AKADEMIA WYCHOWANIA FIZYCZNEGO IM. JERZEGO KUKUCZKI W KATOWICACH

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MOŻLIWOŚĆ ZASTOSOWANIA ULTRASONOGRAFII GŁÓWNYCH MIĘŚNI ODDECHOWYCH W ANALIZIE SZYBKOŚCI I WYTRZYMAŁOŚCI NASTOLETNICH PIŁKARZY NOŻNYCH

Autoreferat rozprawy na stopień doktora nauk o kulturze fizycznej w postaci zbioru opublikowanych i powiązanych tematycznie artykułów naukowych

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1. WPROWADZENIE

Piłka nożna wymaga od zawodników długotrwałej, intensywnej przerywanej aktywności z powtarzającymi się sprintami [1,2]. Jednymi z ważniejszych zdolności motorycznych rozwijanych podczas treningu piłkarskiego są wytrzymałość [1] i szybkość [2,3], co stanowi obiekt zainteresowania wielu naukowców, którzy poszukują możliwości ich optymalnego kształtowania [2–5].

Aktywność fizyczna jest bezpośrednio związana z czynnością układu oddechowego, a więc będzie się przekładać na szybkość [6] i wytrzymałość [7]. Trening fizyczny wpływa na zmiany w układzie oddechowym poprzez zmniejszenie oporu w drogach oddechowych, zwiększenie elastyczności płuc i rozszerzanie się pęcherzyków płucnych [7]. Jednymi z głównych mięśni oddechowych (RM) będących przedmiotem zainteresowań badaczy są przepona (DA) i mięśnie międzyżebrowe (IC). Wynika to z faktu, że DA wpływa na ruch płuc [8] i wykonuje ponad 60% pracy oddechowej podczas wdechu [9]. IC z kolei przyczyniają się do rozszerzania klatki piersiowej [10] oraz mogą zwiększyć objętość wdechową bardziej niż przy samej pracy DA [11]. Zmęczenie RM podczas nasilającego się wysiłku ogranicza zdolności wytrzymałościowe, a ich trening ma wpływ na wydajność wysiłku wytrzymałościowego [12–14]. Trening RM wpływa na wyniki testów obejmujących próby czasowe, czy też wydłuża czas wytrzymałości RM [15]. Z tego też względu uzasadnionym jest ocenianie i analizowanie RM w kontekście kształtowania wybranych zdolności motorycznych – w tym szybkości i wytrzymałości, gdyż są one bezpośrednio powiązane z układem oddechowym.

Obecnie najczęściej ocenia się RM w sposób pośredni poprzez analizę funkcjonowania całego układu oddechowego z wykorzystaniem spirometrii [16–20] jako złotego standardu oceny czynności oddechowej [21]. W literaturze zaleca się jednak, by badania spirometryczne

uzupełniać badaniami obrazowymi i/lub czynnościowymi [18]. Jedną z powszechniej stosowanych metod bezpośredniej analizy RM jest ultrasonografia, odnosząca się konkretnie do badanego mięśnia oddechowego. Od kilku lat do analizy mięśni – w tym RM – można wykorzystać elastografię fali poprzecznej (shear wave elastography), stosowaną do analizy właściwości mechanicznych tkanek w sposób ilościowy [22,23]. W literaturze rola ultrasonografii, a tym bardziej elastografii fali poprzecznej w kontekście oceny układu oddechowego nie jest do końca wyjaśniona.

Literatura potwierdza rzetelność wybranych parametrów ultrasonograficznych DA (tj. grubości [24–26], ruchomości [16,27,28], prędkości przemieszczenia [29,30], echogeniczności [31]). Zaledwie kilka prac analizowało rzetelność parametrów ultrasonograficznych IC (grubość, echogeniczność) [11,32–34]. W odniesieniu do rzetelności modułu ścinania DA i IC, można znaleźć jedynie pojedyncze badania na ograniczonej populacji [33,35,36].W dostępnej literaturze występuje duża rozbieżność w metodyce, co utrudnia ich poprawną interpretację w odniesieniu do użyteczności ultrasonografii RM i adekwatnego określenia jej roli w badaniach układu oddechowego. Tym samym, przed badaniami właściwymi niezbędne jest określenie metodologii badań ultrasonograficznych RM – głównie modułu ścinania – z pełną analizą rzetelności.

Według dostępnej literatury na ten moment nie ma badań analizujących rolę RM w kontekście parametrów szybkości i wytrzymałości u nastoletnich piłkarzy nożnych. Sugeruje się, że ćwiczenia DA powinny być kluczowym elementem w osiąganiu wyników sportowych, zapobieganiu urazom i rehabilitacji [37]. Badania ultrasonograficzne RM u nastoletnich sportowców mogą więc dostarczyć nowej wiedzy na temat fizjologii tych mięśni i potencjalnie wpłynąć na procedury treningowe, diagnostyczne, czy też prognostyczne. Z tego względu niniejsza praca stanowi zbiór publikacji poruszających zagadnienia, które najogólniej odnoszą się do: a) przybliżenia obecnego stanu wiedzy na temat ultrasonografii RM; b) ustalenia metodyki i określenia rzetelności badania ultrasonograficznego RM na grupie nastoletnich piłkarzy nożnych; c) wstępnego oszacowania stopnia powiązania parametrów ultrasonograficznych głównych RM z wytrzymałością i szybkością u nastoletnich piłkarzy nożnych.

2. CEL BADAŃ I PYTANIA BADAWCZE

Celem niniejszych badań była weryfikacja możliwości zastosowania ultrasonografii głównych RM w analizie szybkości i wytrzymałości na grupie nastoletnich piłkarzy nożnych. Z uwagi na tak postawiony cel pracy zdecydowano się na realizację trzy etapowego projektu. Na każdym z etapów postawiono więc ściśle określone cele szczegółowe, których przeprowadzenie pozwoli na realizację celu głównego.

W pierwszym etapie (ETAP I) postanowiono dokonać przeglądu systematycznego literatury nad wykorzystaniem ultrasonografii w ocenie RM i przełożenia tych wyników na funkcję układu oddechowego. Dla tego etapu badań postanowiono odpowiedzieć na następujące pytanie badawcze:

Jaka jest rola/przydatność ultrasonografii głównych RM do oceny układu oddechowego w świetle dostępnej literatury?

Biorąc pod uwagę, że parametry oddechowe zależą od wielu czynników, a ultrasonografia pozwala na bezpośrednią ocenę RM przyjęto również następującą hipotezę roboczą:

Parametry oddechowe bezpośrednio związane z siłą RM powinny wykazywać wyższy związek z parametrami ultrasonograficznymi RM w porównaniu z parametrami oddechowymi niezwiązanymi bezpośrednio z siłą RM.

Z kolei drugi etap badań (ETAP II) związany był z koniecznością ustalenia odpowiedniej metodyki badania ultrasonograficznego głównych RM i określenia jej rzetelności na grupie nastoletnich piłkarzy nożnych. W etapie drugim postawiono następujące pytanie badawcze:

Czy przyjęta metodyka badania modułu ścinania DA i IC nastoletnich piłkarzy nożnych podczas spokojnego oddechu odznacza się odpowiednią rzetelnością?

Ostatni etap projektu (ETAP III) miał na celu wdrożenie przyjętej metodyki badania ultrasonograficznego DA i IC oraz przeprowadzenie analizy zależności uzyskanych parametrów ultrasonograficznych z szybkością i wytrzymałością u nastoletnich piłkarzy nożnych w ramach badań pilotażowych. Tym samym, realizując etap trzeci postawiono następujące pytanie badawcze:

Czy parametry ultrasonograficzne głównych RM są powiązane z szybkością i wytrzymałością u nastoletnich piłkarzy nożnych?

Z uwagi na to, że aktywność fizyczna jest zależna od pracy układu oddechowego, przyjęto również następującą hipotezę roboczą:

Zdolności motoryczne takie jak wytrzymałość i szybkość są powiązane z grubością i elastycznością głównych RM (DA, IC) u nastoletnich piłkarzy nożnych.

2.1. PRZEDMIOT ROZPRAWY

Przedmiot rozprawy doktorskiej stanowi osiągniecie naukowe przedstawione w postaci zbioru trzech prac opublikowanych w czasopismach posiadających Impact Factor. Łączna wartość punktowa opublikowanych prac wynosi: IF=10,874; MNiSW 300 pkt.

Wykaz publikacji będących podstawą rozprawy doktorskiej:

 <u>Pałac M</u>, Rutka M, Wolny T, Podgórski M, Linek P. Ultrasonography in Assessment of Respiratory Muscles Function: A Systematic Review. *Respiration*. 2022;101(9):878-892. doi: 10.1159/000524785.
 IF: 3.966; MNiSW: 100

Autor korespondencyjny: Paweł Linek

Realizacja etapu I

 <u>Pałac M</u>, Linek P. Intra-Rater Reliability of Shear Wave Elastography for the Quantification of Respiratory Muscles in Adolescent Athletes. *Sensors (Basel)*. 2022 Sep 1;22(17):6622. doi: 10.3390/s22176622.

IF: 3.847; MNiSW: 100

Autor korespondencyjny: Paweł Linek

Realizacja etapu II

 <u>Pałac M</u>, Sikora D, Wolny T, Linek P. Relationship between respiratory muscles ultrasound parameters and running tests performance in adolescent football players. A pilot study. PeerJ. 2023 Apr 17;11:e15214. doi: 10.7717/peerj.15214.

IF: 3.061; MNiSW: 100

Autor korespondencyjny: Małgorzata Pałac

Realizacja etapu III

3. MATERIAŁ I METODY BADAWCZE

3.1. ETAP I

3.1.1 Projekt badań i ekstrakcja danych

W celu oceny związku pomiędzy parametrami ultrasonograficznymi RM i parametrami oddechowymi dokonano przeglądu artykułów naukowych. Wyszukiwania artykułów przeprowadzono z uwzględnieniem następujących baz danych: MEDLINE (PubMed), Scopus, Ovid SP, EBSCO Academic Search Ultimate oraz Web of Science. Strategię wyszukiwania oparto o strategię PICO (P – patient, I – intervention, C – comparator, O - outcomes). Kryterium włączenia były wszystkie opublikowane artykuły naukowe (w języku angielskim) analizujące relacje między parametrami ultrasonograficznymi RM a funkcjami oddechowymi.

W pierwszej kolejności analizowano tytuły i streszczenia wyszukanych artykułów. Po wstępnej selekcji artykuły zostały analizowane w całości. Jakość badań oceniono za pomocą Scottish Intercollegiate Guidelines Network (SIGN) z zalecanym poprawionym narzędziem diagnostycznym (QUADAS-2) (art. 1, tab. 1, str. 25).

3.1.2 Synteza danych

Badania zostały poddane syntezie jakościowej. Dane wyodrębnione ze wszystkich włączonych badań (autorzy, charakterystyka grupy badanej, mierzone parametry i kluczowe wyniki korelacji) zostały zestawione w tabelach lub załącznikach. Ze względu na zbyt małą ilość podobnych do siebie prac nie przeprowadzono meta-regresji. Korelacje we włączonych badaniach interpretowano jako nieistotne (0,00–0,10), słabe (0,10–0,39), umiarkowane (0,40–0,69), silne (0,70–0,89) lub bardzo silne (0,90–1,00).

3.2 ETAP II

3.2.1 Projekt i materiał badań

Grupę badaną stanowiło 10 nastoletnich piłkarzy nożnych z klubu piłkarskiego w średnim wieku 17,1 lat (± 0.29). Do oceny parametrów ultrasonograficznych DA i IC (grubość, moduł ścinania) wykorzystano ultrasonograf Aixplorer (wersja produktu 12.2.0, wersja oprogramowania 12.2.0.808, Supersonic Imagine, Aix-en-Provence, Francja) z głowicą liniową (2–10 MHz; SuperLinear 10-2, Vermon, Tours, Francja).

3.2.2 Procedury pomiarowe i analiza danych

Pomiary ultrasonograficzne RM zebrano w spoczynkowej pozycji leżącej (z prawą ręką umieszczoną pod głową), głowica została umieszczona w prawej przestrzeni międzyżebrowej (pomiędzy linią pachową przednią i środkową). Parametry zebrano w dwóch ustawieniach głowicy (poprzecznie i równolegle do żeber) podczas spokojnego oddechu w trybie elastografii fali poprzecznej (art. 2, ryc. 1, str. 3). Każdy z pomiarów wykonywano dwukrotnie przez jednego badacza. Powtórne badania zostały wykonane po 7 dniach. Z każdego obrazu ultrasonograficznego zebrano pomiary grubości i modułu ścinania RM. Do określenia modułu ścinania mięśni zastosowano narzędzie ilościowe Q-Box bezpośrednio w aparacie ultrasonograficznym (art. 2, ryc. 2, str. 4). Do oceny grubości korzystano z programu RadiAnt DICOM Viewer (Medixant, Poznań, Polska).

3.2.3 Statystyka

Do obliczenia rzetelności jednego badacza wykorzystano współczynnik korelacji wewnątrzklasowej (intraclass correlation coefficient - ICC) typu 3,1 (dla pojedynczego pomiaru) i typu 3,2 (dla wartości średniej z dwóch pomiarów). ICC interpretowano według następujących kryteriów: 1,00–0,75 (doskonały), 0,74–0,60 (dobry), 0,59–0,40 (umiarkowany) i poniżej 0,40 (słaby). Do obliczenia zgodności wykorzystano standardowy błąd pomiaru

(SEM = SD × $\sqrt{1 - ICC}$), współczynnik zmienności (coefficient of variation - CV) oraz test Blanda-Altmana (BA). Dane analizowano za pomocą oprogramowania STATISTICA 13.1 PL (Statsoft, Tulsa, OK, USA) oraz Excel 2013 (Microsoft Corporation, Redmond, Washington, DC, USA).

3.3 ETAP III

3.3.1 Projekt i materiał badań

Do oceny korelacji parametrów ultrasonograficznych RM z parametrami wytrzymałości i szybkości zaproszono nastoletnich piłkarzy nożnych spełniających kryterium włączenia. Kryterium włączenia do badań był: brak problemów zdrowotnych lub kontuzji w czasie badania, brak problemów związanych z układem oddechowym w wywiadzie, brak zabiegów chirurgicznych na klatce piersiowej, jamie brzusznej, obręczy biodrowej i/lub kręgosłupie. Pierwszego dnia wykonano test szybkości, drugiego test oceny wytrzymałości, tydzień później ultrasonografię. Przepływ badanych w projekcie przedstawiono na diagramie zamieszczonym w artykule nr. 3 (art. 3, ryc. 2 str. 6).

3.3.2 Pomiary ultrasonograficzne

Parametry ultrasonograficzne grubości i modułu ścinania zostały zebrane z wykorzystaniem ultrasonografu Aixplorer (wersja produktu 12.2.0, wersja oprogramowania 12.2.0.808; Supersonic Imagine, Aix-en-Provence, Francja). Do oceny parametrów grubości i modułu ścinania RM wykorzystano głowicę liniową (2–10 MHz; SuperLinear 10-2, Vermon, Tours, Francja), pomiary były wykonywane w trybie elastografii fali poprzecznej przez dwóch fizjoterapeutów. Uczestnicy byli badani w pozycji leżącej z prawą ręką umieszczoną pod głową, głowica znajdowała się pomiędzy linią pachową przednią i środkową, równolegle do żeber (art. 3, ryc. 1, str. 4). Pomiary zostały zebrane na końcu spokojnego wdechu i wydechu.

Zastosowana w tej części pracy metoda pomiarowa oceny grubości i modułu ścinania RM została szczegółowo opisana i poddana analizie rzetelności w artykule nr. 2. Natomiast ruchomość i prędkość przemieszczenia DA została badana w trybie M-mode z wykorzystaniem głowicy konweksowej (1–6 MHz, Cristal Curved XC6-1; Vermon, Tours, Francja) podczas maksymalnego wdechu, a następnie spokojnego wydechu. Uczestnik leżał na plecach, głowica została umieszczona pod prawym łukiem żebrowym po prawej stronie ciała.

Do obliczenia wartości modułu ścinania RM zastosowano narzędzie ilościowe Q-Box[™]. Parametry grubości mierzono za pomocą programu RadiAnt DICOM Viewer (Medixant, Poznań, Polska). Stosunek grubości i modułu ścinania mierzono również jako wartość końcowo-wdechową podzieloną przez końcowo-wydechową wartość ultrasonograficzną. Ruchomość DA została opisana jako pionowa odległość od minimalnego do maksymalnego punktu przemieszczenia DA podczas danego manewru oddechowego. Prędkość przemieszczenia DA jest interpretowana jako prędkość ruchomości DA.

