ± 1.25[a]	11.73 ± 0.82[a]	7.83 ± 0.94[b]
PSV/EDV	2.97 ± 0.29[a]	2.83 ± 0.27[a]	4.99 ± 0.62[b]
RI	0.62 ± 0.05[a]	0.60 ± 0.04[a]	0.75 ± 0.04[b]
PI	0.83 ± 0.08[a]	0.85 ± 0.06[a]	1.11 ± 0.08[b]

PSV I – first peak systolic velocity; PSV II – second peak systolic velocity; EDV – end-diastolic velocity; RI – resistive index; PI – pulsatility index
Values with different letters in a row are significantly different ($P < 0.05$).

The values of the correlation coefficients are presented in Table 2.

Table 2.
Pearson correlation coefficients (r) between the day of gestation and Doppler parameters in the ductus venosus in the third trimester of pregnancy in goats (n = 14)

	Parameters	Correlation coefficients	Significance level
Ductus venosus	PSV I	0.52	$P < 0.05$
	PSV II	0.75	$P < 0.01$
	EDV	-0.91	$P < 0.01$
	PSV/EDV	0.96	$P < 0.01$
	RI	0.92	$P < 0.01$
	PI	0.93	$P < 0.01$

PSV I – first peak systolic velocity; PSV II – second peak systolic velocity; EDV – end-diastolic velocity; RI – resistive index; PI – pulsatility index

DISCUSSION

The examination showed that the parameters of blood flow and the Doppler spectrum change during the third trimester. To our knowledge, this is the first study to describe haemodynamic changes in the liver venous system of foetuses in goats that have not previously been sedated. The ductus venosus, which originates in the umbilical vein, is the only venous vessel that regulates the nutrient supply via the blood from the umbilical veins between the liver and the heart. Blood flowing into the ductus venosus is

significantly accelerated towards the foramen ovale. This separates the bloodstream from the other venous stream entering the heart and allows nutrient-rich blood to reach the left ventricle instead of flowing through the tricuspid valve into the right ventricle (Seravalli *et al*., 2016). In the present study, the waveform of the Doppler spectrum in the ductus venosus was pulsating with two visible phases of acceleration. The available scientific literature contains no data on the waveform of the Doppler spectrum in the ductus venosus in goats. However, the results are in line with those obtained by Saemundsson *et al*. (2011) in sheep. They also showed that during the onset of tachycardia in sheep foetuses, the Doppler spectrum of the ductus venosus shows three repetitive reversals of flow velocity during atrial contraction (Seravalli *et al*., 2016). The course and waveform of the Doppler spectrum in the ductus venosus may explain the changes in atrial pressure and volume in the systolic and diastolic phases of the heart cycle (Saemundsson *et al*., 2011; Seravalli *et al*., 2016). Abnormal waveforms of the Doppler spectrum in the ductus venosus cause increased cardiac preload, abnormal cardiac function or structure, or increased cardiac afterload (Saemundsson *et al*., 2011; Seravalli *et al*., 2016). Therefore, an abnormal waveform requires careful examination of all potential cardiovascular factors. The scientific literature currently provides no data pertaining to values of Doppler indices in the ductus venosus of goats under physiological conditions. Most research describes the results of studies carried out in human foetuses (Nakagawa *et al*., 2012) and sheep foetuses (Mäkikallio *et al*., 2010). In the present study, peak systolic velocities increased significantly in the final days of the third trimester, while EDV values decreased significantly. Similar observations have been reported in human foetuses (Nakagawa *et al*., 2012).

In this study, for the first time, second peak systolic values were determined for the diastolic peak when the atrioventricular valves open during early passive diastolic ventricular filling. The results of human studies show that in the second phase of acceleration, blood flow velocity values increase during blood flow through the atria (Seravalli *et al*., 2016). In the present study, the mean PI values in the ductus venosus increased with gestational age. Similar observations were reported in studies conducted in sheep (Mäkikallio *et al*., 2010), whereas in humans, the average PI values during pregnancy are similar for most of the time, decreasing slightly at the end of pregnancy (Hofer, 2000; Nakagawa *et al*., 2012). In those studies, PI values were also found to be associated with gestational age (Hofer, 2000; Nakagawa *et al*., 2012). In the present study as well, significant correlations were found between these indices and gestational age. The results of the present study show that the mean RI values in the ductus venosus at the end of the third trimester, as in the case of the PI, were significantly higher than at the beginning of the third trimester of gestation in goats. Human studies have shown that for umbilical cord blood to enter the left atrium, it requires kinetic energy and high pressure to overcome the resistance of the ductus venosus and enter the foramen ovale (Kiserud and Kessler, 2023). This may explain the high values of Doppler parameters such as flow velocity, pulsatility index, and resistance index.

CONCLUSIONS

Summing up, the results presented in this paper may be useful in determining the reference values of haemodynamic ultrasound parameters of the ductus venosus in goats during pregnancy. The Doppler indices obtained indicate that haemodynamics in the ductus venosus change during the third trimester of pregnancy in goats, so these factors should be taken into account in assessment of hepatic venous system flow and heart function in goat foetuses.

