



Can sonographic assessment of pulmonary vascular reactivity following maternal hyperoxygenation predict neonatal pulmonary hypertension? (HOTPOT study protocol)

Ann McHugh^{a,*}, Orla Franklin^b, Afif El-Khuffash^c, Fionnuala Breathnach^a

^a Department of Obstetrics and Gynaecology, Royal College of Surgeons in Ireland, Rotunda Hospital, Dublin, Ireland

^b Department of Paediatric Cardiology, Our Lady's Children's Hospital, Crumlin, Dublin, Ireland

^c Department of Neonatology, Royal College of Surgeons in Ireland, Rotunda Hospital, Dublin, Ireland

ARTICLE INFO

Keywords:

Fetal cardiology
Hyperoxygenation
Maternal haemodynamics
Non-invasive cardiac output monitor
Persistent pulmonary hypertension of the newborn

ABSTRACT

Background: Persistent pulmonary hypertension of the newborn (PPHN) is a condition that occurs in 0.5–7 per 1000 live births and can result in significant cardiovascular instability in the newborn. It occurs when there is a failure of the normal circulatory transition in the early newborn period. Recent studies have shown that fetal pulmonary vasculature reacts to maternal hyperoxygenation (MH). The aim of the study is to assess if the in-utero response to MH can predict pulmonary hypertension in the early newborn period.

Methods: We will perform a prospective cohort study. It will evaluate the use of fetal echocardiographic Doppler assessment of the pulmonary vasculature prior to and following MH to predict fetuses that may develop pulmonary hypertension in the neonatal period. The study will be undertaken in the Rotunda Hospital, Dublin, Ireland. A fetal ultrasound and echocardiography will be performed on fetuses in the third trimester. Blood flow velocity waveforms will be recorded during periods of fetal quiescence. Pulsatility index (PI), Resistance index (RI), Peak systolic (PSV) and end diastolic velocity (EDV), time-averaged velocity (TAV), acceleration time (AT), and ejection time (ET) will be measured within the fetal distal pulmonary artery (PA). The acceleration-to-ejection time ratio (AT: ET) will be used to assess pulmonary vascular resistance (PVR). Doppler measurements will be taken at baseline and repeated immediately following MH for 10 min (O₂ 100% v/v inhalational gas) at a rate of 12L/min via a partial non-rebreather mask. Doppler waveform measurements from the umbilical artery (UAD), middle cerebral artery (MCA) ductus arteriosus (DA), aortic isthmus (AoI) and ductus venosus (DV) will also be obtained. After birth, a comprehensive neonatal functional echocardiogram will be performed within the first 24 hours of life.

Discussion: This study proposes to validate methods described to date in investigating the fetal pulmonary vascular response to MH, with expansion of the study subjects to include fetuses at risk of PPHN. Evaluation of the different at-risk subgroups will be informative in relation to the fetal circulatory adaptation close to term. Prediction of neonatal pulmonary hypertension may help guide the pharmacological and neonatal ICU strategies that optimise postnatal survival.

1. Background

Persistence of the fetal circulation results in pulmonary hypertension, reduced oxygen saturation and may result in right-to-left shunting of blood in the newborn heart. It results in a mortality ranging between 4 and 33% [1–3]. Some degree of pulmonary hypertension complicates the course of more than 10% of all neonates with respira-

tory failure. Increased PVR in the newborn can produce intracardiac shunting of blood which can lead to severe hypoxemia and significant morbidity and mortality. Pulmonary hypertension may be associated with pulmonary hypoplasia when diminished surface area for gas exchange and inadequate pulmonary blood flow lead to hypoxia and remodeling of the resistant pulmonary arterioles [2].

* Corresponding author.

E-mail address: mchughaf@tcd.ie (A. McHugh).

<https://doi.org/10.1016/j.conctc.2020.100610>

Received 25 March 2020; Received in revised form 23 June 2020; Accepted 5 July 2020

Available online 12 July 2020

2451-8654/© 2020 The Authors.

Published by Elsevier Inc.

This is an open access article under the CC BY-NC-ND license

(<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Several methods have been proposed to assess fetal lung volume as a proxy indicator of pulmonary function [4]. Some of these methods are time consuming to perform and there can be considerable variability in measurements between different ultrasonographers. One previously described method of measuring the chest circumference to abdominal circumference ratio cannot be used in fetuses with large abdominal circumferences, therefore fetuses with polycystic kidneys, obstructive uropathy or omphaloceles would be excluded [5].