3.3.3 Testy biegowe

Do analizy wytrzymałości i szybkości uczestników wykorzystano dwa testy biegowe za pomocą fotokomórek firmy Witty System (Microgate Bolzano, Włochy) z dokładnością do 0,01 sekundy. Testy biegowe zostały przeprowadzone przez asystenta trenera przygotowania motorycznego na boisku piłkarskim ze sztuczną nawierzchnią (w fazie przygotowań do kolejnego sezonu piłkarskiego). Przed wykonaniem testów przeprowadzono 20-minutową rozgrzewkę. Pierwszego dnia wykonano test szybkości, który polegał na jak najszybszym przebiegnięciu 30 m w linii prostej między fotokomórkami. Pomiary były automatycznie rejestrowane na 5 m, 10 m i 30 m. Każdy uczestnik biegł dwukrotnie (przerwa pomiędzy dwoma próbami wynosiła 2 minuty), analizowano średni wynik z obydwu prób. Następnego dnia oceniano wytrzymałość przy pomocy wieloetapowego testem biegu wahadłowego na 20 m (multi-stage 20-m shuttle run test - MSRT). Test wymagał biegania w jedną i drugą stronę między dwoma pachołkami oddalonymi o 20 m. Prędkość zmieniała się co minutę. Uczestnikom zalecono bieganie jak najdłużej w tempie sygnałów dźwiękowych. Do analizy wykorzystano parametr "Total" oraz VO_{2max}. Parametr "Total" określał całkowitą liczbę wykonanych 20-metrowych odcinków (podczas całego testu). VO_{2max} oszacowano na podstawie maksymalnej prędkości osiągniętej podczas testu za pomocą wcześniej opracowanego równania –24,4+6,0× maksymalna prędkość tlenowa (s) [38]. Według Altmann et al. [39] obydwa testy biegowe wykazują się wysokim poziomem rzetelności.

3.3.4 Statystyka

Dane analizowano za pomocą oprogramowania STATISTICA 13.1 PL (Statsoft, USA) oraz Excel (Microsoft Corporation, USA). Ze względu na brak normalności rozkładu w teście Shapiro-Wilka dokonano analizy nieparametrycznego współczynnika korelacji rang Spearmana. Wartość korelacji (R) interpretowano w następujący sposób: od 0 do 0,30 lub od 0 do -0,30 uznano za korelację słabą; 0,31 do 0,50 lub -0,31 do -0,50 umiarkowana korelacja; 0,51 do 0,70 lub -0,51 do -0,70 silna korelacja; a 0,71 do 1 lub -0,71 do -1 to bardzo silna korelacja [40]. Poziom istotności ustalono dla $p \le 0,05$. Wielkość próby określono za pomocą G*POWER (wersja 3.1.9.7, Universitat Kiel, Niemcy).

4. WYNIKI

4.1 ETAP I

Podczas przygotowywania przeglądu systematycznego znaleziono 4636 prac spełniających warunki wyszukiwania. Po przejściu całego procesu wykluczania poszczególnych artykułów ostatecznie uwzględniono 31 prac. Szczegółowy proces wykluczenia artykułów z przeglądu przedstawia rycina 1 w artykule nr 1 (art. 1, ryc. 1, str. 30).

Badania naukowe dotyczące powiązania parametrów oddechowych z parametrami ultrasonograficznymi RM obejmują głównie osoby po 50 roku życia, osoby z przewlekłą obturacyjną choroba płuc (POChP), osoby zdrowe lub pacjenci z chorobami nerwowomięśniowymi. Tylko dwie prace brały pod uwagę sportowców (art. 1, tab. 2, str. 26). Najczęściej badanymi parametrami ultrasonograficznymi RM była ruchomość (58% artykułów) i grubość DA (45% artykułów). Na podstawie pomiarów grubości obliczono współczynniki grubości DA (współczynnik grubości - thickening ratio, frakcja grubości thickening fraction, różnica grubości - thickening difference). Prócz powyższych parametrów uwzględniono również prędkość przemieszczania DA oraz grubość i echogeniczność IC. W większości badań parametry ultrasonograficzne były zbierane po prawej stronie ciała.

Wśród parametrów oddechowych najczęściej badano: maksymalne ciśnienie wdechowe (maximal inspiratory pressure - MIP) – 48% prac, natężoną pojemność życiową (forced vital capacity - FVC) – 45% prac, nasilona objętość wydechowa pierwszosekundowa (forced expiratory volume in one second - FEV1) – 32% prac. Szczegóły dotyczące częstości poddawania korelacji wybranych parametrów ultrasonograficznych z parametrami oddechowymi zawarto na rycinie 2 artykułu nr 1 (art. 1, ryc. 2, str. 31).

W większości uwzględnionych artykułów obliczano zależności między parametrami wykorzystując korelację Pearsona lub Spearmana. Zgodność między dwoma oceniającymi

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została ustalona na poziomie 81%. Żadna praca nie uzyskała (w pełni) niskiego ryzyka stronniczości. Szczegółowe wyniki testu QUADAS-2 przedstawiono w artykule nr 1 (art. 1, tab. 1, str. 25).

4.1.1 Grubość i echogeniczność RM

Grubość RM (głównie DA) była poddawana analizie korelacji zazwyczaj z takimi parametrami oddechowymi jak MIP, FVC, FEV1. Grubość DA mierzono głównie na końcu głębokiego wdechu i na koniec spojonego wydechu. W większości artykułów grubość DA (mierzona głównie podczas maksymalnego wdechu) korelowała dodatnio z FVC i FEV1 (umiarkowanie–silnie), z maksymalnym ciśnieniem wdechowym przez nos (sniff nasal inspiratory pressure - SNIP) i MIP (umiarkowanie) oraz z maksymalnym ciśnieniem wydechowym (maximal expiratory pressure – MEP) i pojemnością życiową (vital capacity – VC) (słabo-umiarkowanie). Grubość i echogeniczność IC nie była istotnie skorelowana z parametrami oddechowymi (FEV1 i FEV1%). Szczegóły wszystkich korelacji zostały przedstawione w tab. 3 w art. nr.1 str. 27 oraz w załączniku 1 w art. nr.1 str. 32.

4.1.2 Ruchomość, prędkość przemieszczania i współczynniki grubości DA

Współczynniki grubości DA (thickening ratio, thickening fraction, thickening difference) są wyliczane z grubości DA. Ruchomość DA była najczęściej korelowana z FVC, FEV1, MIP, FVC%, FEV1%, a mierzona w kilku różnych manewrach oddechowych. Ruchomość DA była istotnie umiarkowanie skorelowane z większością parametrów oddechowych. Współczynniki grubości DA (thickening ratio, thickening fraction, thickening difference) również były skorelowane (od słabego do silnego) z większością parametrów oddechowych. Dla lepszej przejrzystości wszystkie dane przedstawiono w tab. 4 w art. 1 str. 28-29 oraz załączniku 2 w art. 1 str. 33.

4.2 ETAP II

4.2.1 Poprzeczne ustawienie głowicy (poprzeczne do żeber)

Rzetelność dla pomiarów modułu ścinania DA i IC z jednego dnia (ICC_{3.1}) niezależnie od fazy oddechu była zazwyczaj doskonała, CV nie przekraczał 3%, nie wykryto błędu systematycznego w teście BA. Natomiast rzetelność średniej z jednego pomiaru po 7-dniowej przerwie (ICC_{3.1}) wahała się od doskonałej do dobrej dla DA i od umiarkowanej do słabej dla IC. Rzetelność dla wartości średniej z dwóch pomiarów w 7-dniowym przedziale czasowym (ICC_{3.2}) dla DA była doskonała, dla IC od umiarkowanej do doskonałej, CV wynosiło poniżej 8%, ale wykryto błąd systematyczny dla modułu ścinania DA na końcu spokojnego wdechu.

Rzetelność pojedynczych pomiarów grubości DA i IC (ICC_{3.1}) była doskonała z pierwszego dnia badań oraz od doskonałej do umiarkowanej w 7. dniu badań, CV dla obydwu dni nie przekraczało 7%. Stwierdzono ujemny błąd systematyczny dla grubości IC na końcu spokojnego wdechu i wydechu oraz dodatni błąd systematyczny dla grubości przepony na końcu spokojnego wydechu. Rzetelność z wartości średniej z dwóch pomiarów przy 7-dniowym interwale (ICC_{3.2}) wahała się od doskonałej do dobrej, a CV nie przekraczało 6,03%, nie wykazano błędu systematycznego. Wszystkie szczegółowe wyniki rzetelności w poprzecznym ustawieniu głowicy przedstawiono w tab. 1, art. 2, str. 5.

4.2.2 Wzdłużne ustawienie sondy (równoległe do żeber)

Rzetelność modułu ścinania DA i IC (na szczycie wdechu i wydechu) dla pojedynczego pomiaru zebrane pierwszego dnia (ICC_{3.1}) wahała się od doskonałej do dobrej, a podczas pomiaru w interwale 7 dni wahała się od słabej do umiarkowanej. Rzetelność wartości średniej z dwóch pomiarów z interwałem 7 dni (ICC_{3.2}) dla modułu ścinania RM była dobra.

W ustawieniu głowicy równolegle do żeber dla modułu ścinania CV wynosiło zawsze poniżej 4%, test BA nie wykazał błędów systematycznych.

Rzetelność pojedynczych pomiarów grubości DA i IC z pierwszego dnia badań (ICC_{3.1}) wahała się od średniej do doskonałej z CV poniżej 9,5%. Rzetelność z interwałem 7 dni była dobra dla DA i od słabej do umiarkowanej dla IC z CV poniżej 5,5%. Rzetelność wartości średniej z dwóch pomiarów grubości DA i IC z interwałem 7 dni (ICC_{3.2}) uległa poprawie (prócz grubości IC podczas wdechu), CV nie przekroczyło 6,5%. Nie wykryto błędów systematycznych dla grubości RM we wzdłużnym ustawieniu głowicy. Wszystkie wyniki rzetelności w podłużnym ustawieniu głowicy przedstawiono w tab. 2, art. 2, str. 6.

4.3 ETAP III

Do analizy zależności parametrów ultrasonograficznych DA i IC z wybranymi zdolnościami motorycznymi (szybkość i wytrzymałość) ostatecznie uwzględniono 22 młodych piłkarzy nożnych w wieku 17-18 lat (art. 3, ryc. 2, str. 6). Dane antropometryczne, parametry ultrasonograficzne oraz wyniki testu wytrzymałościowy i szybkości przedstawiono w tabeli 1 (art. 3, tab. 1, str. 7). Moduł ścinania DA na końcu spokojnego wdechu korelował umiarkowanie ujemnie z wynikiem szybkości na 10m. Współczynniki modułu ścinania DA i IC były ujemnie skorelowane z wynikami prędkości na 10m (umiarkowanie) i 30m (od umiarkowanie do silnie). Ruchomość DA była dodatnio skorelowana z wynikami szybkości na 5m (umiarkowanie) i 10m (silnie). Prędkość przemieszczania się DA była umiarkowanie dodatnio skorelowana z wynikami szybkości na 5m i 30m. Grubość RM nie wykazała istotnych korelacji z wynikami szybkości można znaleźć w tabeli 1. Nie wykazano istotnej korelacji pomiędzy wytrzymałością a parametrami ultrasonograficznymi RM (art. 3, tab. 3, str. 9).

	5	m	10	m	30	m
	R	р	R	р	R	р
Moduł ścinania						
Przepona na końcu spokojnego wdechu	-0.34	0.12	-0.49	0.02*	-0.24	0.29
Przepona na końcu spokojnego wydechu	-0.10	0.66	-0.14	0.55	0.10	0.66
Współczynnik przepony	-0.31	0.16	-0.48	0.02*	-0.41	0.06
Mięśnie międzyżebrowe na końcu spokojnego wdechu	-0.26	0.26	-0.39	0.08	-0.18	0.44
Mięśnie międzyżebrowe na końcu spokojnego wydechu	-0.13	0.58	-0.16	0.49	0.16	0.48
Współczynnik mięśni międzyżebrowych	-0.28	0.22	-0.47	0.03*	-0.54	0.01*
Grubość						
Przepona na końcu spokojnego wdechu	-0.07	0.75	-0.06	0.80	0.22	0.34
Przepona na końcu spokojnego wydechu	-0.27	0.23	-0.12	0.60	0.25	0.25
Współczynnik przepony	0.33	0.13	0.07	0.75	-0.03	0.91
Mięśnie międzyżebrowe na końcu	-0.19	0.42	-0.07	0.78	0.11	0.63
spokojnego wdechu						
Mięśnie międzyżebrowe na końcu	-0.08	0.74	-0.11	0.64	0.05	0.83
spokojnego wydechu						
Współczynnik mięśni międzyżebrowych	-0.04	0.86	0.14	0.56	0.07	0.76
M-mode						
Ruchomość przepony	0.46	0.04*	0.52	0.02*	0.26	0.27
Prędkość przemieszczania się przepony	0.42	0.06	0.34	0.15	0.42	0.07

Tabela 1. Wyniki korelacji parametrów ultrasonograficznych z wynikami testów szybkości

*istotny statystycznie p<0,05; R - współczynnik korelacji; p - wartość prawdopodobieństwa; współczynnik – wartość parametru na końcu wdechu/ na końcu wydechu

5. MOCNE I SŁABE STRONY PROJEKTU ORAZ IMPLIKACJE

5.1 Mocne strony

Główny cel badań wynikał z faktu, iż parametry ultrasonograficzne RM nie były wcześniej bezpośrednio analizowane w kontekście badania wytrzymałości i szybkości u młodych sportowców. Dokonano dogłębnej analizy literatury dotyczącej ultrasonografii RM. Dostępne dane z badań zostały uporządkowane w jednej pracy naukowej (art. 1). Wcześniejsze badania nie analizowały jeszcze rzetelności modułu ścinania oraz grubości DA i IC u dorastających sportowców. Natomiast te wykonane na innych grupach badanych oceniały rzetelność tylko jednego dnia. Wstępne badania wykazały, że niektóre parametry ultrasonograficzne RM mogą być związane ze zdolnościami motorycznymi takimi jak szybkość. Tym samym, te wstępne wyniki potwierdziły zasadność badania RM w aspekcie zdolności motorycznych. Ultrasonografia zapewnia nieinwazyjne i niedrogie badanie. Badania na grupie sportowców przy pomocy elastografii fali poprzecznej potwierdziły jej rzetelność i możliwość wykorzystania do oceny bezpośredniej RM.

5.2 Ograniczenia

Badania dotyczące powiązania parametrów ultrasonograficznych RM ze zdolnościami motorycznymi obejmowały małą grupę oraz osoby uprawiające tylko piłkę nożną. Z tego względu badania potraktowano jako wstępny raport. Uczestnicy nie byli analizowani pod względem zajmowanej pozycji na boisku. Poza tym pomiary ultrasonograficzne były zbierane podczas spokojnego oddechu. Jak wykazał własny przegląd systematyczny, ultrasonografia RM wydaje się być lepiej skorelowana z parametrami układu oddechowego podczas maksymalnego wdechu. Mimo tych danych zdecydowano się na wstępnym etapie dokonać pomiarów w trakcie spokojnego wdechu ze względu na trudności metodologiczne. W przyjętej metodyce podczas maksymalnego wdechu RM były niewidoczne przez przysłaniające je płuco, co uniemożliwiało zebranie pomiaru. Na potrzeby badania, wytrzymałość sportowców określono pośrednio na podstawie wielostopniowego testu biegu wahadłowego na 20m. Wyniku nie należy zatem interpretować jako bezpośredniego pomiaru wytrzymałości, ale jako jego szacunek. Poza tym w badaniach tych analizowano szybkość liniową a nie wielokierunkową, która ma większe znaczenie w piłce nożnej.

5.3 Implikacje do kolejnych badań

Przyszłe badania powinny zawierać szczegółowy opis metodologiczny, uwzględniający pozycje badanego, manewry oddechowego podczas zbierania pomiarów i nazewnictwo tych samym pomiarów. Metodyka podczas badań ultrasonograficznych powinna być bardziej ujednolicona. Warto rozważyć analizę innych RM biorących udział w oddychaniu. Ze względu na rozległą rolę DA nie tylko w układzie oddechowym badanie jej wydaje się mieć duży potencjał. W przyszłych badaniach można by uwzględnić osoby uprawiające różne dyscypliny sportu, a jeśli chodzi o piłkarzy nożnych uwzględnić pozycję na boisku.