REFERENCES

1. Bejdić P., Ćutuk A., Alić A., Čengić B., Avdić R., Tandir F., Hadžiomerović N., Mrvić V. (2021). Comparative Anatomical Studies on Ductus Venosus in Fetuses of Domestic Ruminants. Macedonian Veterinary Review, 44: 29-36. https://doi.org/10.2478/macvetrev-2020-0034
2. Da Silva PDA, Uscategui RAR, Santos VJC, Taira AR, Mariano RSG, Rodrigues MGK, Simões APR, Maronezi MC, Avante ML, Vicente WRR, Feliciano MAR. (2018). Qualitative and quantitative ultrasound attributes of maternal-foetal structures in pregnant ewes. Reproduction in Domestic Animals, 53: 725-732. https://doi.org/10.1111/rda.13163
3. Fasulkov IR, Karadaev M., Vasilev N., Hristov K., Fedev I. (2021). Doppler ultrasound measurements of the blood flow velocity in the fetal heart and aorta in Bulgarian White milk goats. Veterinary Medicine and Science, 7: 1297-1302. https://doi.org/10.1002/vms3.463
4. Haugen G., Kiserud T., Godfrey K., Crozier S., Hanson M. (2004). Portal and umbilical venous blood supply to the liver in the human fetus near term. Ultrasound in obstetrics & gynecology, 24: 599-605. https://doi.org/10.1002/uog.1744
5. Hofer M. (2000). Teaching Manual of Color Duplex Sonography. Georg Thieme.
6. Kiserud T. (2003): Fetal venous circulation. Fetal and Maternal Medicine Review, 14: 57-95. doi:10.1017/S0965539503001037
7. Kiserud T., Kessler J. (2023). The ductus venosus. Doppler Ultrasound in Obstetrics and Gynecology, 449-473. https://doi.org/10.1007/978-3-031-06189-9_28
8. Kumar K., Chandolia RK, Kumar S., Jangir T., Luthra RA, Kumari S., Kumar S. (2015). Doppler sonography for evaluation of hemodynamic characteristics of fetal umbilicus in Beetal goats. Veterinary World, 8: 412. doi: 10.14202/vetworld.2015.412-416
9. Lewis K., Yoshimoto M., Takebe T. (2021). Fetal liver hematopoiesis: from development to delivery. Stem Cell Research & Therapy, 12: 1-8. https://doi.org/10.1186/s13287-021-02189-w
10. Mahadeep S., Kamal S., Suri S., Devi J. (2013). Gross morphological studies on the liver of goat foetii (Capra hircus). Indian Journal of Veterinary Anatomy, 25: 39-40.
11. Mäkikallio K., Acharya G., Erkinaro T., Kavasmaa T., Haapsamo M., Huhta JC, Rasanen J. (2010). Ductus venosus velocimetry in acute fetal acidemia and impending fetal death in a sheep model of increased placental vascular resistance. American Journal of Physiology-Heart and Circulatory Physiology, 298: H1229-H1234. https://doi.org/10.1152/ajpheart.01299.2008

12. Mavrides E., Moscoso G., Carvalho JS, Campbell S., Thilaganathan B. (2001). The anatomy of the umbilical, portal and hepatic venous systems in the human fetus at 14–19 weeks of gestation. Ultrasound in Obstetrics and Gynecology: The Official Journal of the International Society of Ultrasound in Obstetrics and Gynecology, 18: 598-604.https://doi.org/10.1046/j.0960-7692.2001.00581.x
13. Nakagawa K., Tachibana D., Nobeyama H., Fukui M., Sumi T., Koyama M., Ishiko O., Hecher K. (2012). Reference ranges for time-related analysis of ductus venosus flow velocity waveforms in singleton pregnancies. Prenatal Diagnosis, 32: 803-809. https://doi.org/10.1002/pd.3908
14. Ramírez-González D., Poto Á., Peinado B., Almela L., Navarro-Serna S., Ruiz S. (2023). Ultrasonography of Pregnancy in Murciano-Granadina Goat Breed: Fetal Growth Indices and Umbilical Artery Doppler Parameters. Animals, 13: 618. https://doi.org/10.3390/ani13040618
15. Saemundsson Y., Johansson C., Wenling S., Gudmundsson S. (2011). Hepatic venous Doppler in the evaluation of fetal extrasystoles. Ultrasound in Obstetrics & Gynecology, 37: 179-183. https://doi.org/10.1002/uog.7684
16. Seravalli V., Miller JL, Block-Abraham D., Baschat AA. (2016). Ductus venosus Doppler in the assessment of fetal cardiovascular health: an updated practical approach. Acta Obstetricia et Gynecologica Scandinavica, 95: 635-644. https://doi.org/10.1111/aogs.12893
17. Serin G., Gökdal Ö., Tarımcılar T., Atay O. (2010). Umbilical artery doppler sonography in Saanen goat fetuses during singleton and multiple pregnancies. Theriogenology, 74: 1082-1087. https://doi.org/10.1016/j.theriogenology.2010.05.005
18. Sørensen A., Holm D., Pedersen M., Tietze A., Stausbøl-Grøn B., Duus L., Uldbjerg N. (2011). Left–right difference in fetal liver oxygenation during hypoxia estimated by BOLD MRI in a fetal sheep model. Ultrasound in obstetrics & gynecology, 38: 665-672. https://doi.org/10.1002/uog.9044
19. Stankiewicz T., Błaszczyk B., Chundekkad P. (2023). Ultrasound evaluation of biometric and Doppler parameters of kidneys in sheep in the prenatal and postnatal periods. Small Ruminant Research, 219. https://doi.org/10.1016/j.smallrumres.2022.106891
20. Stankiewicz T., Błaszczyk B., Udała J., Chundekkad P. (2020). Morphometric measurements of the umbilical cord and placentomes and Doppler parameters of the umbilical artery through ultrasonographic analysis in pregnant sheep. Small Ruminant Research, 184: 106043. https://doi.org/10.1016/j.smallrumres.2019.106043
21. Wojtasiak N., Stankiewicz T., Błaszczyk B., Udała J. (2023). Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43: 97-102. DOI: 10.29261/pakvetj/2022.067

9.3. Załącznik 3.

Wojtasiak N., Stankiewicz T., Udała J. (2020): Ultrasound examination of pregnancy in the domestic goat (Capra hircus) - a Review. Scientific Annals of the Polish Society of Animal Production, 16, (2), 65-78.