Recent studies have shown that the fetal pulmonary vasculature reacts to MH [6–8]. Following maternal oxygen administration, a decrease in the PVR as demonstrated by the pulmonary artery Doppler, is deemed to indicate vasoreactivity in the pulmonary vascular bed [9]. Small studies to date indicate that a lack of vasoreactivity in response to MH, may serve as a useful clinical tool in predicting lethal pulmonary hypoplasia in those at-risk fetuses [10,11]. The measurement of peripheral pulmonary velocity waveforms before and after MH may therefore help in determining the risk of developing PPHN. The ability to accurately predict the occurrence of PPHN by a method that is non-invasive and reproducible would be extremely beneficial in both obstetric management and in parental counseling.

2. Study hypothesis

The overall hypothesis of the study is that a lack of reactivity in response to MH, as assessed using pulmonary artery Doppler ultrasound can predict the presence of PPHN in the early newborn period.

This study proposes to validate methods previously described in investigating the fetal pulmonary vascular response to MH.

To evaluate our study hypothesis, we will ascertain what fetal response occurs in-utero following MH. We will establish if any changes can be identified on neonatal echocardiography in those fetuses that responded to MH in-utero. We will compare those findings with the findings in fetuses that did not respond to MH.

3. Primary objective

The overall aim of the study is to assess the ability of the hyperoxygenation test to predict the presence of PPHN in the early newborn period.

The primary outcome of interest is the presence of PPHN on Day 1 of life.

4. Defining PPHN

Persistent pulmonary hypertension will be defined by echocardiography as well as by clinical indicators as follows:

- 1) A requirement of at least 0.4 Fractional Inspired Oxygen to maintain a preductal saturation of $\geq 95\%$; and,
- 2) Normal structural anatomy of the heart on echocardiogram; and,
- 3) In the presence of a tricuspid regurgitant (TR) jet, an estimated right ventricular systolic pressure (using the Bernoulli Equation) $\geq 50\%$ of the systemic systolic pressure measured at the start of the echocardiogram; or
- 4) In the presence of a patent ductus arteriosus (PDA of a low velocity shunt across the PDA from left to right such that) the estimated right ventricular/pulmonary artery pressures was $> 50\%$ systemic
- 5) In the absence of a TR jet or a PDA, an intraventricular septum bowing into the left ventricular cavity.

Secondary outcomes will include the following:

Neonatal respiratory morbidity in all neonates.

(Including oxygen requirement, intubation and continuous positive airway pressure (CPAP) requirement).

5. Methods

5.1. Overview

This prospective cohort study will be undertaken in the Department of Obstetrics and Gynecology in the Rotunda Hospital Dublin, Ireland. The Rotunda Hospital is a tertiary-level, stand-alone maternity hospital in Dublin, with over 8500 deliveries per year. There is a large Maternal Fetal Medicine unit and a neonatology department that accepts national referrals, with over 1500 admissions to the neonatal unit per year.

5.2. Study population

Two groups of pregnant participants in the third trimester will be recruited to the study, those at risk of PPHN and a group of gestational age matched controls. Participants will be identified through the hospital records system (Current inpatients, ultrasound department [anatomy scans], fetal medicine meetings), and will be offered participation in the study.

5.3. Inclusion criteria

The following groups of participants will be recruited to the study:

5.4. Group A

Women who are carrying a fetus at risk of pulmonary hypoplasia: including those with mid-trimester preterm prelabour rupture of membranes (PPROM), congenital diaphragmatic hernia (CDH) and skeletal dysplasia.

This group has been chosen due to the increased risk of PPHN in this cohort. The critical phase in fetal lung development is between 16 and 28 weeks' gestation [1]. If a PPROM occurs prior to 26 weeks gestation, fetal lung development can be impaired and result in pulmonary hypoplasia [2]. Congenital diaphragmatic hernia is associated with pulmonary hypoplasia and pulmonary hypertension. Pulmonary hypertension in CDH is driven by lung hypoplasia and may result in alterations in the pulmonary vasculature and in the pulmonary vasoreactivity [3]. Skeletal dysplasia is associated with PPHN and many complex respiratory complications due to small and poorly compliant chest wall movements, airway anomalies, pulmonary hypoplasia, and central apnea [4].

5.5. Group B

Women attending for scheduled Caesarean section (CS) prior to 38 weeks' gestational age.