8. WNIOSKI

- Parametry ultrasonograficzne RM są częściowo powiązane z parametrami oddechowymi. Nie ma jednak wystraczających dowodów, by używać ultrasonografii RM do całościowej oceny układu oddechowego. Aktualne wyniki sugerują, że pomiary ultrasonograficzne RM mogą wyłącznie uzupełniać spirometrię, a dokładna rola RM w ocenie układu oddechowego wymaga dalszych badań.
- Przyjęta metodyka oceny grubości i modułu ścinania RM może być wykorzystana u
 nastoletnich sportowców. Na tym etapie trudno jednoznacznie określić lepsze położenie
 głowicy (poprzeczne vs. podłużne). Należy nadal poszukiwać rozwiązań pozwalających
 na rzetelną ocenę RM podczas maksymalnego wdechu i wydechu, gdyż może to
 pozwolić na pełniejszą analizę w kontekście szybkości i wytrzymałości.
- Badania pilotażowe wykazały, że moduł ścinania RM, ruchomość i prędkość przemieszczania DA są związane z szybkością i niezwiązane z wytrzymałością u dorastających piłkarzy nożnych. Obecny stan wiedzy nie pozwala nam jednak jednoznacznie określić jaką rzeczywiście rolę mogą pełnić parametry ultrasonograficzne RM w analizie szybkości i wytrzymałości u młodych sportowców. Z tego względu konieczne są dalsze badania na liczniejszej i mniej jednorodnej grupie nastoletnich sportowców.

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10. KOPIE PUBLIKACJI BĘDĄCE PODSTAWĄ ROZPRAWY DOKTORSKIEJ

10.1 Artykuł nr 1

Respiration

Systematic Review and Meta-Analysis

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Ultrasonography in Assessment of Respiratory Muscles Function: A Systematic Review

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Keywords

Ultrasonography · Pulmonary function · Respiratory muscles · Diaphragm

Abstract

Introduction: The purpose of this study was to evaluate the potential utility of respiratory muscles ultrasound (US) imaging for assessing respiratory function and identify US variables that best correlate with pulmonary parameters. Materials and Methods: A search of 5 databases was conducted. Initially, there was no language, study design, or time frame restrictions. All studies assessing the relationship between pulmonary and US parameters were included. Two reviewers independently extracted and documented data regarding to examined population, age, gender, health condition, methodology, US, and pulmonary function measurements. All studies were qualitative synthesis. Results: A total of 1,272 participants from 31 studies were included. Diaphragm thickness, diaphragm thickening ratio, and diaphragm excursion amplitude were mainly used as US parameters. Forced vital capacity, forced expiratory volume_{1sec}, and maximal inspiratory pressure were mainly used as pulmonary parameters.

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varied from negligible to strong (depend on examined population and methodology used). Data were not quantitatively synthesis due to high heterogeneity in terms of study design, population examined, and various pulmonary and US parameters. **Conclusion:** A strong relationship between US measurements and pulmonary parameters was demonstrated in some studies but not others. This review confirmed that US measurements can complement spirometry, but the exact role of the US remains to be confirmed. Further studies using standardized methodology are needed to obtain more conclusive evidence on the usefulness of US for assessing respiratory function. © 2022 S. Karger AG, Basel

The relationships between pulmonary and US parameters

Introduction

Spirometry and plethysmography are the most commonly used tools in the evaluation of respiratory system function [1–9]. These instruments require the use of dynamic functional trials [1, 8], potentially limiting their application. The primary parameters measured during

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Written permission to distribute the PDF will be granted against payment of a permission fee, which is based on the number of accesses required. Please contact permission@karger.com spirometry and plethysmography are forced vital capacity (FVC), vital capacity (VC), forced expiratory volume in the first second (FEV₁), and FEV₁/VC or FEV₁/FVC, which estimate the VC of the lungs. The strength of the respiratory muscles can be assessed with maximal inspiratory pressure (MIP) and maximal expiratory pressure (MEP) [2, 3, 7, 10–14]. Spirometry is the gold standard for assessing pulmonary function and is the most widely performed respiratory function test used clinically to diagnose and assess the severity of respiratory disorders [2, 4, 15–17]. Spirometry should also be complemented with imaging and/or functional tests [2] in certain clinical situations. It therefore seems reasonable to seek other methods of assessing pulmonary function to complement spirometry or replace it in some clinical conditions.

Ultrasound (US) can be successfully used to assess the respiratory muscles [4, 11, 18-22]. It is noninvasive, cost effective, and commonly available and compared to spirometry or plethysmography, does not always require such coordinated patient effort in performing respiratory manoeuvres [4, 11]. Overall, the relationship between pulmonary and US parameters has been evaluated [3, 4, 7]. The pulmonary parameters are mainly related to the thickness, change in thickness (ratios), and excursion of the diaphragm or echogenicity of certain respiratory muscles [11, 14, 19, 23-26]. However, there are conflicting opinions concerning the utility of using US to assess pulmonary function [23, 27-29]. Thus, the aim of this systematic review was to evaluate the potential utility of respiratory muscles US for assessing respiratory function. To reach this aim, we gathered studies that have assessed the relationship between pulmonary and US parameters. We also hypothesized that pulmonary parameters directly related to respiratory muscles strength should present a higher relationship with US parameters compared to pulmonary parameters not directly related to respiratory muscles strength. These data may be useful in clinical practice for screening out individuals in whom it is difficult to perform spirometry and to provide direction for future research in this area.

Materials and Methods

The original protocol of this systematic review was registered in the PROSPERO database (CRD42020211441).

Study Design and Search Strategy

A review was carried out of research articles evaluating the relationship between the US parameters of the respiratory muscles and pulmonary parameters. Original articles published in the English language without any restrictions on publication time were searched in MEDLINE (PubMed), Scopus, Ovid SP (Sports Medicine, American Journal of Physical Medicine & Rehabilitation, Drugs in R & D, Spine, MEDLINE, Strength and Conditioning Journal, Journal of Strength & Conditioning Research), EBSCO Academic Search Ultimate, and Web of Science. The search strategy was created based on the PICO strategy: P (patient) – adults and children (healthy and all diseases), athletes; I (intervention) – US of the respiratory muscles (e.g., diaphragm, abdominal muscles, intercostal muscles); C (comparator) – spirometry, respiratory muscle strength, plethysmography; O (outcomes) – relationship, correlation, association.

The systematic review took into account studies on healthy populations (children and adults), athletes, and people with any diseases affecting pulmonary function, regardless of age, gender, level of physical activity, and health status. All studies analysing the relationship between the US parameters of the respiratory muscles (e.g., diaphragm, abdominal muscles, intercostal muscles) and respiratory function were included.

Data Extraction

Initially, we reviewed the titles and summaries of potentially important articles. Database searches were conducted based on the terms specified in the search strategy. Two authors (M.P. and M.R.) selected publications for more detailed analysis. After the initial selection, the selected articles were read in full. All discrepancies in decisions to include or exclude studies were discussed. In case of doubt, the final selection of articles was made by consultation with other researchers (P.L. and T.W.). The following data were extracted from each article: research group, age, gender, and health condition. We also took into account important information on the methodology of the study and measured parameters (both US and lung function assessment). The data were collected and combined using the EndNote x9 (version 19.2.0.13018) programme. Duplicate searches were deleted. The references of selected studies were also searched for eligible studies. If any data were lacking, we contacted the authors. If this was not possible, the article was excluded.

Risk of Bias (Quality) Assessment

The quality of the studies was evaluated using the Scottish Intercollegiate Guidelines Network (SIGN) quality checklist according to the recommended revised diagnostic accuracy tool (QUA-DAS-2). Study quality was determined by testing its internal and external validity. The validity was evaluated using the QUADAS-2 tool. QUADAS-2 involves individualized scoring of seven components. Each of the seven steps is scored as "yes," "no," or "unclear." Two independent researchers (M.P. and M.R.) evaluated the articles. Uncertainties were resolved through discussion, or in the case of disagreement, third (T.W.) and fourth investigators (P.L.) resolved the problem. Irrespective of the outcome of the risk of bias, they were included in the review.

Data Synthesis

Data extracted from all included studies were tabulated, including the study authors and sample characteristics, the measurements of the outcome variables, and key results. All the identified studies were included in a qualitative synthesis and are presented in the tables and/or appendices. Initially, it was intended to synthesize the data quantitatively. However, because of high heterogeneity in terms of study design, population examined, and various pulmonary and US parameters, we could not perform a meta-



Fig. 1. Summary of included studies.

regression as it was not possible to select 10 similar articles. Any correlations in the included studies were interpreted as negligible (0.00–0.10), weak (0.10–0.39), moderate (0.40–0.69), strong (0.70–0.89), or very strong (0.90–1.00) [30].

Results

A total of 4,639 articles were identified during five database searches and hand searches of relevant articles (shown in Fig. 1). After removing duplicates and read-

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ing the titles and abstracts, 74 papers remained. Thirty of these were subsequently excluded due to lack of correlation between US parameters and respiratory function. Three papers were not in English, and another 10 papers were rejected because of lack of US measurements of respiratory muscles or spirometry. Full text analysis allowed 31 articles to be included in the review, according to the established criteria. The detailed process of inclusion of articles in the review is shown in Figure 1.

Table 1. Characteristics of the included studies
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Study	Characteristics of the correlation group	Patients, n	Age	Sex/ gender: male, %
Bennett et al. [4]	CP	36	13.9	58
Brown et al. [24]	Powerlifters and control group	20	26.5	100
Cordenas et al. [11]	Healthy	64	46	47
Carrie et al. [10]	Neuromuscular diseases (ALS or myotonic dystrophy)	45	56	42
Cohen et al. [21]	Healthy	10	36	50
Corbellini et al. [5]	COPD with/without healthy*	30 or 46*	*	76*
Dos Santos Yamaguti et al. [7]	COPD	54	62	78
Fantini et al. [25]	ALS	41	63	73
Henke et al. [31]	Facioscapulohumeral muscular dystrophy	14	53.4	64
Hiwatani et al. [19]	ALS	36	66.9	50
Holtzhausen et al. [26]	Students (athletes at the university level and leading a sedentary lifestyle)	55	21.2	62
Jung et al. [32]	Stroke patients	10	59.7	80
Kim et al. [39]	Stroke patients and healthy subject	94	48.8	61
Kim et al. [6]	Patients undergoing liver lobectomy	35	48.9	77
Kwon and Kim [12]	CP	43	7.7	61
Lim et al. [33]	COPD	10	79.8	100
Miyagi et al. [34]	Patients with heart failure	77	72	56
Noda et al. [27]	Neuromuscular diseases with/without control*	47 or 37*	58.9*	61*
Ogan et al. [29]	COPD	34	71	85
Pinto et al. [20]	ALS	42	58.9	48
Santana et al. [18]	Interstitial lung disease with/without control*	56 or 40*	55*	55*
Scott et al. [9]	Healthy	36	50.6	69
Smargiassi et al. [35]	COPD	23	71.2	100
Souza et al. [28]	Elderly woman	25	68.3	0
Souza et al. [36]	COPD	21	65.8	62
Spiesshoefer et al. [40]	Charcot-Marie-Tooth disease with/without being healthy*	19 or 38*	47*	32*
Spiesshoefer et al. [14]	Healthy	70	34	36
Summerhill et al. [37]	Patients with clinically suspected diaphragm paralysis	16	52.6	81
Wallbridge et al. [23]	COPD	20	71.5	80
Zanforlin et al. [38]	Patients with airway obstruction	124	53	65
Zhu et al. [41]	Spinal cord injury	30	39.6	90

CP, cerebral palsy; COPD, chronic obstructive pulmonary disease; ALS, amyotrophic lateral sclerosis. * Unclear information in the study.

Characteristics of the Included Studies

Most (64%) studies included participants over 50 years of age [5, 7, 9, 10, 18–20, 23, 25, 27–29, 31–38]. Two articles included children [4, 12]. The studies commonly included: people with chronic obstructive pulmonary disease (COPD) (23% of all included articles) [5, 7, 23, 29, 33, 35, 36], healthy individuals – 23% articles [9, 11, 14, 21, 26, 28, 39], and people with neuromuscular disease – 23% articles [10, 19, 20, 25, 27, 31, 40]. Other study populations appeared only once [6, 18, 24, 34, 37, 38, 41]. More details are shown in Table 1.

In all the included studies, US was performed on the diaphragm (right hemidiaphragm, 22 studies; left side, 1 study; both sides, 7 studies; lack of information, 1 study). In one of the studies, the intercostal muscles were also examined [23]. The diaphragm thickness (D_T) was used as a US parameter in 45% of the articles. Based on thick-

ness measurements available in 45% of articles, the thickness ratios (diaphragm thickening ratio [DTR], diaphragm thickening fraction [DTF], diaphragm thickening difference [DTD]) were calculated. Diaphragm excursion amplitude (DE_a) was collected in 58% of articles, and diaphragm excursion velocity (DE_v) was collected in three articles. With regards pulmonary parameters, the investigators mainly used parameters such as MIP (48% of articles) [7, 10–12, 14, 20, 24, 26, 28, 31, 34, 36, 37, 39, 40], FVC (45% of articles) [4, 7, 10–12, 14, 20, 25, 27, 32, 33, 35, 40, 41] and FEV₁ (32% of articles) [4, 7, 11, 12, 23, 29, 32, 33, 35, 41].

Due to the high number of studies included in the review, the variety of US and pulmonary parameters, and the need for clarity, the results are presented in two separate tables (shown in Tables 2, 3) and in the appendices (shown in Appendix 1 and 2; for all online supplementary

Study	US		Pulmonary	v paramet	ers				
	parameters	conditions	FVC	FEV1	MIP	SNIP	VC	MEP	FEV _{1%PRED}
Brown et al. [24]	D _T (right)	At end-tidal expiration			0.52* , 1			0.67* , 1	
Cordenas et al. [11]	D _T (right)	At the end of maximal inspiration	0.48 * ^{, 1}	0.48 * ^{, 1}	0.47* , 1	0.45 * ^{, 1}			
Fantini et al. [25]	D_T (both sides)	At the end of maximal inspiration At end-tidal inspiration	0.52* ^{, 2} 0.28 ²			0.53* ^{, 2} 0.30 ²			
Holtzhausen et al. [26]	D _T (right)	At end-tidal expiration			0.52* ^{, 2}				
Miyagi et al. [34]	D _T (right)	At the end of maximal inspiration At end-tidal expiration			0.24 * ^{, 1} 0.11 ¹		0.24* ^{, 1} 0.09 ¹	0.33* ^{, 1} 0.12 ¹	
Noda et al. [27]	D _T (both sides; average)	At end-tidal expiration	0.74 * ^{, 1}						nc
Ogan et al. [29]	D _T (right)	At end-tidal expiration At the end of maximal inspiration		0.19 ² -0.18 ²					
Pinto et al. [20]	D _T (right)	At the end of maximal inspiration	0.46 * ^{, 1}		0.44 * ^{, 1}	0.50 * ^{, 1}		0.51 * ^{, 1}	
Smargiassi et al. [35]	D _T (right)	At end-tidal expiration At the end of maximal inspiration At the end of maximal expiration					0.46* ^{,3} 0.65* ^{,3} 0.36* ^{,3}		
Spiesshoefer et al. [40]	D _T (right)	At the end of maximal inspiration At end-tidal expiration At end-tidal expiration	nc (data not shown)	nc (data not shown)				
Spiesshoefer et al. [14]	D _T (right)	At the end of maximal inspiration			0.30 * ^{,1}				
Wallbridge et al. [23]	D _T (right)	At end-tidal expiration		0.19 ⁴					-0.02 ⁴
	$2nd IC_{T} (left)$ $3rd IC_{T} (left)$ $2nd IC_{T} (right)$ $3rd IC_{T} (right)$ $2nd IC_{E} (left)$ $3rd IC_{E} (left)$ $2nd IC_{E} (right)$ $3rd IC_{E} (right)$ $IC_{E} (both sides)$	At end-tidal inspiration		$\begin{array}{c} 0.30^4 \\ 0.14^4 \\ 0.28^4 \\ 0.23^4 \\ 0.01^4 \\ 0.07^4 \\ -0.33^4 \\ -0.20^4 \end{array}$					$\begin{array}{c} 0.36^4 \\ 0.11^4 \\ 0.35^4 \\ 0.34^4 \\ -0.16^4 \\ -0.12^4 \\ -0.45^4 \\ -0.24^4 \\ -0.32^4 \end{array}$
Zhu et al. [41]	D _T (right) D _T (left)	At the end of maximal inspiration At end-tidal expiration	0.71* , ² 0.80* , ²	0.70* , ² 0.79* , ²					

D_T, diaphragm thickness; ICT, intercostal thickness; IC_E, intercostal echogenicity; FVC, forced vital capacity; FEV₁, forced expiratory volume in 1 s; SNIP, sniff nasal inspiratory pressure; VC, vital capacity; MEP, maximal expiratory pressure; MIP, maximal inspiratory pressure; FEV_{1%PRED}, forced expiratory volume in 1 s % predicted; nc, no correlation. * $p \le 0.05$ interpretation: negligible (0.00–0.10); weak (0.10–0.39); moderate (0.40–0.69); strong (0.70–0.89); very strong (0.90–1.00). ¹ Pearson correlation. ² Spearman correlation. ³ Beta regression. ⁴ Multiple linear regression.

material, see www.karger.com/doi/10.1159/000524785). The tables include those pulmonary variables that were correlated with US parameters in at least two separate studies. The appendices include those pulmonary variables that were correlated with US parameters in only one study. Table 2 and Appendix 1 show the studies in which the diaphragmatic thickness (a single US measurement)

was correlated with various pulmonary parameters. Table 3 and Appendix 2 show the studies in which the US ratios or excursion (US measurements under two different conditions) were analysed alongside various pulmonary parameters. A detailed description is given in synthesis of the results.