Scientific Annals of the Polish Society of Animal Production - Vol. 16 (2020), No 2, 65-78

DOI: 10.5604/01.3001.0014.2019

Review article

Ultrasound examination of pregnancy in the domestic goat (*Capra hircus*) - a review

Natalia Wojtasiak#, Tomasz Stankiewicz, Jan Udała

West Pomeranian University of Technology in Szczecin,
Faculty of Biotechnology and Animal Husbandry,
Department of Animal Reproduction Biotechnology and Environmental Hygiene
ul. Janickiego 29, 71-270 Szczecin, Poland

Ultrasonography (USG) for embryo-foetal foetometry is widely used in the management of goat breeding. Tissues and organs of the embryo/foetus are measured and evaluated to assess gestational age. Transrectal, transabdominal and transvaginal probes are used to perform the ultrasound examination. Technological advances, especially with regard to ultrasound image resolution, enable precise visualization of embryo-foetal structures in goats. The article reviews the foetometric measurements used in ultrasound examination of pregnancy in goats. Performing this examination during specific periods of gestation enables effective monitoring of embryonic and foetal growth and development. In addition to measurements of embryo-foetal structures, measurements of foetal-maternal structures such as the placentomes and umbilical cord are important as well. The role of ultrasound in monitoring goat pregnancy, both normal and pathological, was also emphasized.

KEY WORDS: ultrasound examination, embryo-foetal foetometry, pregnancy, goat

In recent years, ultrasonography has become one of the most important imaging techniques used to manage breeding of small ruminants. It is the preferred diagnostic tool because it is a relatively simple and non-invasive technique (Erdogan, 2012). The main practical applications of ultrasound in breeding of small ruminants are to confirm that mating has successfully resulted in pregnancy and to carry out precise biometric measurements, referred to as embryo-foetal foetometry (Santos et al., 2007; Karen et

#Corresponding author e-mail: wojtasiak1natalia@gmail.com
Received: 2.03.2020
Accepted: 5.05.2020

al., 2009; Samir et al., 2016). Available reports describe this diagnostic imaging as one of the most important elements of monitoring pregnancy in goats (Padilla-Rivas et al., 2005; Raja-Khalif et al., 2014). It is a valuable and practical method for monitoring embryonic and foetal development in most mammals, including goats. This paper presents a review of currently used ultrasound measurements of embryonic and foetal structures and their potential practical uses in monitoring the course of pregnancy in goats.

Pregnancy ultrasonography in goats - the most important parameters of embryo-foetal biometry

Ultrasound examination can be used to determine gestational age (GA) based on foetal development and thus to determine the date of parturition (Karen et al., 2009; Raja-Ili Airina et al., 2011). In goats, ultrasound imaging for the purpose of performing foetal biometric measurements is primarily used to assess gestational age (Padilla-Rivas et al., 2005; Raja-Ili Airina et al., 2011; Yazici et al., 2018). Ultrasound foetometry involves measurement of selected anatomical structures of the uterus and embryo/foetus, as well as assessment of foetal growth in successive stages of gestation.

Foetometric measurements include the following parameters:
– uterine lumen diameter – ULD (Karadaev et al., 2016)
– crown-rump length – CRL (Kumar et al., 2015b; Rasheed, 2017)
– trunk diameter – TD (Kumar et al., 2015b)
– biparietal diameter – BPD (Roukbi, 2013)
– foetal orbit diameter – OD (Kandiel et al., 2015)
– heart longitudial (long) axis – HLA (Karadaev et al., 2016)
– heart transverse (short) axis) – HTA (Lee et al., 2005; Karadaev et al., 2016)
– foetal heart rate – FHR (Karen et al., 2009; Karadaev et al., 2016)
– chest diameter – CD (Kandiel et al., 2015)
– femur length – FemL (Kandiel et al., 2015, Zongo et al., 2018)
– tibia length – TL (Kandiel et al., 2015, Zongo et al., 2018)
– humerus length – HL (Abdelghafar et al., 2012)
– length of six thoracic vertebrae – L6TV (Kandiel et al., 2015)
– length of six lumbar vertebrae – L6LV (Kandiel et al., 2015)
– occipitonasal length – ONL) (Kandiel et al., 2015)
– inner placentome diameter – IPD (Karadaev et al., 2016)
– outer placentome diameter – OPD (Roukbi, 2013)
– umbilical cord diameter (UCD) and diameter of umbilical vessels (Lee et al., 2005)
– aortic diameter – AD (Karadaev et al., 2018)

Preparation of the female goat and ultrasound examination techniques

Early detection of pregnancy in goats is performed using brightness mode (B-Mode) ultrasonography in real time. The presence of a fertilized egg with an embryo and amnio-

tic fluid together with observation of the embryonic heartbeat clearly confirm pregnancy in ultrasound imaging during this period (Raja-Ili Airina et al., 2011; Samir et al., 2016; Anya et al., 2017). B-mode ultrasonography presents echogenicity specific to individual embryo-foetal structures in the form of images with darker or lighter areas (Kharche and Kouamo, 2015). Linear high-frequency probes used in early pregnancy diagnosis allow for precise imaging of individual foetometric parameters in transrectal examination (Kumar et al., 2015a). In goats, ultrasound foetometry can be performed using transrectal probes generating ultrasonic waves with a frequency of 5 to 10 MHz (Kumar et al., 2015a). Transabdominal probes with frequencies from 3.5 to 5 MHz (Raja-Ili Airina et al., 2011; Samir et al., 2016) and transvaginal probes with frequencies from 5 to 7.5 MHz may be used as well (Koker et al., 2012, Philip et al., 2017). During the ultrasound examination, pregnant females may stand (Koker et al., 2012), sit, or lie on their backs (Vinoles-Gil et al., 2010). For examination with a transrectal probe, first the faeces are removed from the rectum, and then a gel-coated ultrasound probe is inserted. The head of the probe is placed against the abdomen and its position is adjusted until the bladder becomes visible. At that time the uterine horns appear in the ultrasound image in the cranial position. Then, by moving the probe backward and forward and rotating it 90° to the right and left, the other elements of the reproductive system can be examined (Raja--Khalif et al., 2014; Samir et al., 2016).

For transabdominal ultrasound examination, the hair of the goat should be cut short on an area of 100-200 cm^2 on the right side just above the lower abdomen. The examination is then performed by guiding the probe head placed on this region of the skin (Suguna et al., 2008).

Transvaginal ultrasound in goats is usually performed in a standing position. The probe is coated with ultrasound gel and gently inserted into the vagina. First, the head of the probe is positioned dorsally at a 45° angle, and then moved forward cranially. By further rotating the head 90°, the reproductive system of the goat is visualized. To optimize the ultrasound image, the abdominal wall may be gently raised (Koker et al., 2012; Philip et al., 2017).

Ultrasound biometric features of the embryo and foetus in goats

Gestational age is estimated on the basis of measurements of embryonic and foetal structures obtained from ultrasound images. Determining gestational age by ultrasound foetometry has become a useful method in management of goat breeding, especially when the exact date of mating is unknown (Kandiel et al., 2015; Samir et al., 2016; Jones and Reed, 2017). Depending on the stage of pregnancy, different foetometric parameters can be visualized and measured.

In goats, pregnancy lasts about 150 days and can be divided into three trimesters. The first trimester lasts up to the 49th day of pregnancy, the second lasts from days 50 to 100, and the third begins on the 101st day of pregnancy (Karadaev et al., 2018). In the ultrasound image, pregnancy is confirmed by the presence in the uterus of anechoic amniotic

fluid in the fertilized egg and a hyperechogenic embryo (Suguna et al., 2008; Raja-Ili Airina et al., 2011; Raja-Khalif et al., 2014).

Sometimes it is also possible to visualize the embryonic heartbeat and to perform biometric measurements of the heart (Raja-Ili Airina et al., 2011; Mali et al., 2019). In goats, this type of examination can be performed from the 21st day of gestation (Suguna et al., 2008), and according to some authors as early as the 15th day of gestation (Yazici et al., 2018).

Uterine lumen diameter

The uterine lumen diameter is one of the first foetometric measurements taken during obstetric examination of goats. It can be measured from the 21st day of gestation until the end of the first trimester (Karadaev et al., 2016). The examination is performed by determining the maximum transverse diameter of the uterus on the ultrasound image (Martinez et al., 1998). In the later stages of pregnancy, uterine lumen diameter is measured in the embryonic region (Karadaev et al., 2016; Karadaev et al., 2018).

Crown-rump length

In the early stages of pregnancy, the structures of the embryo are not yet differentiated. In this case, crown-rump length is defined as the length of the entire embryo/foetus (Karadaev et al., 2018). In the later stages of gestation, CRL is measured from the top of the foetal skull to the end of the sacrum (Abdelghafar et al., 2007; Roukbi, 2013; Pati et al., 2016). When the foetus is in a curved position, crown-rump length is measured first from the head to the heart, and then from the heart to the end of the sacrum (Abdelghafar et al., 2011; Karadaev et al., 2018). CRL in goats can be measured from the 21st to the 49th day of gestation (Karadaev et al., 2018). Some authors, however, have shown that this parameter can be measured in the goat foetus as early as day 19 of gestation, and in the later stages of foetal development up to day 75 (Kuru et al., 2018). Table 1 presents CRL measurements in goats taken at various times during pregnancy.

Biparietal diameter, foetal orbit diameter, and occipitonasal length

During ultrasound examination in the first trimester of pregnancy in goats, it is also possible to determine the biparietal diameter of the foetal head. The cross-section of this structure should be visualized in the ultrasound image, because image of the foetal head in this projection must be symmetrical to enable an accurate and reliable measurement (Lee et al., 2005). BPD measurement is performed on the visualized closed contour of the skull and both well visualized eye orbits (Lee et al., 2005). Next, BPD length is measured from the outer to the inner surface of the upper part of the skull (Amer, 2008; Karadaev et al., 2018). In the ultrasound image of the foetal head, in addition to the biparietal diameter, it is also possible to measure the foetal orbit diameter and occipitonasal length. Accurate measurement of the foetal orbit diameter requires lateral visualization of the head so that the orbit is visible on the ultrasound image in

Table 1

Crown-rump length determined by ultrasound in the first and second trimester of pregnancy in goats of different breeds

Day of gestation	Crown-rump length (mm)	Breed	Author
19	5.3	Anglo-Nubian	Martinez et al., 1998
21	4.8	Bulgarian	Karadaev et al., 2018
	5,2	Bulgarian	Karadaev et al., 2016
	7	Shiba	Kandiel et al., 2015
25	12	Damascus	Karenet al., 2009
30	16	Gürcü	Kuru et al., 2018
	17	Abaza	
35	23	Saanen	Abdelghafar et al., 2011
	27.82	Osmanabadi	Pati et al., 2016
37	33	Jamnapari	Abubakar et al., 2016
40	35.9	Damascus	Amer, 2008
46	34.4	Saanen	Abdelghafar et al., 2007
60	65	Abaza	Kuru et al., 2018
	72	Gürcü	
75	92	Abaza	Kuru et al., 2018
	99	Gürcü	

the form of a spherical structure with a pronounced closed hyperechogenic contour (Lee et al., 2005; Nwaogu et al., 2010; Karadaev et al., 2018). Occipitonasal length is defined as the distance between the top of the head and the tip of the nose (Yazici et al., 2018). BPD (Table 2) in goats can be determined from the 30th day of gestation (Karen et al., 2009], and ONL from the 37th day (Yazici et al., 2018). Foetal orbit diameter in goat foetuses can usually be determined at about the 49th day of gestation (Karadaev et al., 2016), although some authors suggest that earlier imaging is possible (Kandiel et al., 2015; Yazici et al., 2018). Variation in the time when OD measurements can be made may be due to differences in the characteristics of specific breeds (Table 2).