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Study	US		Pulmon	ary paramete	rs											
	parameters	conditions	FVC	FVC FE %PRED	N1 FE	ev, N PRED	AIP FE	c RV	RV/TLC IG	c/TLC S	NIP	V V	MEP	LLC IC	ER	2
Bennett et al. [4]	DE _a (right) DE _a (left)	DB	0.56*, ¹ 0.65*, ¹	0.52*, ¹ 0. 0.52*, ¹ 0.	52*, ¹ 0. 61*, ¹ 0.	58*, ¹ 48*, ¹										
Cordenas et al. [11]	DE _a (right) DTF (right)	DB At TLC/at FRC	0.54*, ¹ 0.59*, ¹	00	53*, ¹ 58*, ¹	00	.57*, ¹ .55*, ¹			00	.59*, ¹ .49*, ¹					
Carrie et al. [10]	DE _a (right) DE _a (right) DE _v (right)	FB SV SV	0.68 *, ²	0.75 * ^{, 2}		0	. 49 *, ² c c									
Corbellini et al. [5]	DE _a (right)	At TLC TB			7 7	0.74*, ¹ 0.80*, ¹								Ŷ).64* ^{,1} –0).63 *, ¹
Dos Santos Yamaguti et al. [7]	DE _a (right)	FB	0.60*, 2	0.	55 *, ²	1	0.11 ²	-0.60*, 2	° -0.76* ^{,2} 0	.55 *, ²		0	0.032	-0.28* ^{, 2} 0.	41*,2 0.4	43 *, ²
Fantini et al. [25]	DTR (both sides)	At TLC/at VT	-0.45*							1	-0.58*, ²					
Henke et al. [31]	DTR (right) DE _a (right)	At TLC/at FRC DB				00	.74*, ^L .70*, ^L									
Hiwatani et al. [19]	DTR (right)	At TLC/at FRC					0.0	15 ³								
Jung et al. [32]	DE _a (left)	DB	0.86 *, ²	0	70 *, ²											
Kim et al. [39] stroke patients	DTR (both sides)	At TLC/at FRC				0	.57*,1					-0.14 ¹				
Kim et al. [39] healthy subjects	DTR (both sides)	At TLC/at FRC				0	.32*,1					0.31 *, ¹				
Kim et al. [6]	DE _a (right)	DB).84 *, ²				
Kwon and Kim [12] Gl	DE _a (L)	TB DB	0.04 ¹ -0.11 ¹	0.0	0.43 ¹ 01 ¹		0.03 ¹ 0.20 ¹						-0.01 ¹ -0.18 ¹			
Kwon and Kim [12] GII	DE _a (L)	TB DB	-0.26 ¹ 0.02 ¹	9 9	0.50 ¹ 0.24 ¹	0 1	.42 ¹ 0.14 ¹					0 0).32 ¹).30 ¹			
Kwon and Kim [12] GIII	DE _a (L)	TB DB	0.10 ¹ 0.04 ¹	9 9).22 ¹).06 ¹	0	.11 ¹ 0.05 ¹						-0.26 ¹ -0.07 ¹			
Lim et al. [33] initial (during exacerbation phase)	DTF (right) DTF (left)	At TLC/at FRC	0.09 ² 0.08 ²	0.17 ² 0.3 0.11 ² 0.3	29 ² 0.4	44 ² 31 ²										
	DE _a (right) DE _a (left)	At TLC	0.59 ² 0.91 *, ²	0.20 ² 0.4 0.81 *, ² 0.4	43 ² 0.0	01 ² 36 ²										
Lim et al. [33] follow-up (during stable phase)	DTF (right) DTF (left)	At TLC/at FRC	0.39 ² 0.44 ²	0.50 ² 0.1 0.39 ² 0. 1	51 ² 0.	89*, ² 74*, ²										
	DE _a (right) DE _a (left)	At TLC	0.58 ² 0.86 *, ²	0.10 ² 0.5 0.62 ² 0.5	21 ² 0.3 56 ² 0.3	29 ² 31 ²										

Table 3. US ratios and excursion in relation to pulmonary parameters

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Table 3 (continued)																
Study	US		Pulmonary	/ parameters												
	parameters	conditions	FVC	-VC FEV1 %PRED	FEV ₁ %PRE	MIP	FEV ₁ / R FVC	>	3V/TLC	IC/TLC		V V	MEP	TLC	<u>∪</u>	ERV
Santana et al. [18]	DE _a (linear fitting) (richt)	DB	U).72 *,4	0.68*	4										
	DE _a (right)		U).73 *,4	0.70*	4										
	DTF (exponential fitting) (right)	At TLC/at FRC		0.224	0.23 ⁴											
	DTF (right)		0	0.24 ⁴	0.22 ⁴											
Scott et al. [9]	DE _a (right)	SV									0	0.034				
Smargiassi et al. [35]	DTD (right) DTD (right)	FB At TLC/at FRC	0.58* ^{,5} 0.49* ^{,5}	0.46 0.42	*,5 *,5		0.10 ⁵ 0 0.05 ⁵ 0	.28 ⁵ (0.54* ^{, 5} 0.46* ^{, 5}	0.49* ^{, 5} 0.53* ^{, 5}).58 *, ⁵).48*, ⁵		0.10 ⁵ 0.05 ⁵	0.51* ^{,5} 0.51* ^{,5}	
	DE_a (right)	At TLC/at FRC	0.05 ⁵	0.14			0.14 ⁵ 0	.175	0.01 ⁵	0.17 ⁵	0).14 ⁵	0	0.24 ⁵	0.32 ⁵	
Souza et al. [28]	DTR (right)	At TLC/at FRC				0.43* ^L										
Souza et al. [36]	DE _a (right)	At IC				-0.15 ¹										
Spiesshoefer et al. [40]	DE _a (right)	TB At TLC SV	nc (data not shown)			nc (dat: not shown)										
	DE _v (right)	TB SV	l													
	DTR (right)	At TLC/at FRC														
Spiesshoefer et al. [14]	DE _a (right) DE _v (right)	At TLC SV	0.63*, ¹ 0.43*, ¹			0.44*, ¹ 0.36*, ¹						00	0.47*, ¹ 0.34*, ¹			
Summerhill et al. [37]	DTF (both sides)	At TLC/at FRC				0.56 *, ⁶).66 *, ⁶				
Zhu et al. [41]	DE _a (right)	DB	0.70 *, ²	0.74	*, 2											
DE _a , diaphragm excursion am functional residual capacity/expira FVC [%] PRED, forced vital capacity % p maximal expiratory pressure; ERV, ¹ Pearson correlation. ² Spearman c	Jitude; DEv, diaphra; tory after TLC; DB, de redicted; FEV1, forcec expiratory reserve vol orrleation. ³ Polynom	gm excursion vel ep breathing; TB d expiratory volur lume. nc-no corre ial regression an	ocity; DTR, tidal breat ne in 1 s; FE elation; L, la alysis. ⁴ Reg	diaphragm th hing; FB, forc :V1 % _{PRED} , forc ck. * $p \leq 0.05$, iression analy	ickening r ed breathi ced expirat interpreta sis. ⁵ Beta r	atio; DTF, di ng; SV, volu ory volume tion: negligi egression. ⁶	aphragm th ntary sniffir in 1 s % pre ble (0.00–0 Multiple lir	nickening ig; MIP, m dicted; R ¹ i 10); weal	fraction; [laximal ins /, residual <(0.10–0.3 ssion.	DTD, diaph piratory pi volume; Sh 9); modera	ragm thic essure; IC NIP, sniff n ite (0.40–0	kening di , inspiratc asal inspir 0.69); stror	fference; ory capaci ratory pre ng (0.70⊣	TLC, tota ity; FVC, f sssure; VC 0.89); ver	l lung cap orced vita , vital capa / strong ((acity; FRC, l capacity; acity; MEP, 0.90–1.00).

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Table 4. Risk of bias - result of QUADAS-2

Study	Risk of bias				Applicabili	ty concerns	
	patient selection	index test	reference standard	flow and timing	patient selection	index test	reference standard
Bennett et al. [4]		?		\odot	\odot	0	
Brown et al. [24]		?	\odot		\odot	\odot	\odot
Cardenas et al. [11]	(C)	?	?	\odot	\odot	\odot	\odot
Carrié et al. [10]	(* *)	(* *)	()	(***)	\odot	\odot	\odot
Cohen et al. [21]	?	?	?	(<u>;</u>)	(<u>;</u>)	\odot	(:)
Corbellini et al. [5]		?	?		C	(C)	?
Dos Santos Yamaguti et al. [7]		?		?	\odot	\odot	\odot
Fantini et al. [25]		\odot	\odot	\odot	\odot	\odot	\odot
Henke et al. [31]		?	?	?	\odot	\odot	\odot
Hiwatani et al. [19]		?	\odot	\odot	\odot	\odot	\odot
Holtzhausen et al. [26]		?	\odot	\odot	\odot	\odot	\odot
Jung et al. [32]		?	?	\odot	\odot	\odot	\odot
Kim et al. [39]		?	?	?	\odot	\odot	\odot
Kim et al. [6]		?	\odot		\odot	\odot	\odot
Kwon and Kim [12]	٢	?	?	\odot	\odot	\odot	\odot
Lim et al. [33]	٢	٢	?		\odot	\odot	\odot
Miyagi et al. [34]		?	?	?	\odot	\odot	\odot
Noda et al. [27]		?	?	\odot	\odot	\odot	\odot
Ogan et al. [29]	(* *)	?	?	?	\odot	\odot	\odot
Pinto et al. [20]	()	?	\odot	\odot	\odot	\odot	\odot
Santana et al. [18]	()	?	?	\odot	\odot	\odot	\odot
Scott et al. [9]	()	?	\odot	٢	\odot	\odot	\odot
Smargiassi et al. [35]	()	?	\odot	\odot	\odot	\odot	\odot
Souza et al. [28]		?	\odot	\odot	\odot	\odot	\odot
Souza et al. [36]		?	?	?	\odot	\odot	\odot
Spiesshoefer et al. [40]		?	?	?	\odot	\odot	\odot
Spiesshoefer et al. [14]	(P)	\odot	?	?	?	Ū	$\overline{\odot}$
Summerhill et al. [37]	Ċ	?	?	?	\odot	Ō	\odot
Wallbridge et al. [23]	ē	?	?	?	õ	õ	õ
Zanforlin et al. [38]	ē		\odot	?	õ	õ	õ
Zhu et al. [41]	<u>.</u>	?	?				0

(i), low risk; (i), high risk; ?, unclear risk.

Risk of Bias

Concordance between the two evaluators was set at 81%. None of the papers received a low-risk result in any domain. The highest proportion (96%) of low risk of bias was found in all parts of the applicability concerns assessment. The results of the QUADAS-2 are described in Table 4.

Synthesis of Results

The relationship between US and pulmonary parameters was calculated using different statistical approaches. Most included articles used Pearson (35% of articles) or Spearman correlation (29% of articles). Some papers used regression analysis (23% of articles), whereas four papers did not provide information on the tool used.

US Thickness or Echogenicity Measurements

There were 13 studies in which D_T , intercostal muscle thickness (1 study), or intercostal muscle echogenicity (1 study) were correlated with MIP (7 studies), FVC (6 studies), FEV₁ (4 studies), sniff nasal inspiratory pressure (SNIP) (3 studies), MEP (3 studies), FEV1_{%PRED} (3 studies), and VC (2 studies) (Fig. 2). In ten studies, only the right hemidiaphragm was analysed, and in three studies, both sides were analysed. Intercostal muscle thickness and echogenicity were measured on both sides of the body [23]. D_T was measured at the end of deep inspiration (nine studies), at the end of tidal inspiration (one study), at the end-tidal expiration (one study). Intercostal muscle



Fig. 2. Percentage distribution of articles correlating pulmonary parameters with a selected US measurement (e.g., D_T and MIP 50% mean that out of 14 articles where D_T was calculated, in 7, the D_T was correlated with MIP).

thickness and echogenicity were only measured at the end of tidal inspiration (shown in Table 2).

The relationship between D_T at maximal inspiration and FVC was statistically significant in all studies (with the exception of Spiesshoefer et al. [40]) and varied from moderate to strong. Similarly, D_T in maximal inspiration was related to SNIP, MIP (apart from one study where no data was provided – Spiesshoefer et al. [40]), MEP, and VC in all studies but varied from weak to moderate. The relationship between D_T at the end of maximal inspiration and FEV₁ was statistically significant in two studies (out of three) and varied from moderate to strong. Intercostal muscle thickness and echogenicity at the end of tidal inspiration were not correlated with FEV₁ and FEV_{1%PRED}. The correlations of the remaining parameters are presented in Table 2 and Appendix 1.

US Ratios and Diaphragm Excursion (Amplitude, Velocity)

Twenty-two studies in which three types of US parameters were used are listed in Table 3. The first parameter was the amount of diaphragm excursion (diaphragm shift) and was used in 18 studies. The second was based on an analysis of the rates of change in thickness (DTR,

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DTF, DTD) and was used in 11 studies. The last one was DE_v and was used in three studies. US parameters were correlated with the following pulmonary parameters: FVC (12 studies), FEV₁ (8 studies), MIP (11 studies), VC (4 studies), FVC_{%PRED} (4 studies), FEV1_{%PRED} (4 studies), inspiratory capacity (IC) (3 studies), MEP (3 studies), total lung capacity (TLC) (2 studies), FEV1/FVC (2 studies), residual volume (RV) (2 studies), RV/TLC (2 studies), IC/TLC (2 studies), SNIP (2 studies), expiratory reserve volume (2 studies) (Fig. 2). In 15 studies, only the right hemidiaphragm was analysed; in 5 studies, it was analysed on both sides; in one study, only on the left side; and in one other study, the side examined was not specified. A wide variety of breathing patterns were used in the US evaluation of the diaphragm in the eligible studies, with as many as eight different descriptions of such patterns and times when US measurements were collected. Depending on the breathing conditions, DE_a was correlated with most but not all pulmonary parameters. DE_v was not correlated with pulmonary parameters in two out of three studies. In turn, the thickness ratios measured at TLC and at functional residual capacity (FRC) were correlated (from weak to strong) with most of the pulmonary parameters. Detailed information with regard to the relationships between US ratios and excursion in relation to pulmonary parameters is presented in Table 3 and Appendix 2.

Discussion

The main aim of this systematic review was to evaluate the potential utility of respiratory muscles US for assessing respiratory function. To the best of our knowledge, there have not been any reviews of studies that have assessed the relationship between pulmonary function and US parameters. Thus, we have decided to gather studies that have assessed the association between pulmonary and US parameters in order to calculate meta-regression. Based on the applied search strategy, 31 articles were included in this review. However, we were unable to find 10 homogeneous studies among these 31 that would be needed to perform a meta-regression.

This review found that three different types of US measurements were most commonly analysed in relation to pulmonary function: D_T, diaphragm thickening (fraction, ratio, difference), and diaphragm excursion (DE_a and DE_v). Not surprisingly, the diaphragm was most commonly analysed by researchers to assess pulmonary function as the diaphragm is the major respiratory muscle [42]. It contracts during inhalation and enlarges the chest space, reducing the internal pressure. Diaphragm dysfunction may impair respiratory function, including dyspnoea, and reduce performance during physical activity [43]. Thus, lung-related conditions are reflected in the functioning of the diaphragm and vice versa [44]. A number of studies have also indicated that US is a reliable tool for assessing the diaphragm [4, 5, 19, 26, 27, 39]. This information confirms the relevance of US examination of the diaphragm.

Among all the included studies, intercostal muscles were analysed in only one [23]. Intercostal muscles contribute to chest expansion [45]. The methodology of intercostal muscle examination is well described in the literature, and the reliability of US measurements has been proven [23, 45]. Thus, it is surprising that there is such limited research interest in the intercostal muscles. This may be because the intercostal muscles play a smaller role than the diaphragm during inspiration or due to methodological problems related to the triangularis sterni muscle located between the ribs [23] or their relatively small changes in echostructure/thickness during breathing (own experience). However, it seems that the role of intercostal muscles should be more extensively examined because it has been shown that inspiratory volume can be increased by the action of the intercostal muscles to a greater extent than the diaphragm alone in some conditions [45]. Other accessory respiratory muscles such as the scalenus, sternocleidomastoid, abdominal muscle, etc., have never been examined in relation to pulmonary function [46]; however, it would be interesting to do so in patients with chronic conditions who require their support (e.g., patients with dyspnoea due to COPD).