Table 2

Biparietal diameter, occipitonasal length and orbit diameter of the foetus determined by ultrasound in the first and second trimester of pregnancy in goats of different breeds

Foetal biometric parameter	Day of gestation	Measurement (mm)	Breed	Author
BPD	37	7	Saanen	Yazici et al., 2018
	40	11.9	Damascus	Amer, 2008
	42	9.4	Bulgarian	Karadaev et al., 2016
		9.9	Bulgarian	Karadaev et al., 2018
	46	11.6	Saanen	Abdelghafar et al., 2007
	57	20	Sokoto	Nwaogu et al., 2010
OD	42	3	Shiba	Kandiel et al., 2015
	44	4	Saanen	Yazici et al., 2018
	49	6.5	Bulgarian	Karadaev et al., 2016
		6.4	Bulgarian	Karadaev et al., 2018
	57	6	Sokoto	Nwaogu et al., 2010
	60	9,5	Korean Black	Lee et al., 2005
ONL	37	10	Saanen	Yazici et al., 2018
	42	13.37	Shiba	Kandiel et al., 2015
	57	5	Sokoto	Nwaogu et al., 2010

BPD – biparietal diameter; OD – orbit diameter; ONL – occipitonasal length

Foetal trunk diameter and chest diameter

The foetal trunk is also analysed during ultrasound examination of pregnancy in goats (Table 3). Its diameter (TD) is determined in lateral imaging as the maximum length measured from the spine through the abomasum to the abdominal wall (Karadaev et al., 2016; Karadaev et al., 2018). Kandiel et al. (2015) define the trunk diameter as the diameter measured at the height of the stomach and liver or the entry of the umbilical cord to the

foetus. In the ultrasound image, TD measurements in goats are performed in the transverse or sagittal plane (Gosselin et al., 2018). The first TD measurement can be made from the 28th day of gestation (Karadaev et al., 2016).

The chest is another element of the ultrasound examination of the foetus in goats. Its diameter (CD) is defined as the distance between the ventral and dorsal border of the thoracic cavity, at the height of the centre of the heart (Kandiel et al., 2015). CD measurement can be performed from the 37th day of gestation (Yazici et al., 2018).

Table 3
Diameter of the trunk and chest determined by ultrasound in the first trimester of pregnancy in goats of different breeds

Foetal biometric parameter	Day of gestation	Measurement (mm)	Breed	Author
TD	28	6.8	Bulgarian	Karadaev et al., 2016
		7.1	Bulgarian	Karadaev et al., 2018
	40	12	Damascus	Karen et al., 2009
	42	16.32	Shiba	Kandiel et al., 2015
CD	37	10	Saanen	Yazici et al., 2018
	4	10.93	Shiba	Kandiel et al., 2015

TD – trunk diameter; CD – chest diameter

Foetal heart rate and the heart transverse (short) and longitudinal (long) axis

As the pregnancy develops, changes are observed in the echogenicity of the embryonic/foetal heart. These changes can be seen in the varied greyscale typical of B-mode ultrasound imaging of the structures forming the heart of the embryo/foetus (Raja-Ili Airina et al., 2011). Until the 30th day of gestation, the foetal heart is highly echogenic, which is seen in the ultrasound image as a distinct white colour. Between weeks 4 and 8, the echogenicity of this organ decreases, as seen in the whitish-grey colours of the ultrasound images. Between 8 and 12 weeks of gestation in goats, the foetal heart is seen as a clearly outlined grey shape. From weeks 12 to 16 it is greyish-black, and around the 21st week it appears black in ultrasound images (Raja-Ili Airina et al., 2011). According to Anya et al. (2017), imaging of the embryonic heartbeat in goats is first possible from the 23rd day of gestation, and its frequency can be measured from the 25th day of gestation (Karen et al., 2009). In

this case, simultaneous recording in M-mode (motion mode) is used. In this technique, the onset or more often the peak of the contraction serves as an indirect timestamp of the corresponding electrical event (Dancea et al., 2000). Therefore, FHR is measured as the distance between systolic waves and then calculated automatically by the software of the ultrasound apparatus (Karadaev et al., 2016).

The longitudinal (long) and transverse (short) axes of the foetal heart (Table 4) should be measured in the diastolic phase while the foetus is not making any movements of its own. The measurement is performed in cross-section and in a four-chamber view of the heart (Lee et al., 2005). The longitudinal and transverse axes of the foetal heart can be measured from the 42nd day of gestation (Karen et al., 2009).

Table 4
Longitudinal and transverse axis of the heart determined by ultrasound in the first and second trimester of pregnancy in goats

Foetal biometric parameter	Day of gestation	Measurement (mm)	Breed	Author
HTA	42	3	Shiba	Kandiel et al., 2015
	44	3	Saanen	Yazici et al., 2018
	49	4.4	Bulgarian	Karadaev et al., 2016
	60	7.4	Korean Black	Lee et al., 2005
HLA	42	5	Shiba	Kandiel et al., 2015
	49	6.1	Bulgarian	Karadaev et al., 2016
	60	10.4	Korean Black	Lee et al., 2005

HTA – Transverse axis of heart; HLA – Longitudinal axis of heart

Femur, tibia, and humerus length

In the second trimester of pregnancy in goats, more foetal anatomical structures are subjected to ultrasound examination. The length of the femur and tibia can be measured at this time. In the ultrasound image, the foetal limbs should be visible in a longitudinal cross--section, so they can be measured from one end of the femoral and tibial shaft to the other (Kandiel et al., 2015). In measuring the length of the humerus, imaging of the foetal heart

and scapula is helpful. The humerus is then measured according to zones of intense calcification (Abdelghafar et al., 2012). The first FemL, TL and HL measurements in goat foetuses can be made from the 56th day of gestation (Abdelghafar et al., 2012; Kandiel et al., 2015).

Also from the 8th week of gestation in goats, ultrasound foetometry can be used to measure the length of six thoracic vertebrae and six lumbar vertebrae (Kandiel et al., 2015).

Umbilical cord diameter and foetal aortic diameter

Ultrasound foetometry also includes measurements of the diameter of the umbilical cord (Table 5) and umbilical vessels (Lee et al., 2005). In goats, umbilical vessels are represented by two arteries and two veins (Kumar et al., 2015c; Elmetwally and Meinecke-Tillmann, 2018). UCD measurement is performed when the umbilical cord and the foetus are visible in the ultrasound image at the same time. Umbilical cord diameter can be measured in two places: at the entry to the foetus and at a distance of one centimetre from the entry (Lee et al., 2005; Yazici et al., 2018). Umbilical vessels should be measured in a perpendicular projection (at maximum magnification) and where

Table 5
Diameter of the umbilical cord and aorta determined by ultrasound in the first and second trimester of pregnancy in goats of different breeds

Foetal biometric parameter	Day of gestation	Measurement (mm)	Breed	Author
UCD	30	2.5	Damascus	Karen et al., 2009
	42	3.18	Shiba	Kandiel et al., 2015
	51	3.8	Saanen	Yazici et al., 2018
	56	6	Saanen	Abdelghafar et al., 2011
	57	4	Red Sokoto	Nwaogu et al., 2010
	60	3.4	Korean Black	Lee et al., 2005
AD	60	2.2	Korean Black	Lee et al., 2005
	77	3.2	Bulgarian	Karadaev et al., 2018

UCD – umbilical cord diameter; AD – aortic diameter

the umbilical cord is closest to the foetus. In this way, differences in diameter occurring along the length of the umbilical cord are taken into account (Lee et al., 2005; Kandiel et al., 2015). The first measurements of the umbilical cord can be made from the 30th day of gestation (Karen et al., 2009).

In goats between 60 and 135 days of gestation, the aortic diameter of the foetus can be measured (Table 5). The measurement is taken in a cross-section of the foetal aorta, along the longitudinal axis of the left ventricle, when the foetus is not making any movements on its own (Lee et al., 2005; Karadaev et al., 2018).

In ultrasound examination of pregnancy development, in addition to determining the biometric features of embryos and foetuses, it is also very important to make precise measurements of extra-foetal structures. The placenta is the organ that should be analysed to assess the course of the pregnancy.

In pregnant goats, the placenta is perceived as a concave structure with a round shape. In the ultrasound image, it appears as a grey image in the shape of the letter C or O, depending on the imaging plane (Rasheed, 2016). This is a characteristic image ascribed to placentomes, whose size in ultrasound imagine is determined by measuring two diameters. The first is the outer placentome diameter, between its two most distal parts

Table 6
Diameter of the placentomes determined by ultrasound in the first and second trimester of pregnancy in goats of different breeds

Foetal biometric parameter	Day of gestation	Measurement (mm)	Breed	Author
OPD	27	3	Damascus	Roukbi, 2013
	30	4	Saanen	Yazici et al., 2018
	35	7.5	Saanen	Rasheed, 2016
	42	9.3	Bulgarian	Karadaev et al., 2016
	57	14	Sokoto	Nwaogu et al., 2010
	60	14.7	Korean Black	Lee et al., 2005
IPD	42	3.8	Bulgarian	Karadaev et al., 2016
		3.5	Bulgarian	Karadaev et al., 2018

OPD – outer placentome diameter; IPD – inner placentome diameter

(Nwaogu et al., 2010; Karadaev et al., 2016), while the inner diameter is located in the anechoic part of the placentome (Karadaev et al., 2016). To obtain accurate measurements, it is necessary to measure the placentomes located near the foetus and to use the arithmetic mean of the measurements (Lee et al., 2005; Karadaev et al., 2018). In goats, the placentome diameters can first be measured from about the 30th day of gestation (Roukbi, 2013; Rasheed, 2016; Yazici et al., 2018) – Table 6.

Conclusions

The information presented here indicates that ultrasonography is a helpful diagnostic tool in management of reproduction in goats. The use of linear high-frequency ultrasound probes can confirm pregnancy at an early stage of its development. The most foetal biometric measurements can be made in the early and middle periods of gestation.

Implementation of this technique in practice allows tests to be performed in cyclic repetitions in the same embryos/foetuses. This is crucial for monitoring the growth and development of embryo-foetal and extra-foetal structures at every stage of pregnancy, both normal and pathological. The data presented in this study show that ultrasound provides an accurate and non-invasive way to monitor pregnancy in goats. Moreover, technological advances in the field of ultrasonography continue to improve mainly image resolution, which allows more accurate imaging of more details of the goat embryo and foetus, especially those of very small size.

REFERENCES

Abdelghafar R.M., Ahmed B.H., Bakhiet A.O. (2007). Ultrasonic measurements of crown-rump length and bi parietal diameter to predict gestational age in Saanen goats. Journal of Animal and Veterinary Advances, 6: 454–457.

Abdelghafar R.M., Ahmed B.H., Ibrahim M.T., Mantis P. (2011). Prediction of gestational age by transabdominal real-time ultrasonographic measurements in Saanen goats (*Capra hircus*). Global Veterinaria, 6: 346–351.

Abdelghafar R.M., Ahmed B.H., Abdelrahim M.S., Ibrahim M.T. (2012). The accuracy of gestational age predicted from femur and humerus length in Saanen goat using ultrasonography. Acta Veterinaria Brno, 81: 295–299.

Abubakar F., Kari A., Ismail Z., Rashid B.A., Haruna U.T. (2016). Accuracy of transrectal ultrasonography: in estimating the gestational age of Jamnapari goats. Malaysian Applied Biology Journal, 45: 49–54.

Amer H.A. (2008). Determination of first pregnancy and foetal age using ultrasonic measurements of different foetal parameters in red Sokoto goats (*Capra hircus*). Veterinaria Italiana, 44: 429–437.