Diaphragm Thickness

D_T is a parameter of the diaphragm measured between the pleural membrane and the peritoneal membrane. It is widely accepted that muscle strength is correlated with the muscle's thickness [47]. D_T measured at the end of maximal inspiration was correlated with pulmonary parameters in almost all relevant studies (except for two [29, 40]). The strength of the relationship varied from strong to weak with FVC, FEV₁, MIP, SNIP, VC, and MEP [11, 14, 20, 25, 26, 34, 35, 41]. Higher lung volumes were correlated with greater D_T at the end of maximal inspiration [11, 25, 35]. This may explain the relationship between D_T at the end of maximal inspiration and FVC and FEV1. Additionally, there was a moderate correlation between MIP, MEP and FVC, FEV₁ parameters because all are at least indirectly related to respiratory muscle strength [48]. Parameters like MIP and SNIP are collected during maximal inspiration, and hence, they are also related to D_T at the end of maximal inspiration. Janssens et al. [49] confirmed that transdiaphragmatic pressure is moderately correlated with SNIP. It has been suggested that D_T at the end of maximal inspiration can be used as an indicator of diaphragm contractility [35]. Among these pulmonary parameters, it is difficult to indicate definitively which parameter correlates most strongly with D_T during maximal inspiration. This is probably the result of the moderate positive correlation observed with the pulmonary parameters [48]. However, three studies found a weak or no relationship between pulmonary parameters and D_T measured at the end of maximal inspiration [29, 34, 40]. Potential reasons for this include (1) the overweight status of the examined population because an increase in BMI leads to greater difficulty in examining the diaphragm due to reduced echogenicity and (2) the small sample size with additional dementia and physical disability as in Miyagi et al.'s [34] study. It is worth mentioning that each of these three studies included at least two domains assessed as having an unclear or high risk of bias (shown in Table 4).

Most pulmonary parameters are measured during maximal inspiration or expiration because there is no correlation between these parameters and D_T measured at the end of tidal inspiration [25, 40]. It has been shown that during tidal breathing, there is a small (less than 10%) or no change in the thickness of the diaphragm [50]. In turn, the relationship between D_T measured at the end of tidal expiration and pulmonary parameters was inconsistent, some showing a significant relationship [24, 27, 35, 41] and others showing no such relationship [23, 29, 34, 40]. At this stage, it is difficult to explain such inconsistencies between studies, but they might be associated with the technique of tidal breathing (different breathing patterns, pace, or intensity).

Diaphragm Thickening (Fraction, Ratio, Difference)

DTF, DTR, or DTD is a group of parameters obtained from the analysis of the D_T in two different breathing phases (TLC and FRC). The only difference is the use of a different formula to analyse the same raw results. DTF is the per cent change in D_T . DTR is calculated by dividing two results, whereas DTD is the difference between the two results. These parameters are interchangeable, and this is probably why they did not appear together in any of the relevant studies. DTF was analysed in six studies, DTF was analysed in four, and DTD was analysed in only one study. Most frequently D_T measurements were taken at the moment of maximal inspiratory effort (TLC) and tidal expiration (FRC).

The strength of the relationship of DTF or DTR with MIP varied from strong to weak in all relevant studies [11, 28, 31, 39]. This variability in correlation strength may be due to the large heterogeneity between the examined cohorts. The relationship between DTF or DRT and MIP is stronger in patients with neuromuscular disease [31] and in poststroke patients [39] than in healthy or elderly individuals. This might be related to changes in the diaphragm, which tends to atrophy in stroke, and/or potential changes in diaphragm movement due to impairments in the central nervous system [51].

In turn, the relationship between DTR, DTF, or DTD measurements and FVC and FEV₁ was inconsistent. Some studies showed a moderate correlation [11, 33, 35], and others showed no correlation [18, 33]. Again, such incoherence in the results may be related to the examined population or phase of disease. In the study by Lim et al. [33], there was clearly a stronger relationship between DTF and FEV₁ (FEV_{1%PRED}) in COPD patients in the stable phase compared to the exacerbation phase. This may be due to impaired diaphragm function caused by sys-

temic inflammation and functional exhaustion during the exacerbation phase in COPD patients. Steroids administered in exacerbation also contribute to respiratory muscle weakness [52].

Some other pulmonary parameters (SNIP, VC, RV, IC, TLC, etc.) were also considered but mainly in a single study. This was expected as spirometry most commonly measures parameters like FVC and FEV₁. Not all spirometry devices can measure VC and IC, whereas RV and TLC are measured using less accessible device – plethysmography. In other studies, different breathing phases were shown to explain the time at which D_T measurements were taken – tidal volume or forced breathing [25, 35]. However, changes in D_T during tidal breathing were lower than 10% in almost 30% participants, and in some, there were no changes [50]. Thus, it is more useful to examine the diaphragm at its maximum performance [53].

Diaphragm Excursion (Amplitude or Velocity)

DE_a is described as the upright perpendicular distance from the minimum to the maximum point of diaphragm displacement during a given breathing manoeuvre. This US parameter was most commonly used in studies included in this review, but different breathing patterns were used, which complicates analysis of the results. DE_a was most frequently linked with FVC, presenting a moderate to very strong relationship. The breathing patterns during DE_a were described as deep breathing, forced breathing, or TLC. However, three studies [12, 35, 40] showed no relationship between DE_a and FVC. Smargiassi et al. [35] suggested that BMI has a significant impact on DE_a, which was confirmed in our review. The exception to this was Kwon et al.'s [12] study, where, however the authors used a completely different methodology. FVC%PRED was also correlated with DEa, and a similar relationship to that for FVC was observed. Based on the study by Lim et al. [33], it seems that in some conditions, DE_a is slightly more closely correlated with FVC than FVC_{%PRED}. FVC_{%PRED} is calculated on the basis of age, sex, body weight, and height and thus does not take into account the subjects' diseases. This is probably the reason for the higher correlation of DE_a with FVC than with FVC_{%PRED}. FEV₁ was also moderately to strongly correlated with DE_a except in Lim et al.'s [33] study. In articles analysing FEV₁ (FEV_{1%PRED}) and FVC (FVC_{%PRED}) together, a similar relationship with DE_a was observed.

MIP, another pulmonary parameter, was moderately to strongly correlated with DE_a measured during deep breathing, forced breathing, or TLC and not correlated with DE_a measured during voluntary sniffing, IC, or

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tidal breathing. MIP is determined during forced breathing and is directly related to respiratory muscle strength. Thus, only papers where maximal respiratory effort was analysed showed significant correlations with DE_a . Other pulmonary parameters were much less frequently analysed. Taking all results together, it can be suggested that FVC, FEV₁ and MIP were most commonly related to DE_a and showed the highest and similar correlations (in cases in which all parameters were analysed together).

 DE_v is described as the velocity of diaphragm displacement during a given breathing pattern. DE_V was only analysed in three studies (in two, it was recorded during voluntary sniffing and in one, during voluntary sniffing and tidal breathing) in relation to MIP, FVC, or MEP [10, 14, 40]. In most cases, there were no significant relationships. This is probably due to the way the DE_v was measured (during tidal breathing or voluntary sniffing). It was mentioned earlier that MIP is calculated during forced breathing and is directly related to respiratory muscle strength. However, a recent study [14] found a moderate relationship between DE_v and FVC and a weak relationship between DE_v and MIP or MEP, also during the sniff manoeuvre.

Limitations of Current Review

There were several limitations in this review. First, the number of studies included was relatively small, and the search strategy was limited to studies in English. Second, the sample size of included studies was rather small and heterogeneous. Although a meta-regression was initially planned (to find the best variables), we were unable to find the requisite 10 studies that were similar in terms of study design, population examined, and pulmonary and US parameters measured. Additionally, the high degree of variability in the parameters analysed and the inconsistent descriptions of the conditions in which the US measurements were performed made it very difficult to interpret them correctly, potentially creating an error in the data analysis. For example, some authors used the terms deep breathing (requires the diaphragm to contract after the diaphragm relaxes, air passively leaves the lungs) and forced breathing (requires muscle contractions during both inspiration and expiration) interchangeably, whereas the exhalation phase is different in these two manoeuvres. Another problem was the lack of analysis of all parameters obtainable from the pulmonary assessment. Some studies analysed FVC but not FEV₁ and vice versa. This makes it more difficult to interpret and estimate the potential usefulness of each US parameter in assessing pulmonary function. It is also necessary to ensure the methodological quality of future work as none of the included papers received a low-risk result in any domain.

The limitations are also related to US measurements itself. Overall, the US measurements of the respiratory muscles (intercostal muscles and diaphragm) are reliable [5, 23, 34, 41, 45, 50], but in most studies, the diaphragm is assessed only on the right body side. US visualization of the left hemidiaphragm is considered more difficult due to the smaller window of the spleen (compared to the liver window) [53, 54] and the presence of gas in the stomach and intestine which blocks sound waves [9]. It is also difficult to obtain an adequate angle of the left hemidiaphragm visualization, impairing the measurement accuracy. In practice, Boussuges et al. [54] were only able to assess left hemidiaphragm in 21% of participants. Additionally, numerous approaches (different body positions, probe orientations, breath manoeuvres) of respiratory muscles US were used. In some studies, the description of the US measurement methods was so unclear that it was challenging to determine how the data was collected. The reason for this is the lack of international guidelines for US measurements of respiratory muscles, which negatively affects the understanding of the role of US in the evaluation of the respiratory function in the current review.

Implications for Future Research

Based on this systematic review, several recommendations for future work can be made. Future studies should meet the basic requirements that minimize risk of bias. A detailed description of the respiratory cycle and the points when US measurements were captured is required. It is worth considering using terms established in the literature for the pulmonary test or describing in detail the timing of US measurements. Future studies should also present all measured pulmonary parameters and respiratory muscle strength (when accessible) in relation to US parameters. The methodology of spirometry should also be standardized because in some articles, the measurements were performed in the standing position, contrary to recommendations [13]. Elaboration of international guidelines for respiratory muscles US is also needed. Due to the relatively small changes in D_T during tidal breathing, it seems more reasonable to perform US of the diaphragm during maximal respiratory effort when possible. Additionally, the included studies were mainly limited to diaphragm US. The analysis of other muscles involved in breathing is worth considering - for example, intercostal muscles, which are able to increase inspiratory volume to a greater extent than the diaphragm alone in some conditions [45]. A statistical analysis of all correlated parameters should be made available (at least as an appendix). To better understand the potential usefulness of US measurements, it is important not to limit the examination to people with diseases. Studies of healthy people (adults and children) and even athletes are worth considering.

Clinical Context

Based on the results of this systematic review, respiratory muscle US as a surrogate of pulmonary function in clinical practice cannot be clearly recommended. An application of US for the assessment of pulmonary function (instead of spirometry) should be used with caution as the results correlating US measurements of the diaphragm with pulmonary function are conflicting and rely on the methodology used and population examined. It is worth noting that information obtained by US refers specifically to a given respiratory muscle, while spirometry parameters depend on many factors (air volumes, airflows, pulmonary compliance, respiratory muscles function, etc.). This was the reason why we initially hypothesized that pulmonary parameters directly related to strength of the respiratory muscles should present higher relationship with US measuring respiratory muscles. Putting all the results together, this systematic review failed to definitively indicate a higher correlation of MIP, MEP, or SNIP with US measurements compared to other parameters determining respiratory function. In turn, there was a visibly higher correlation between pulmonary and US parameters in selected neurological diseases, which may indicate that US measurements are more useful for assessing the respiratory system in some clinical conditions. Thus, further studies on patients are needed to obtain more conclusive evidence on the usefulness/nonusefulness of US for assessing pulmonary function in clinical practice. From this perspective, US measurements will rather supplement than replace pulmonary function assessment. At present, US measurements are useful (i) in assessing diaphragm dysfunction (paralysis [37], asymmetrical movements [55], congenital anomalies [56]) or (ii) as a prognostic marker for frequent exacerbations in COPD patients [32]. It is also worth considering US measurements of the diaphragm in surgical management/repair of congenital diaphragmatic hernia [57-59] and as an additional indicator in the assessment of effort tolerance in pulmonary rehabilitation or rehabilitation progress [60].

Conclusion

A strong relationship between US measurements and pulmonary function was demonstrated in some studies but not others. Based on the current state of knowledge, it is not possible to definitively identify the US measurements that best correlate with pulmonary parameters, especially as these relationships differ between healthy subjects and patients with particular health problems. Additionally, pulmonary parameters directly related to respiratory muscles strength did not present higher (compared to other pulmonary parameters) relationship with US measuring respiratory muscles. Thus, there is not enough evidence to use US measurements alone to assess pulmonary function. Further studies using standardized methodology are needed to obtain more conclusive evidence on the usefulness of US for assessing pulmonary function. Current evidence suggests that US measurements can successfully complement spirometry, but the exact role of the US measurements remains to be confirmed.

Statement of Ethics

An ethics statement is not applicable because this study is based exclusively on published literature.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

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Author Contributions

Pałac M.: project administration, conceptualization of ideas, preparation, writing – original draft, data curation, methodology, and formal analysis. Rutka M.: data curation, methodology, formal analysis, and writing – review and editing. Wolny T.: formal analysis and writing – review and editing. Podgórski M.: formal analysis, writing – review and editing, and supervision. Linek P.: project administration, visualization, conceptualization of ideas, preparation, methodology, validation, writing – review and editing, and supervision.

Data Availability Statement

All data generated or analysed during this study are included in this article. Further enquiries can be directed to the corresponding author.

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Article Intra-Rater Reliability of Shear Wave Elastography for the Quantification of Respiratory Muscles in Adolescent Athletes

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Abstract: The aim of this study was to assess the intra-rater reliability and agreement of diaphragm and intercostal muscle elasticity and thickness during tidal breathing. The diaphragm and intercostal muscle parameters were measured using shear wave elastography in adolescent athletes. To calculate intra-rater reliability, intraclass correlation coefficient (ICC) and Bland–Altman statistics were used. The reliability/agreement for one-day both muscle measurements (regardless of probe orientation) were at least moderate. During the seven-day interval between measurements, the reliability of a single measurement depended on the measured parameter, transducer orientation, respiratory phase, and muscle. Excellent reliability was found for diaphragm shear modulus at the peak of tidal expiration in transverse probe position (ICC_{3.1} = 0.91-0.96; ICC_{3.2} = 0.95), and from poor to excellent reliability for the intercostal muscle thickness at the peak of tidal inspiration with the longitudinal probe position (ICC_{3.1} = 0.26–0.95; ICC_{3.2} = 0.15). The overall reliability/agreement of the analysed data was higher for the diaphragm measurements (than the intercostal muscles) regardless of the respiratory phase and probe position. It is difficult to identify a more appropriate probe position to examine these muscles. The shear modulus/thickness of the diaphragm and intercostal muscles demonstrated good reliability/agreement so this appears to be a promising technique for their examination in athletes.

Keywords: shear wave elastography; adolescent; athletes; diaphragm; intercostal muscles

1. Introduction

Respiratory muscles are considered not only in the context of the respiratory system, but also in relation to spine stability, intra-abdominal pressure [1,2], pain sensation [3,4] and body balance [5]. Respiratory muscle morphology (mainly the diaphragm and intercostal muscles) can be assessed using ultrasound imaging (US) [6–9]. Recently, shear wave elastography (SWE) as a new, non-invasive US imaging technique, has allowed the assessment of muscle's mechanical properties [10,11]. It has been suggested that SWE may be used as an index of diaphragmatic force change [12], and that the diaphragm shear modulus measured using SWE is related to transdiaphragmatic pressure [13,14], which is considered the gold standard in diaphragm evaluation.

There are a number of studies assessing the reliability of US diaphragm thickness [3,15,16], echogenicity [17], excursion [18–20] or velocity [21,22] measurements. The reliability of intercostal muscle US measurements were, in turn, evaluated only in four studies [7,23–25]. The reliability of the diaphragm SWE was only measured in two studies [26,27] on a limited population (healthy adults or chronic obstructive pulmonary disease and critically ill patients), whereas intercostal muscles were only analysed in one study [23]. To the best of our knowledge, there is only one reliability study on intercostal muscle SWE [23] and no study of diaphragm SWE in adolescents. Although, diaphragm and intercostal muscle SWE



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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). or thickness is usually measured in adults or patients with impaired respiratory system, it could be useful to assess the SWE of these muscles in adolescent athletes. The reliability results for the adults (who were sometimes critically ill) should not be transferred to healthy athletes in whom the functioning of the respiratory system (and respiratory muscles) function is expected to be at or above the population norm. It was confirmed that athletes have a greater diaphragm thickness [16] and higher pulmonary parameters than non-athletes [28,29].