Anya K.O., Ekere S.O., Ogwu D.O. (2017). Early pregnancy diagnosis using transabdominal ultrasonography in west African dwarf goats. Nigerian Veterinary Jounral, 4: 311–318.

Dancea A., Fouron J.C., Miró J., Skoll A., Lessard M. (2000). Correlation between electrocardiographic and ultrasonographic time-interval measurements in fetal lamb heart. Pediatric Research, 47: 324–328.

Elmetwally M.A., Meinecke-Tillmann S. (2018). Simultaneous umbilical blood flow during normal pregnancy in sheep and goat foetuses using non-invasive colour Doppler ultrasound. Animal Reproduction, 15: 148–155.

Erdogan G. (2012). Ultrasonic assessment during pregnancy in goats – a review. Reproduction in Domestic Animals, 47: 157–163.

Gosselin V.B., Volkmann D.H., Dufour S., Middleton J.R. (2018). Use of ultrasonographic fetometry for the estimation of days to kidding in dairy does. Theriogenology, 118: 22–26.

Jones A.K., Reed S.A. (2017). Benefits of ultrasound scanning during gestation in the small ruminant. Small Ruminant Research, 149: 163–171.

Kandiel M.M.M., Watanabe G., Taya K. (2015). Ultrasonographic assessment of fetal growth in miniature „Shiba" goats (*Capra hircus*). Animal Reproduction Science, 162: 1–10.

Karadaev M., Fasulkov I., Vassilev N., Petrova Y., Tumbev A., Petelov Y. (2016). Ultrasound monitoring of the first trimester of pregnancy in local goats through visualization and measurements of some biometric parameters. Bulgarian Journal of Veterinary Medicine, 19: 209–217.

Karadaev M., Fasulkov I., Stanimir Y., Atanasova S., Vasilev N. (2018). Determination of gestational age through ultasound measurements of some uterine and foetal parameters in Bulgarian local goats. Reproduction in Domestic Animals, 53: 1456–1465.

Karen A.M., Fattouh E.S.M., Abu-Zeid S.S. (2009). Estimation of gestational age in Egyptian native goats by ultrasonographic fetometry. Animal Reproduction Science, 114: 167–174.

Kharche S.D., Kouamo J. (2015). An overview of pregnancy diagnosis in small ruminants. Indian Journal of Animal Sciences, 85: 331–342.

Koker A., Ince D., Sezik M. (2012). The accuracy of transvaginal ultrasonography for early pregnancy diagnosis in Saanen goats: A pilot study. Small Ruminant Research, 105: 277–281.

Kumar K., Chandolia R.K., Kumar S., Pal M., Kumar S. (2015a). Two-dimensional and three-dimensional ultrasonography for pregnancy diagnosis and antenatal fetal development in Beetel goats. Veterinary World, 8: 835–840.

Kumar K., Chandolia R.K., Kumar S., Pal M., Kumar S., Pandey A.K. (2015b). Prediction of gestational age in beetal goats by ultrasonic fetometry. Indian Journal of Small Ruminants, 27: 35–44.

Kumar K., Chandolia R.K., Kumar S., Jangir T., Luthra R.A., Kumari S., Kumar S. (2015c). Doppler sonography for evaluation of hemodynamic characteristics od fetal umbilicus in Beetal goats. Veterinary World, 8: 412–416.

Kuru M., Oral H., Kulaksiz R. (2018). Determination of gestational age by measuring defined embryonic and foetal indices with ultrasonography in Abaza and Gurcu goats. Acta Veterinaria Brno, 87: 357–362.

Lee Y., Lee O., Cho J., Shin H., Choi Y., Shim Y., Choi W., Shin H., Lee D., Lee G., Shin S. (2005). Ultrasonic measurement of fetal parameters for estimation of gestational age in Korean black goats. Theriogenology, 67: 497–502.

Mali A.B., Amle M.B., Markandeya N.M., Kumawat B.L. (2019). Comparative efficacy of transrectal and trasnsabdominal ultrasonography for early diagnosis of pregnancyn and embryonic ageing in goats. Indian Journal of Small Ruminants, 25: 171–175.

Martinez M.F., Bosch P., Bosch R.A. (1998). Determination of early pregnancy and embryonic growth in goats by transrectal ultrasound scanning. Theriogenology, 49: 1555–1565.

Nwaogu I.C., Anya K.O., Agada P.C. (2010). Estimation of foetal age using ultrasonic measurements of different foetal parameters in red Sokoto gaots (Capra hircus). Veterinarski Arhiv, 80: 225–233.

Padilla-Rivas G.R., Sohnrey B., Holtz W. (2005). Early pregnancy detection by real-time ultrasonography in Boer goats. Small Ruminant Research 58: 87–92.

Pati P., Sahatpure S.K., Patil A.D., Kumar U., Jena B., Nahak A.K., Sharma L.P. (2016). Prediction of gestational age in Osmanabadi goats by ultrasonic measurement of crown-rump length. The Indian Journal of Veterinary Sciences & Biotechnology, 12: 75–78.

Philip L.M., Abhilash R.S., Francis B.P. (2017). Accuracy of transvaginal ultrasonography for early pregnancy diagnosis in attappady black goats. Malaysian Journal of Veterinary Research, 8: 35–41.

Raja-Ili Airina R.K., Mohad Nizam A.R., Abdullah R.B., Wan-Khadijah W.E. (2011). Using fetal-heart measured from ultrasound scanner images to estimate age of gestation in goat. Journal of Animal and Veterinary Advances, 10: 2528–2540.

Raja-Khalif R.I.A., Rahman M.M., Wan-Khadijah W.E., Abdullah R.B. (2014). Pregnancy diagnosis in goats by using two different ultrasound probes. The Journal of Animal and Plant Sciences, 24: 1026–1031.