Vicente-Campos et al. [3] have suggested that diaphragm exercise should be a crucial component of sports performance, injury prevention and rehabilitation strategy. Therefore, it is important to consider investigation of respiratory muscles (especially the diaphragm) in a broader (not just respiratory-related) context and on heterogeneous populations. As an example, there is a relationship between diaphragm thickness and non-specific lumbopelvic pain in athletes [9]. We believe that extensive research considering respiratory muscle measurements by SWE in adolescent athletes could provide new knowledge about the physiology of these muscles and potentially influence training, diagnostic, prognostic, or rehabilitation procedures. However, the reliability and agreement of SWE will be important to ensure the study and measurement quality in future studies assessing respiratory muscles in adolescent healthy athletes. Thus, the aim of this study was to assess the intra-rater reliability and agreement of diaphragm and intercostal muscle elasticity and thickness during tidal breathing.

2. Materials and Methods

2.1. Setting and Study Design

This study was conducted in Musculoskeletal Elastography and Ultrasonography Laboratory in accordance with the Declaration of Helsinki. The protocol was approved by local Ethics Committee (Decision No. 9/2020). All participants and their parents were informed about the procedures performed and provided written informed consent to participate in the study.

2.2. Participants

Ten male footballers (mean age: 17.1 ± 0.29 ; mean body mass: 71.5 ± 7.57 ; mean body height: 179.9 ± 5.67 ; BMI: $22.1 \pm 1.90 \text{ kg/m}^2$; football participation from 7 to 9 years) were selected using convenience sampling from an elite youth football club.

2.3. Investigator

Ultrasound data (SWE, thickness) were collected and analysed by a physiotherapist. Prior to the study, examiner had 3 years of experience in musculoskeletal SWE. Before the study, the examiner was additionally trained by an experienced radiologist in evaluating the respiratory muscles and underwent 3 months practical training.

2.4. Equipment

An Aixplorer ultrasound scanner (Product Version 12.2.0, Sofware Version 12.2.0.808, Supersonic Imagine, Aix-en-Provence, France) coupled with a linear transducer array (2–10 MHz; SuperLinear 10-2, Vermon, Tours, France) was used to evaluate the diaphragm and intercostal muscles' shear modulus and thickness.

2.5. Measurement Procedures

The measurements were performed on the right body side in the supine position using SWE mode. The patient's right hand was placed under the head in order to better visualize the diaphragm on the US. At the beginning, the examiner marked anterior and mid-axillary line on the chest, and positioned the US probe between them (the right intercostal space). The probe was positioned in the first intercostal space (counting from the bottom) where the lungs did not obscure the diaphragm during tidal breathing. The US measurements were performed in two probe orientations: transversally to the ribs—long body axis (Figure 1A) and parallel to the ribs—space between two ribs (Figure 1B).



Figure 1. Illustration showing the placement of the transversally (**A**) and parallel to the ribs (**B**) ultrasound probe position.

The participants were asked to stay calm and breath quietly throughout the measurement procedure. US data was collected twice at the end-tidal inspiration and at the end-tidal expiration, separately. The moment of determining the end stage of inspiration and expiration was based on the visual inspection of diaphragm movement on real-time US. The end of diaphragm movement during tidal breathing was defined as the end of tidal inspiration or expiration.

After 7 days, the procedure was replicated in order to calculate reliability in a more extended time interval. The examiner was encouraged to apply minimum force by US probe to the skin because this may have affected the study results [30]. The left side was not examined due to the smaller acoustic window affecting reliability [20].

2.6. Data Analysis

From each US image collected in SWE, mode thickness and shear modulus (elasticity) measurements were collected. The Q-Box quantitative tool was used to quantify muscle shear modulus. Three circles were positioned in the middle of the image and inside the fascial edge of each muscle between the ribs. The circles were always next to each other and omitted potential artefacts (when they were detected).

In order to measure thickness precisely, the images were saved on an external drive in DICOM format and transferred to a computer where they were further processed using RadiAnt DICOM Viewer (Medixant, Poznań, Poland). If needed, images were sharpened, enlarged and contrasted to better visualize the pleural line and the peritoneal line. The diaphragm thickness was measured between these two hyperechoic lines. The intercostal muscles were measured as the first muscle placed was more superficial than the diaphragm (Figure 2). Shear modulus and thickness of the muscles were measured manually based solely on the examiner's experience.

2.7. Statistical Analyses

To calculate intra-rater reliability, intraclass correlation coefficient (ICC) type 3.1 (for single measurement) and type 3.2 (for mean value from two measurements) were used. The ICC was interpreted according to the following criteria: 1.00–0.75 (excellent), 0.74–0.60 (moderate), 0.59–0.40 (fair), and below 0.40 (poor reliability) [31]. In order to calculate agreement, the standard error of measurement (SEM = SD × $\sqrt{1 - ICC}$), the coefficient of variation (CV), and the results of the Bland–Altman test (BA) were used. The only reason to use the BA test was to find potential biases between the two measures. Due to the sample size not being large enough (more than 50 is preferred to allow a good estimation of the limits of agreement), plots with limits of agreement were not included [32]. The significance



level was set at p < 0.05. Data were analysed using STATISTICA 13.1 PL (Statsoft, Tulsa, OK, USA) and Excel 2013 (Microsoft Corporation, Redmond, Washington, DC, USA) software.

Figure 2. Data extraction from the images collected in SWE mode in transverse (**A**) and longitudinal (**B**) probe view. D—diaphragm; IC—intercostal muscle.

3. Results

В

3.1. Transverse Probe Orientation (Transversally to the Ribs)

The one-day intra-session reliability (ICC_{3.1}) of diaphragm and intercostal muscles shear modulus at peak of tidal expiration and inspiration was generally excellent. The corresponding CV was always below 3% at inspiration phase and below 1% at expiration phase; no systematic errors in BA test were detected. The intra-session reliability for single measurement (ICC_{3.1}) during the 7-day interval varied from excellent to moderate for diaphragm and from fair to poor for intercostal muscles. The intra-session reliability (ICC_{3.2}) for the mean value from two measurements was improved (excellent for diaphragm, fair to excellent for intercostal muscles). Corresponding CV was always below 8%, but systematic error was detected for diaphragm shear modulus at peak tidal inspiration. The expiration phase always corresponds with higher ICC and lower SEM and CV.

The diaphragm and intercostal muscle thickness demonstrated excellent one-day intrasession reliability (ICC_{3.1}) with CV below 9%. The BA test showed negative mean bias with systematic error for intercostal muscles at peak inspiration and expiration and positive bias for diaphragm. The intra-session reliability for single measurement (ICC_{3.1}) during the 7-day interval varied from excellent to fair, but no systematic errors were detected. The intra-session reliability (ICC_{3.2}) for the mean value from the two measurements varied from excellent to moderate, and the corresponding CV did not exceed 6.03%. The mean bias was close to zero for the diaphragm and intercostal muscle thickness measurements during peak tidal inspiration and expiration. All results of the reliability and variability in transverse probe orientation are included in Table 1.

		Inspi	ration	Expir	ation
		D	IC	D	IC
	Mean (kPa) ¹	27.36	24.96	25.42	23.81
Shear modulus	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	5.31	5.20		
	$\frac{D}{P} = \frac{D}{P} = \frac{D}$	0.96	0.95		
Inter consist with the (1 dow)	SEM (kPa)	2.13	1.81	1.07	1.03
Intra-session reliability (1 day)	CV (%)	2.98	1.60	0.71	0.17
	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	-0.26	0.06		
	$\begin{array}{c ccccc} \textbf{(ay)} & CV (\%) & 2.98 & 1.60 \\ \hline Bias ^3 (kPa) & 1.22 & 0.58 \\ \hline ICC_{3.1} & 0.66 & 0.35 \\ SEM (kPa) & 3.40 & 4.00 \\ CV (\%) & 10.63 & 5.69 \\ \hline Bias ^3 (kPa) & 4.14^2 & 1.99 \\ ICC_{3.2} & 0.78 & 0.47 \\ SEM (kPa) & 2.53 & 3.66 \\ CV (\%) & 7.32 & 3.36 \\ \hline Bias ^3 (kPa) & 3.34^2 & 1.09 \\ \hline \hline Mean (mm) ^1 & 1.79 & 3.65 \\ SD (mm) ^1 & 0.62 & 0.74 \\ \hline \end{array}$	0.91	0.51		
Intra-cossion reliability (after 7 days)	SEM (kPa)	3.40	4.00	1.61	3.68
	CV (%)	10.63	5.69	3.14	0.54
Intro cossion reliability (after 7 days)	Bias ³ (kPa)	4.14^{2}	1.99	1.13	-0.18
intra-session renability (after 7 days)	ICC _{3.2}	0.78	0.47	0.95	0.76
	SEM (kPa)	2.53	3.66	1.18	2.53
	CV (%)	7.32	3.36	2.94	0.50
	ty (after 7 days) $ICC_{3\cdot 2}$ 0.78 0.47 0.95 SEM (kPa) 2.53 3.66 1.18 CV (%) 7.32 3.36 2.94 $Bias^3 (kPa)$ 3.34 ² 1.09 1.28 Mean (mm) ¹ 1.79 3.65 1.48 $SD (mm)^1$ 0.62 0.74 0.32	-0.07			
March di Lanca	ickness Mean (mm) 1 1.79 3.6 SD (mm) 1 0.62 0.7		3.65	1.48	3.71
Muscle thickness	$ \frac{\text{CV}(\%)}{\text{Bias}^{3}(\text{kPa})} = 3 $ Muscle thickness $ \frac{\text{Mean}(\text{mm})^{1}}{\text{SD}(\text{mm})^{1}} = 0 $ ICC _{3·1}	0.62	0.74	0.32	0.88
	ICC _{3·1}	$\begin{array}{cccc} 2.53 & 3.66 \\ 7.32 & 3.36 \\ 3.34^2 & 1.09 \\ \hline 1.79 & 3.65 \\ 0.62 & 0.74 \\ \hline 0.93 & 0.76 \\ 0.93 & 0.76 \\ \hline \end{array}$	0.76	0.80	0.93
Intro cossion valiability (1 day)	SEM (mm)	0.20	0.43	0.15	0.25
Intra-session renability (1 day)	CV (%)	$\begin{array}{cccc} n \ (mm)^{1} & 1.79 & 3.65 \\ (mm)^{1} & 0.62 & 0.74 \\ \hline CC_{3\cdot 1} & 0.93 & 0.76 \\ \mathcal{M} \ (mm) & 0.20 & 0.43 \\ V \ (\%) & 2.21 & 8.66 \end{array}$	6.37	4.48	
	Bias ³ (mm)	0.06	-0.44 2	0.14 ²	-0.23
	ICC _{3·1}	0.80	0.51	0.48	0.84
	SEM (mm)	0.29	0.54	0.25	0.36
	CV (%)	0.00	8.11	6.55	3.72
Intra-cossion reliability (after 7 days)	Bias ³ (mm)	-0.001	-0.41	0.14	-0.19
inna-session renavinty (antel / udys)	ICC _{3·2}	0.85	0.65	0.75	0.92
	SEM (mm)	0.24	0.46	0.17	0.25
	CV (%)	1.48	6.03	4.91	2.75
	Bias ³ (mm)	-0.01	-0.04	0.08	-0.06

Table 1. Reliability and validity of stiffness and thickness values measured in diaphragm (D) and intercostal (IC) muscles in transverse probe orientation.

CV—coefficient of variation; ICC—intraclass correlation coefficient; SEM—standard error of the mean; ¹ from all (four) measurements; ² systematic error as the line of equality is not in the 95% confidence interval; ³ Bland–Altman test.

3.2. Longitudinal Probe Orientation (Parallel to the Ribs)

The one-day intra-session reliability (ICC_{3.1}) of the diaphragm and intercostal muscles' shear modulus at peak of tidal expiration and inspiration varied from excellent to moderate. Corresponding CV was always below 4%. The intra-session reliability for single measurement (ICC_{3.1}) during the 7-day interval varied from fair to poor but the corresponding CV was always below 4%. The intra-session reliability (ICC_{3.2}) for the shear modulus mean value from two measurements was improved (moderate for all measurements) and the CV was still below 4%. In the longitudinal probe orientation, the BA test showed bias below 2 kPa with no systematic errors. The expiration and inspiration phases showed similar reliability and agreement results.

The diaphragm and intercostal muscles' one-day intra-session reliability (ICC_{3.1}) of thickness measurements varied from moderate–excellent with CV below 10%. The BA test showed a mean bias close to zero with no systematic errors. The intra-session reliability for single measurement (ICC_{3.1}) during the 7-day interval was moderate for the diaphragm and varied from poor to fair for the intercostal muscles. The corresponding

CV was always below 6%. The intra-session reliability (ICC_{3.2}) for the mean value from two thickness measurements was improved with the exception of the intercostal muscles during inspiration. The corresponding CV did not exceed 6.37% and no systematic errors were detected in any cases. All results of reliability and variability in longitudinal probe orientation are included in Table 2.

Table 2. Reliability and validity of stiffness and thickness values measured in diaphragm (D) and intercostal (IC) muscles in longitudinal probe orientation.

		Inspir	ation	Expir	ation
		D	IC	D	IC
Shear modulus	Mean (kPa) ¹	30.69	26.66	28.60	25.94
	SD (kPa) ¹	6.36	6.26	7.15	5.72
	ICC _{3.1}	0.94	0.85	0.68	0.44
Intra-session reliability (1 day)	SEM (kPa)	1.27	1.99	3.85	4.09
intra-session renability (1 day)	CV (%)	0.41	3.18	2.03	3.44
	Bias ² (kPa)	-0.18	1.20	0.83	1.24
	$ICC_{3\cdot 1}$	0.43	0.50	0.38	0.52
	SEM (kPa)	5.33	4.52	5.68	3.90
	CV (%)	0.23	1.04	3.08	0.12
Intra-session reliability (after 7 days)	Bias ² (kPa)	0.10	0.40	1.25	0.04
intra-session renability (arter 7 days)	ICC _{3·2}	0.63	0.65	0.65	0.66
	SEM (kPa)	3.92	3.74	4.33	3.52
	CV (%)	3.14	2.10	1.84	2.58
	Bias ² (kPa)	1.00	0.15	0.63	-0.72
Marsala (bislansaa	Mean (mm) 1	1.83	3.81	1.56	3.85
Muscle thickness	SD (mm) ¹	0.61	0.68	0.42	0.83
	ICC _{3·1}	0.96	0.95	0.68	0.84
Intra-cossion reliability (1 day)	SEM (mm)	0.15	0.19	0.29	0.39
Intra-session renability (1 day)	CV (%)	0.29	1.18	9.41	2.18
	Bias ² (mm)	-0.007	-0.06	0.21	-0.12
	ICC _{3·1}	0.67	0.26	0.73	0.59
	SEM (mm)	0.33	0.68	0.27	0.55
	CV (%)	1.58	0.35	5.13	2.87
Intra-sossion reliability (after 7 days)	Bias ² (mm)	-0.04	-0.02	0.12	0.15
intra-session remaining (arter / udys)	ICC _{3·2}	0.85	0.15	0.78	0.64
	SEM (mm)	0.24	0.67	0.21	0.51
	CV (%)	1.69	1.69	6.37	3.57
	Bias ² (mm)	-0.05	-0.05	0.06	0.22

CV—coefficient of variation; ICC—intraclass correlation coefficient; SEM—standard error of the mean; ¹ from all (four) measurements; ² Bland–Altman test.

4. Discussion

The main aim of the study was to assess the reliability and agreement of shear modulus measurements in diaphragm and intercostal muscles at peak of tidal expiration and inspiration. To the best of our knowledge, no other studies have calculated the reliability and agreement of these muscle measurements in adolescent athletes. In our study, we showed that regardless of the probe orientation and the muscle tested, the reliability/agreement for one-day measurements were at least moderate. However, at transverse probe positioning, a bias in the thickness measurements of the diaphragm and intercostal muscle was detected (in the second measurement, higher values were found compared to the first measurement). From a clinical perspective, it is more reasonable to analyse reliability/agreement at longer intervals. During the seven-day interval between measurements, the reliability of a single measurement depended on the measured parameter, transducer orientation, respiratory phase, and muscle. Excellent reliability was found for diaphragm shear modulus at the peak of tidal expiration in transverse probe position, and poor reliability for intercostal

muscle thickness at the peak of tidal inspiration with the longitudinal probe position. At the 7-day interval, the analysis of the mean values from the two measurements allowed moderate reliability for almost all variables analysed (with the exception being the reliability of intercostal muscle thickness at the peak of tidal inspiration in the longitudinal probe position), and the CV for all variables was remarkably below 10%. The overall reliability/agreement of the analysed data was higher for the diaphragm elasticity and thickness measurements (in relation to the intercostal muscles) regardless of the respiratory phase and probe position. The longitudinal probe position is characterised by a lack of bias and slightly lower CV values.

In the literature, there are a number of studies assessing the reliability of diaphragm and intercostal muscle thickness in adults with diseases [7,27,33], healthy adults [19,25] or athletes [3,15]. Out of these, there were only two studies evaluating the diaphragm thickness reliability in adolescents [34,35], where ICC or bias at the peak of tidal expiration was similar to the present study's results [34,35]. These results were similar despite the use of a different methodology, a larger age span, and the use of a different interval between repeated measurements. In studies on adults, the reliability of diaphragm thickness at the end of maximal inspiration [18,36–40], at the end of tidal expiration [3,15,18] and at the end of tidal inspiration [18,39,41] was confirmed. In turn, the reliability of intercostal muscles thickness at the peak of tidal expiration [7,24,25] and at the end of maximal inspiration ranged from 0.6 to 0.9 [24,25], which was also consistent with the results obtained in this study.

The reliability of diaphragm shear modulus was only assessed in adults [26,27], and intercostal muscles in adolescents as well [23]. The intra-rater ICC for diaphragm elasticity was excellent for measurements at the end of tidal expiration [26] and at apnea after expiration [27]. The ICC for intercostal muscles was also excellent during normal breathing and in apnea [23]. In all of these studies, the reliability was calculated for data collected during the same day, and was similar to the present study results (the one-day reliability of the diaphragm and intercostal muscle elasticity was also excellent in the transverse probe position).

In the present study, we also attempted to determine the reliability/agreement of the intercostal muscles and diaphragm US measurements, taking into account the probe orientation (transverse vs. longitudinal). This is particularly important in the assessment of elasticity by SWE, as evidence shows that the probe orientation in relation to muscular fibres may affect the results [42]. The diaphragm shear modulus reliability was evaluated in longitudinal [26,27] and transverse [26] probe orientation, but intercostal muscle shear modulus was only analysed in the transverse probe orientation [23]. In all of these studies, the reliability for one-day measurements was excellent. Only in the study by Flattres et al. [26] was the obtained reliability poor for the diaphragm assessment in the transverse probe orientation. In our study, longitudinal and transverse probe orientation resulted in excellent reliability for assessing diaphragm and intercostal muscle elasticity during tidal inspiration. During expiration, we found better reliability in the transverse probe orientation, which is only contrary to the results obtained in the study by Flattres et al. [26], where better reliability was observed in the longitudinal probe position. This may be due to differences in the population studied. In our study, there were slender adolescent athletes, whereas in the study by Flatters et al. [26] adults were recruited. Regular sporting activity influences the lungs and chest elasticity [28], and may explain the differences in reliability of the elasticity measurements between longitudinal and transverse probe position.

The present study had a number of limitations. First, the sample size was small and homogenous (adolescent athletes), and the results should be applied with caution to different populations. Second, the examiner had relatively little experience in the diaphragm and intercostal muscle assessment. However, SWE does not require much examiner experience in assessing the diaphragm [26]. In the present study, one-day reliability was excellent for most of the variables analysed. Third, probe compression was not controlled by an external device or specialised US gel pad. Another study showed that probe stabilizing

grips may affect the muscle's elasticity [30]. Fourth, in the present study, we evaluated only intra-examiner reliability and an inter-examiner calculation needs to be performed. Fifth, the athletes were only measured in supine position. It is frequent practice to examine the diaphragm in other body positions (e.g., semi-supine, seated)—so it is worth remembering that the reliability values in the present study (and other work cited) only apply to the supine position (body position can affect diaphragm relaxation). Six, measurements were only collected during tidal breathing.

5. Conclusions

Shear modulus/thickness of the diaphragm and intercostal muscles during tidal breathing demonstrated good reliability/agreement in adolescent athletes. However, the diaphragm had better reliability. SWE appears to be a promising technique to examine the diaphragm and intercostal muscles in athletes. At this stage, it is difficult to unambiguously identify a more appropriate probe position (transverse vs. longitudinal). We currently recommend taking at least two repeated measurements and analysing the mean value. Further studies are needed to establish an optimal measurement procedure and improve the reliability (in particular during intercostal muscle assessment at tidal inspiration).

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Relationship between respiratory muscles ultrasound parameters and running tests performance in adolescent football players. A pilot study

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ABSTRACT

Purpose. Assessing the relationship between ultrasound imaging of respiratory muscles during tidal breathing and running tests (endurance and speed) in adolescent football players.

Methods. Ultrasound parameters of the diaphragm and intercostal muscles (shear modulus, thickness, excursion, and velocity), speed (30-m distance), and endurance parameters (multi-stage 20-m shuttle run test) were measured in 22 male adolescent football players. The relation between ultrasound and running tests were analysed by Spearman's correlation.

Results. Diaphragm shear modulus at the end of tidal inspiration was moderately negatively (R = -0.49; p = 0.2) correlated with the speed score at 10 m. The diaphragm and intercostal muscle shear modulus ratio was moderately to strongly negatively correlated with the speed score at 10 m and 30 m (about R = -0.48; p = 0.03). Diaphragm excursion was positively correlated with the speed score at 5 m (R = 0.46; p = 0.04) and 10 m (R = 0.52; p = 0.02). Diaphragm velocity was moderately positively correlated with the speed score at 5 m (R = 0.42; p = 0.06) and 30 m (R = 0.42; p = 0.07). Ultrasound parameters were not significantly related to all endurance parameters ($R \le 0.36; p \ge 0.11$).

Conclusions. Ultrasound parameters of the respiratory muscles are related to speed score in adolescent football players. The current state of knowledge does not allow us to clearly define how important the respiratory muscles' ultrasound parameters can be in predicting some performance parameters in adolescent athletes.

Subjects Kinesiology, Sports Medicine Keywords Athlete, Ultrasonography, Motor skills, Respiration, Diaphragm, Intercostal muscle

INTRODUCTION

It is well known that respiratory function is related to physical activity and affects exercise performance in athletes. Respiratory muscles (RMs) are an integral part of the respiratory system and physical activity. Their morphology and contractile properties make them useful in endurance types of training (*Welch, Kipp & Sheel, 2019*). RMs are susceptible to

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fatigue, resulting in reduced performance (*Aliverti, 2016; Welch, Kipp & Sheel, 2019*) and insufficient oxygen supply to the working muscles (*Mcconnell & Lomax, 2006*). Studies have shown that RMs training improves RMs' parameters and decreases muscle fatigue, resulting in a change in respiratory system function (*Welch, Kipp & Sheel, 2019*). It is also indicated that inspiratory muscle training affects the test results involving time trials or exercise endurance time (*Hajghanbari et al., 2013*). The main RMs are the diaphragm (DA) and intercostal muscles (IMs). Physiologically, the DA executes about 65% of the respiratory work during inspiration (*Moeliono, DM & Nashrulloh, 2022*) and affects to a greater extent lung movements (*Welch, Kipp & Sheel, 2019*). IMs, in turn, contribute to chest expansion (*Yoshida et al., 2021*), leading to increased inspiratory volume (*Yoshida et al., 2019*). During inspiration, while the IMs contract, the abdominal muscles gradually relax, and vice versa during expiration. This mechanism has some effects: (a) it prevents rib cage distortion; (b) the DA is unloaded and can act as a flow generator; and (c) the abdominal volume decreases below resting levels (*Aliverti, 2016*).

In football, RM training improves RMs' strength, which helps to improve exercise tolerance and lower blood lactate levels (*Guy, Edwards & Deakin, 2014*). Respiration exercises also improve muscle oxygen supply during high-intensity exercise (*Archiza et al., 2018*). This process can be translated into an improvement in fatigue tolerance and running efficiency of football players (*Archiza et al., 2018*). Additionally, it was confirmed that in youth football players, the RMs improve aerobic endurance, which is one of the most important parameters of motor preparation in football (*Mackala et al., 2020*).

Spirometry, as a gold standard of assessing respiratory function (*Durmic et al., 2015*), allows reproducible and standardised assessment of pulmonary function (*Lazovic-Popovic et al., 2016*). However, spirometry performance is the result of many factors (including airway obstruction, respiratory compliance, and RM strength) that do not allow direct analysis of the RMs (*Palac et al., 2022*). In contrast, ultrasound (US) imaging can directly and reliably assess the thickness, excursion, and shear modulus (elasticity) of the RMs (*Palac et al., 2019*). *Palac et al. (2022*) also confirmed the reliability of RMs US measurements in adolescent football players. In the literature, some studies have shown the relationship between US parameters of the RMs and spirometry parameters in different populations (*Palac & Linek, 2022*). However, a recent systematic review by *Palac & Linek (2022)* has shown that the relationship between US parameters and lung function (measured, for example, by spirometry) is inconclusive. Thus, the two methods of measurement should not be used interchangeably, as they measure different aspects (*Palac & Linek, 2022*).

Taking into account that RMs training affects motor skills and has implications for sports training, it is worth considering these muscles in athletes. Running tests are usually used to assess motor skills such as speed and endurance. According to the literature, speed and endurance depend on the thickness of the lower-extremity muscles, which has been measured using US in young athletes (*Stock et al., 2017*). Other US parameters have been related to motor skills in elite sports (*Sarto et al., 2021*). For example, RMs function correlates with postural stability in footballers (*León-Morillas et al., 2021*), and thus potentially affects motor skills as well. To the best of our knowledge, however, there

have been no studies relating US measurements of RMs with motor skills (endurance and speed) in adolescent football players. We believe that such an analysis is justified, as it may launch the exploration of RMs US measurements that are potentially useful in predicting motor skill performance in athletes. The aim of this preliminary report was to assess the relationship between US of RMs during tidal breathing and selected motor skill (endurance and speed) performance in adolescent football players. Based on the current state of the art, we hypothesised that endurance and speed parameters should be related to the thickness and elasticity of RMs (DA and IMs) in adolescent football players.

MATERIALS & METHODS

Informed consent

The study was approved by the Ethics Committee of the Jerzy Kukuczka Academy of Physical Education in Katowice (Decision No. 9/2020) and conducted in accordance with the guidelines of the Declaration of Helsinki. Before the study, participants and their parents were informed about all procedures performed and have given written consent to participate. All participants provided written informed consent to participate in the study. This research did not receive any external funding.

Setting and study design

US data were collected in a laboratory setting (Institute of Physiotherapy and Health Sciences, Musculoskeletal Elastography and Ultrasonography Laboratory) by two physiotherapists, whereas endurance and speed measurements were performed by a motor preparation assistant on a football field with an artificial ground surface. Speed and endurance tests were conducted during two consecutive training days. During the first day, speed tests were performed, and on the next day, an endurance test was conducted. All measurements were performed in a preparation phase for the next football season. Due to organisational issues, US was collected one week after the endurance measurements.

Participants

Adolescent footballers from the professional football academy were considered for the study. We invited all male individuals from a randomly selected team (one age group). The basic criteria of eligibility for the study were (a) all players had to be free of any health or injury issues at the time of testing; (b) no respiratory-related medical history; and (c) no surgical procedure on the pectoral chest, abdominal cavity, pelvic girdle, and/or spine. Information regarding the athletes' health was obtained by a short interview with the footballers and a coach or physiotherapist working with these athletes in the club.

Ultrasound measurements

All US measurements were collected by an Aixplorer US scanner (Product Version 12.2.0, Software Version 12.2.0.808; Supersonic Imagine, Aix-en-Provence, France). Linear transducer array (2–10 MHz; SuperLinear 10-2, Vermon, Tours, France) in the SWE mode was used to evaluate the shear modulus and thickness of the ICs and DA on the right side of the body. Each participant laid in the supine position with the right hand



Figure 1 Illustration showing the ultrasound probe placement and orientation (parallel to the ribs). Full-size DOI: 10.7717/peerj.15214/fig-1

placed under the head in order to better visualise the DA. At the beginning, anterior and mid-axillary lines were marked on the participant's chest, and the US probe was positioned between the lines (Fig. 1). The probe was positioned in the first intercostal space (counting from the bottom) where the lungs did not obscure the DA during tidal breathing. The US measurements were performed in a longitudinal probe position (parallel to the ribs). The participants were asked to relax and breath quietly throughout the procedure. US data were collected twice at the end-tidal inspiration and at the end-tidal expiration, separately. The reliability of RM measurements has been confirmed in previous studies on healthy adolescent football players (*Pałac & Linek, 2022*).

DA excursion was collected in the M-mode on the Aixplorer US scanner coupled with convex transducer array (1–6 MHz, Cristal Curved XC6-1; Vermon, Tours, France). For the excursion measurement, the participant was in the supine position with the upper limbs along the trunk. The probe was placed in the right subcostal area. The participant was asked to take a maximal inspiration and then quietly expire. For the excursion DA measurement, a video collecting the work of breathing before maximal inspiration (tidal expiration) and during maximal inspiration and tidal expiration was recorded. The reliability of DA excursion was confirmed on athletes (*Calvo-Lobo et al., 2019*). DA excursion amplitude was described as the upright perpendicular distance from the minimum to the maximum point of DA displacement during a given breathing manoeuvre. DA excursion velocity is described as the velocity of DA displacement (during a given breathing pattern).

Shear modulus and thickness were calculated from the US images. The Q-Box[™] quantitative tool was used to quantify muscle shear modulus. Three separate circles were positioned inside the fascial edge of each muscle, and the shear modulus was automatically calculated. The images were then saved on an external drive in DICOM format and transferred to a computer, where the muscle thickness was measured using RadiAnt DICOM Viewer (Medixant, Poznań, Poland). The DA thickness was measured between the pleural and peritoneal lines. The ICs were measured as the first more superficial

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muscle than the DA. The thickness and shear modulus ratio was also measured as the end-inspiratory US value divided by the end-expiratory US value.

Running tests

Two running tests were used to analyse the participants' endurance and speed. All measurements were collected by using photocells of the Witty System (Microgate Bolzano, Italy) with an accuracy of 0.01 s. The Witty System was coupled with Witty Manager (1.14.32 version; Microgate Bolzano, Italy) and connected to a laptop, allowing data collection (*Altmann et al., 2019*). Both tests were performed on a dry grass football pitch on a sunny day, and the participants wore football kit and boots.

Endurance was assessed by a progressive, multi-stage 20-m shuttle run test (MSRT) as a modification of the beep test (*Green et al., 2013*). The beep test requires athletes to run back and forth ("shuttle") between two cones separated by 20 m. The initial speed was 2.22 m/s for 1 min. At the end of the first min, the speed increased to 2.5 m/s and progressively increased by 0.14 m/s each min thereafter. The speed was imposed by audible beeps from pre-recorded audio. Each min stage (level) consisted of multiple "shuttles", and the number depended on the stage speed. Participants were advised to keep running at the pace of the beeps for as long as possible. Once the participant could no longer keep pace with the beeps (*i.e.,* failed to complete two consecutive shuttles in time), the test was terminated (*Green et al., 2013*). For the purpose of the study, we calculated the parameter "Total" as the total number of completed 20-m repetitions (during the whole test). The following parameters were used for further analysis: Total and calculated VO_{2max} (ml · kg⁻¹· min⁻¹). VO_{2max} was estimated from the maximal speed attained during the test *via* the previously developed prediction equation $-24.4 + 6.0 \times$ maximum aerobic speed (sec) (*Léger et al., 1988*).

The speed test involved running 30 m as fast as possible in a straight line between the photocells. Before the test began, the participants stood adjacent to (*i.e.*, their toes were not touching) the starting line in a standing split-stance position. They were instructed to run as fast as possible and slow down after crossing the finish line. A sound signal marked the beginning of each test. The timer was switched on when the starting line was passed, and measurements were automatically taken at 5 m, 10 m, and 30 m by the photocells positioned at those distances. The timer stopped when the finishing line was passed. Each participant ran the course twice, and the mean scores from both were analysed (*Altmann et al., 2019*).