Rasheed Y.M. (2016). Ultrasonic estimation of gestation age in goats via placentome diameter. The Iraqi Journal of Veterinary Medicine, 40: 100–106.

Rasheed Y.M. (2017). Assessment of gestational age in goats by real-time ultrasound measuring the fetal crown-rump length and bi-parietal diameter. The Iraqi Journal of Veterinary Medicine, 41: 106–112.

Roukbi M. (2013). The use of ultrasonography for early detection of pregnancy and measurement of some foetal pattern in Damascus goats. Iraq Journal of Vetereinary Sciences, 27: 35–44.

Samir H., Karen A., Ashmawy T., Abo-Ahmed M., El-Sayed M., Watanabe G. (2016). Monitoring of embryonic and fetal losses in different breeds of goats using real-time B-mode ultrasonography. Theriogenology, 85: 207–215.

Santos M.H.B., Rabelo M.C., Filho C.R.A., Dezzoti C.H., Reichenbach H.D., Neves J.P., Lima P.F., Oliveira M.A.L. (2007). Accuracy of early fetal sex determination by ultrasonic assessment in goats. Research in Veterinary Science, 83: 251–255.

Suguna K., Mehrotra S., Agarwal S.K., Hoque M., Singh S.K., Shanker U., Sarath T. (2008). Early pregnancy diagnosis and embryonic and fetal development using real time B mode ultrasound in goats. Small Ruminant Research, 80: 80–86.

Vinoles-Gil C., Gonzalez-Bulnes A., Martin G.B., Zlatar F.S., Sale S. (2010). Sheep and Goats. In: DesCoteaux L., Colloton J, Gnemmi G. (eds), Ruminant and Camelid Reproductive Ultrasonography. Willey-Blackwell, Hong Kong, pp. 181–210.

Yazici E., Ozenc E., Celik H.A., Ucar M. (2018). Ultrasonographic foetometry and maternal serum progesterone concentrations during pregnancy in Turkish Saanen goats. Animal Reproduction Science, 197: 93–105.

Zongo M., Kimsé M., Kulo E.A., Sanou D. (2018). Fetal growth monitoring using ultrasonographic assessment of femur and tibia in Sahelian goats. Journal of Animal & Plant Sciences, 36: 5763–5768.

9.4. Załącznik 4.

Oświadczenia współautorów o procentowym udziale w przygotowaniu publikacji

Szczecin, dnia 1.09.2023

Dziekan
Wydziału Biotechnologii i Hodowli Zwierząt
ZUT w Szczecinie
dr hab. inż. Arkadiusz Pietruszka, prof. ZUT
w/m

Oświadczenia współautorów pracy zespołowej wskazujące ich wkład do pracy doktorskiej mgr inż. Natalii Wojtasiak

Niniejszym oświadczam, że udział **mgr inż. Natalii Wojtasiak** w przygotowywaniu publikacji: 1. Wojtasiak N., Stankiewicz T., Błaszczyk B., & Udała J. (2023). Ultrasound Parameters of Embryo-Fetal Morphometry and Doppler Indices in the Umbilical Artery During the First Trimester of Pregnancy in Goats. Pakistan Veterinary Journal, 43:97-102, **wynosi 75%**

Wojtasiak
(podpis czytelny doktoranta)

Jednocześnie udział pozostałych współautorów w przygotowanie powyższej publikacji jest szacowany na:
dr hab. inż. Tomasz Stankiewicz, prof. ZUT: 10%
dr hab. inż. Barbara Błaszczyk, prof. ZUT: 10%
prof. dr hab. inż. Jan Udała: 5%

Błaszczyk Barbara

Szczecin, dnia 1.09.2023

Dziekan
Wydziału Biotechnologii i Hodowli Zwierząt
ZUT w Szczecinie
dr hab. inż. Arkadiusz Pietruszka, prof. ZUT
w/m

Oświadczenia współautorów pracy zespołowej wskazujące ich wkład do pracy doktorskiej mgr inż. Natalii Wojtasiak

Niniejszym oświadczam, że udział **mgr inż. Natalii Wojtasiak** w przygotowywaniu publikacji: Wojtasiak N., Stankiewicz T., Błaszczyk B., Szewczuk M.A. (2023). Doppler parameters ductus venosus during the third trimester of pregnancy in goats. Animal Science and Genetics, 3:17-25, **wynosi 75%**

Wojtasiak
(podpis czytelny doktoranta)

Jednocześnie udział pozostałych współautorów w przygotowanie powyższej publikacji jest szacowany na:

dr hab. inż. Tomasz Stankiewicz, prof. ZUT: 10% Stankiewicz
dr hab. inż. Barbara Błaszczyk, prof. ZUT: 10% Błaszczyk Barbara
dr hab. inż. Małgorzata A. Szewczuk, prof. ZUT: 5% M. Szewczuk

Szczecin, dnia 1.09.2023

Dziekan
Wydziału Biotechnologii i Hodowli Zwierząt
ZUT w Szczecinie
dr hab. inż. Arkadiusz Pietruszka, prof. ZUT
w/m

Oświadczenia współautorów pracy zespołowej wskazujące ich wkład do pracy doktorskiej mgr inż. Natalii Wojtasiak

Niniejszym oświadczam, że udział **mgr inż. Natalii Wojtasiak** w przygotowywaniu publikacji: Wojtasiak N., Stankiewicz T., Udała J. (2020). Ultrasound examination of pregnancy in the domestic goat (Capra hircus) - a Review. Scientific Annals of the Polish Society of Animal Production, 2:65-78, **wynosi 90%**

Wojtasiak
.....
(podpis czytelny doktoranta)

Jednocześnie udział pozostałych współautorów w przygotowanie powyższej publikacji jest szacowany na:

dr hab. inż. Tomasz Stankiewicz, prof. ZUT: 5% .....

prof. dr hab. inż. Jan Udała: 5% .....