Statistical analysis

Data were analysed using Statistica 13.1 PL (Statsoft, Tulsa, OK, USA) and Excel (Microsoft Corporation, USA) software. Due to the non-normality of the distribution in the Shapiro–Wilk test, we decided to use Spearman's correlation in the analysis. The correlation value (R) was interpreted as follows: 0 to 0.30 or 0 to -0.30 was considered a weak correlation; 0.31 to 0.50 or -0.31 to -0.50 a moderate correlation; 0.51 to 0.70 or -0.51 to -0.70 a strong correlation; and 0.71 to 1 or -0.71 to -1 a very strong correlation (*Hopkins et al., 2009*). The significance level was set at $p \le 0.05$. For the a priori analysis, the sample



size was determined using G*POWER (Version 3.1.9.7; Universität Kiel, Kiel, Germany) using an alpha of 0.05, a power of 0.80, and an effect size of 0.50 for a two-tailed test. Because Spearman's rank correlation coefficient is computationally identical to Pearson's product-moment coefficient, we used the software to calculate the latter.

RESULTS

Participants

Based on the assumptions, the required sample size was determined to be 26. Out of 30 initially invited footballers, 24 met the inclusion criteria. However, during the measurements, two athletes were at a camp with the senior team. Thus, a total of 22 adolescent footballers (two goalkeepers, eight defenders, nine midfielders, three forwards) were included in the final analysis (Fig. 2). Basic data and all parameters measured are shown in Table 1.

Speed test vs US

DA shear modulus at the end of tidal inspiration was moderately negatively correlated with the speed score at 10 m. The DA shear modulus ratio was moderately negatively correlated with the speed score at 10 m and 30 m. The IC shear modulus ratio was moderately negatively correlated with the speed score at 10 m and strongly negatively correlated with the speed score at 30 m. Additionally, DA excursion was positively correlated with the speed score at 5 m (moderate) and 10 m (strong). DA velocity was moderately positively correlated with the speed score at 5 and 30 m, but statistical significance was borderline (p = 0.06). Detailed R values for each correlation are presented in Table 2.

Characteristic $(n = 22)$	mean \pm SD	median
Anthropometric data		
Age (yr)	17.1 ± 0.29	17.0
Body mass (kg)	71.4 ± 7.74	70.0
Body height (cm)	180 ± 5.76	180
BMI (kg/m ²)	22.1 ± 1.95	22.0
Football practice (yr)	7.77 ± 0.75	8.0
SWE - Shear modulus (kPa)		
Diaphragm at the end of tidal inspiration	31.2 ± 6.26	31.5
Diaphragm at the end of tidal expiration	29.4 ± 5.60	27.9
Diaphragm ratio	1.07 ± 0.18	1.05
Intercostal muscle at the end of tidal inspiration	27.1 ± 6.23	26.6
Intercostal muscle at the end of tidal expiration	27.0 ± 6.00	25.7
Intercostal muscle ratio	1.01 ± 0.15	0.97
B-mode –Thickness (mm)		
Diaphragm at the end of tidal inspiration	2.09 ± 0.85	1.82
Diaphragm at the end of tidal expiration	1.71 ± 0.59	1.48
Diaphragm ratio	1.21 ± 0.21	1.20
Intercostal muscle at the end of tidal inspiration	3.98 ± 0.85	4.05
Intercostal muscle at the end of tidal expiration	4.09 ± 0.89	3.97
Intercostal muscle ratio	0.99 ± 0.15	0.95
M-mode		
Diaphragm excursion (cm)	4.73 ± 1.45	4.59
Diaphragm velocity (cm/s)	2.13 ± 0.89	1.83
Multi stage 20-m shuttle run test		
Total	127 ± 13.2	122
calculated VO ₂ max (ml \cdot kg ⁻¹ \cdot min ⁻¹)	56.2 ± 3.54	55.1
Speed test (s)		
Distance 5 m	1.03 ± 0.05	1.03
Distance 10 m	1.87 ± 0.52	1.77
Distance 30 m	4.19 ± 0.20	4.14

 Table 1
 Experimental group characteristics: anthropometric data, ultrasound parameters, endurance test (multi-stage 20-m shuttle run test), and speed test (straight line speed in 5, 10, and 30 m).

Notes.

SD, standard deviation; BMI, Body Mass Index; SWE, shear wave elastography; ratio, diaphragm at the end of tidal inspiration/diaphragm at the end of tidal expiration; Total, total number of completed 20-m repetitions.

MSRT vs US

US parameters were not significantly related to endurance parameters, although correlations varied from weak to moderate. Detailed R values for each correlation are presented in Table 3.

DISCUSSION

The preliminary report was designed to assess the relationship between US of RMs during tidal breathing and selected motor skill (endurance and speed) performance in adolescent football players. To the best of our knowledge, there has not yet been a

Table 2 Correlations between ultrasound parameters and speed test results.

	5	5 m 10		m	30 m	
	R	р	R	р	R	р
Shear modulus						
Diaphragm at the end of tidal inspiration	-0.34	0.12	-0.49	0.02*	-0.24	0.29
Diaphragm at the end of tidal expiration	-0.10	0.66	-0.14	0.55	0.10	0.66
Diaphragm ratio	-0.31	0.16	-0.48	0.02*	-0.41	0.06
Intercostal muscle at the end of tidal inspiration	-0.26	0.26	-0.39	0.08	-0.18	0.44
Intercostal muscle at the end of tidal expiration	-0.13	0.58	-0.16	0.49	0.16	0.48
Intercostal muscle ratio	-0.28	0.22	-0.47	0.03*	-0.54	0.01^{*}
Thickness						
Diaphragm at the end of tidal inspiration	-0.07	0.75	-0.06	0.80	0.22	0.34
Diaphragm at the end of tidal expiration	-0.27	0.23	-0.12	0.60	0.25	0.25
Diaphragm ratio	0.33	0.13	0.07	0.75	-0.03	0.91
Intercostal muscle at the end of tidal inspiration	-0.19	0.42	-0.07	0.78	0.11	0.63
Intercostal muscle at the end of tidal expiration	-0.08	0.74	-0.11	0.64	0.05	0.83
Intercostal muscle ratio	-0.04	0.86	0.14	0.56	0.07	0.76
M-mode						
Diaphragm excursion	0.46	0.04^{*}	0.52	0.02*	0.26	0.27
Diaphragm velocity	0.42	0.06	0.34	0.15	0.42	0.07

Notes.

SWE, shear wave elastography.

*statistically significant p < 0.05.

R, correlation coefficient; p, probability value; ratio, diaphragm at the end of tidal inspiration/diaphragm at the end of tidal expiration.

study relating the shear modulus, thickness, excursion, and velocity of the DA and ICs with parameters of speed and aerobic endurance based on MSRT in adolescent football players. This preliminary study has shown that US of RMs measurements (shear modulus, thickness, excursion, velocity) corresponded to speed in adolescent athletes. Thus, our initial hypothesis was partially confirmed because footballers with higher values of DA shear modulus at the end of tidal inspiration obtained better results in the 10-m speed test. Similarly, a higher DA and IC shear modulus ratio corresponded to a better speed score at 10 and 30 m, and a higher value of DA excursion and velocity was related to worse scores during the speed test. In turn, our results rejected the hypothesis that RMs are related to endurance in adolescent footballers.

Speed

Taking all the results together, our study shows that RM shear modulus during tidal breathing may be partially related to the speed score in adolescent footballers. The shear modulus value is related to passive muscle force (*Koo & Hug*, 2015) and can be used to estimate changes in muscle force (*Ateş et al.*, 2015). *Chino et al.* (2018) showed that DA shear modulus is non-linearly related to inspiratory mouth pressure, increasing rapidly at low inspiratory mouth pressure levels, but less rapidly as mouth pressure reaches higher levels. It can therefore be stated that a higher value of the DA shear modulus indicates greater inspiratory muscle strength. Another study confirmed that DA stiffness increases during

	Total		VO ₂ max	
	R	р	R	р
Shear modulus				
Diaphragm at the end of tidal inspiration	-0.16	0.49	0.07	0.76
Diaphragm at the end of tidal expiration	-0.17	0.46	-0.05	0.83
Diaphragm ratio	0.03	0.88	0.18	0.42
Intercostal muscle at the end of tidal inspiration	0.03	0.90	0.33	0.15
Intercostal muscle at the end of tidal expiration	-0.05	0.84	0.17	0.47
Intercostal muscle ratio	0.18	0.43	0.32	0.16
Thickness				
Diaphragm at the end of tidal inspiration	0.01	0.98	0.17	0.45
Diaphragm at the end of tidal expiration	0.10	0.66	0.29	0.18
Diaphragm ratio	-0.14	0.53	-0.12	0.59
Intercostal muscle at the end of tidal inspiration	0.20	0.40	0.36	0.11
Intercostal muscle at the end of tidal expiration	-0.01	0.97	0.10	0.66
Intercostal muscle ratio	0.16	0.48	0.25	0.27
M-mode				
Diaphragm excursion	0.19	0.41	-0.03	0.91
Diaphragm velocity	0.20	0.41	0.17	0.49

 Table 3
 Relationship between ultrasound parameters and endurance test (multi-stage 20-m shuttle run) results.

Notes.

SWE, shear wave elastography; Total, total number of completed 20-m repetitions; VO_2max , calculated VO_2max (ml · kg⁻¹ · min⁻¹); R, correlation coefficient; p, probability value; ratio, diaphragm at the end of tidal inspiration/diaphragm at the end of tidal expiration.

inspiration (*Sendur et al., 2022*). Our study shows that a stiffer (higher shear modulus value) DA during tidal inspiration characterised athletes with a better score in the speed test. This may indicate that a stiffer DA improves speed performance.

The DA shear modulus value is also related to transdiaphragmatic pressure (*Bachasson et al., 2018*), which is considered the gold standard for DA examination (*Ricoy et al., 2019*). Transdiaphragmatic pressure is the main measurement for determining DA strength (*Hamnegard et al., 1995*) and is clinically relevant because it represents the actual force that drives changes in lung volume and therefore ultimately alveolar ventilation (*Bachasson et al., 2018*). Sprint running (up to 6 s/up to 40 m) is characterised by anaerobic effort (*Sanders et al., 2017*). In our study, therefore, it can be assumed that the athletes had an anaerobic effort at the 30-m distance, so they were running at apnoea. It has been suggested that there is increased chest pressure during the initial phase of the speed test, which is linked to the Valsalva test (*Turban, 2010*). The Valsalva manoeuvre initiates with deep inhalation and DA downward movement (*Talasz et al., 2012*). Thus, the DA seems to be the main muscle involved in the Valsalva manoeuvre. The increased DA shear modulus during tidal breathing may predispose to a stronger DA contraction during the speed trial, resulting in a better score in the initial phase of running.

At a distance of 30 m, the DA and IC shear modulus ratio seems to be more significant. The ratio is calculated by dividing the shear modulus value at the peak of tidal inspiration by the shear modulus value at the peak of tidal expiration. In our study, the higher the DA and IC shear modulus ratio, the better the speed test score. The ratio score is therefore determined not only by the shear modulus value during inspiration but also during expiration. This means that the best speed scores were achieved by athletes who had a higher RM shear modulus value during tidal inspiration and simultaneously a lower RM shear modulus value during tidal expiration. It may be that a better ability to relax the RMs allows for their greater contraction. When a muscle lengthens, the muscle spindle located inside the muscle is stretched, causing the muscle fibres to contract (*Bhattacharyya, 2017*). In turn, the comparable correlation values between each of the RMs and speed is probably due to the similar function of the DA and ICs. These muscles both affect chest movement (*Ratnovsky, Elad & Halpern, 2008*), produce axial rotations of the thorax (*Whitelaw et al., 1992*), and are important respiratory pump muscles (*Han et al., 1993*). Consequently, their work must be coordinated (*Han et al., 1993*). In addition, although the DA is the main RM, when the respiratory workload increases (high breathing efforts), the activity of ICs plays an important role (*Ratnovsky, Elad & Halpern, 2008*).

In view of the previous considerations, it is difficult to explain why footballers characterised by greater DA excursion and velocity during maximal inspiration had worse running scores. It was assumed that the increased stiffness of the DA during tidal breathing allowed greater stiffness of the DA during the Valsalva test because greater stiffness may result in lower DA excursion and velocity. Unfortunately, there are no studies connecting US assessment of RMs to speed in athletes, which greatly limits the interpretability of these preliminary findings.

Endurance

Some studies have shown that exercises involving the RMs improve endurance by reducing energy demand (*Bahenský et al., 2021*) and increase aerobic tolerance (*Mackała et al., 2020*) in youth athletes. It has also been indicated that breathing technique can affect endurance through reduced respiratory work and delayed RM fatigue (*Bahenský et al., 2021*). This was the reason we hypothesised that endurance should be related to US of RMs in our study. This was not confirmed, as there was no relationship between the endurance and US parameters of RMs. In cited studies (*Bahenský et al., 2021*; *Mackała et al., 2020*), RMs strength was measured indirectly by analysing maximal inspiratory and expiratory pressure/forces. In the present study, for the first time, we have evaluated and related RMs with endurance directly by analysing US measurements (shear modulus, thickness, excursion, and velocity). An indirect method of assessing respiratory function is the result of many factors (including airway obstruction, respiratory compliance, and RM strength) that do not allow direct analysis of the RMs (*Pałac & Linek, 2022*). This may mean that the improvement in endurance in athletes is a more complex phenomenon unrelated to an exclusive change in RM morphology.

It is particularly surprising that there was no correlation between DA excursion and aerobic endurance in the present study. DA excursion is related to exercise capacity (*Shiraishi et al., 2020*) and can predict the improvement in exercise tolerance (*Shiraishi et al., 2020*) in patients (especially with problems related to the respiratory system). DA

excursion is related to pulmonary parameters like FVC, FEV1, and MIP, whereas DA velocity is related to FVC, MIP, and MEP (*Palac & Linek*, 2022). All of these spirometry parameters are related to RM strength (*Palac & Linek*, 2022). Thus, it was expected that greater DA excursion would predispose to better endurance in examined football players. Possibly in healthy people (and athletes who achieve higher performance in endurance tests than the non-athlete population), the DA excursion is not as important in order to improve endurance. An alternative explanation of the lack of correlation between DA excursion and aerobic endurance may be the relatively similar endurance (training) level of the footballers studied. However, there is a lack of scientific studies determining the significance of DA excursion in athletes. Hence, the present study results are difficult to interpret definitively.

Limitations

Due to the small sample size, this study is of a preliminary nature. The study group consisted exclusively of football players from one team and age group, which may explain the high homogeneity of the participants' motor skills and US parameters. This, in turn, may have influenced the narrow dispersion of the variables and, ultimately, the correlation values. The results should not therefore be generalised to other sports. The participants were included in the analysis regardless of their position; studies have shown that footballers' profiles can vary according to where they play on the pitch (Oliva-Lozano et al., 2020). US examinations were performed only in the supine position. Another limitation was the collection of US measurements only during tidal breathing (except for excursion-maximal inspiration and tidal expiration). It seems necessary to include US assessment of the RMs during maximal respiratory efforts in future studies. For the purposes of this study, the athletes' endurance was indirectly determined. The MSRF is used as a test of aerobic capacity (Voss & Sandercock, 2009). The beep test can be used as a health indicator in children and adolescents (Mayorga-Vega et al., 2016), but it is a field test. Thus, the result should not be interpreted as a direct measurement of cardiorespiratory fitness, only as an estimation (Mayorga-Vega et al., 2016).

Strength and implications

To date, RMs have never been directly investigated in the context of their association with athletes' performance. Although this is a pilot study, we have shown for the first time that some US parameters of the RMs may be related with motor skills (like speed in our study). From this perspective, we have confirmed that such exploration is justified. US provides an inexpensive and non-invasive tool for assessing RMs on wide populations. The methodology used in this report to assess RMs is easy accessible and reliable. Thus, it seems that the US of RMs in elite athletes is warranted in order to provide deeper insights into the role of RMs in the context of different motor abilities. Previous studies have confirmed the relationship between athletic performance and US parameters of lower-limb muscles (*Sarto et al., 2021*). It is also worth noting that RMs (mainly DA) function itself is related to pain sensation, stability, and balance. All these aspects are important in high-performance sport.

CONCLUSIONS

Shear modulus of the RMs, DA excursion, and velocity are related to speed score in adolescent football players. In the examined population, endurance parameters were not related to any US parameters of RMs. The current state of knowledge does not allow us to conclusively determine how important US parameters of RMs can be in predicting performance parameters (for example endurance and speed) in young athletes. However, the results of the present study point to the need for further research into the role of US measurements of RMs in the development of motor skills.

ADDITIONAL INFORMATION AND DECLARATIONS

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Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Małgorzata Pałac conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Damian Sikora performed the experiments, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Tomasz Wolny performed the experiments, authored or reviewed drafts of the article, and approved the final draft.
- Paweł Linek conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.

Human Ethics

The following information was supplied relating to ethical approvals (i.e., approving body and any reference numbers):

The study was approved by the Ethics Committee at the Jerzy Kukuczka Academy of Physical Education in Katowice

Data Availability

The following information was supplied regarding data availability: The raw data is available in the Supplementary File.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.15214#supplemental-information.

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