This group has been chosen for recruitment following numerous publications that have reported higher rates of PPHN in neonates delivered by CS compared to those born by vaginal delivery [25][26] Caesarean sections have been reported to carry an approximately five-fold higher risk for PPHN when compared to vaginal deliveries [5]. In addition, higher rates of RDS and concomitant increases in endothelin-1 levels have been reported in babies born by CS, which might indirectly lead to a higher risk of developing PPHN [6].

5.6. Group C

In addition, a group of gestation-matched uncomplicated singleton pregnancies will be recruited to serve as a control group. This group

has been included to provide information on the in-utero response to MH that occurs in normal pregnancies.

- Subjects must be able and willing to give written informed consent and to comply with the requirements of the study protocol.
- Subjects must be female, aged 18 years or above at baseline.
- Subjects who are judged to be in generally good health by the investigator based upon the results of the medical history.

5.7. Exclusion criteria

- Age <18 years
- Known fetal chromosomal abnormality excluding Trisomy 21
- Gestational age <18 weeks and >40 weeks
- Maternal chronic respiratory disease (including COPD, Cystic Fibrosis, Pulmonar Fibrosis)
- Maternal congenital heart disease (CHD)
- Maternal use of bleomycin or amiodarone (due to interactions with oxygen)
- Subjects unable to provide written informed consent
- Subjects who have any other significant medical disease or disorder (including uncontrolled diabetes, unstable ischemic heart disease, moderate to severe congestive heart failure, recent cerebrovascular accident) which, in the opinion of the investigator, may either put the subject at risk by participation in the study, or may influence the result of the study.

5.8. Justification for inclusion and exclusion criteria

Fetuses' ≥ 31 weeks' gestation will be included as the hyperoxygenation test is known to become responsive after this gestational age (GA) [7]. Those with a GA >40 weeks will be excluded given the potential for advanced GA to effect the acquisition of or the result of various Doppler indices. A window of 31–40 weeks GA was chosen to increase uniformity and to acquire better data. Participant's with a non-smoking status were chosen given the hazards associated with smoking and high flow oxygen [8] and to eliminate any effect that smoking may have on Doppler velocity waveforms [9–11]. Although many women with CHD can go through pregnancy with a low risk to themselves, there remains a higher incidence of miscarriage, premature births, low birth weights and an increase of CHD in the fetus, in women with cyanotic CHD than that found in the normal population [12]. Notwithstanding that there is an increased risk of cardiac and neonatal complications associated with maternal CHD in pregnancy [13], there is limited data on the haemodynamic changes in pregnancy in response to MH and for this reason we have excluded both women with CHD and those with any chronic respiratory disease.

5.9. Study procedure

Doppler echocardiography will be performed on fetuses between 31 and 40 weeks gestation. A fetal echocardiogram will be performed according to an agreed protocol to exclude any major structural defect. Image-directed pulsed and colour Doppler equipment (Voluson E8, GE Healthcare) with a 5 MHz sector probe will be used to obtain blood velocity waveforms. The lowest high-pass filter level (100 Hz) will be used and the spatial peak temporal average power output for colour and pulsed Doppler kept at <100 mW/cm. An angle of insonation <15° between the vessel and Doppler beam as assessed by colour Doppler will be accepted for analysis. The fetal pulmonary artery (PA) will be visualised by rotating the transducer from the four-chamber view to the short-axis view of the fetal heart. The pulmonary valves and the bifurcation of the right and left branches of the PA will be identified. The distal PA (DPA) can be located beyond the first bifurcation of the branch PA and this is the area that will be used for as-

essment, in keeping with previous studies [7]. The following measurements specific to the branch PA Doppler waveform will be recorded:

1. The peak systolic velocity (PSV)
2. End-diastolic velocity (EDV)
3. The pulsatility index (PI; defined as the difference between peak systolic and diastolic velocity divided by time averaged velocity)
4. The resistance index (RI; defined as the difference between the peak systolic and diastolic velocity divided by the peak systolic velocity)
5. The ejection time (ET; defined as the whole time of systole)
6. The acceleration time (AT; defined as the time from the initial increase in velocity to the time of peak velocity)

The PA PI and PA RI measurements are markers of vascular resistance and both require PSV and EDV measurements for their calculation. The AT and ET measurements will serve as markers for pulmonary vascular resistance. Following MH, a repeat fetal echocardiogram will be performed. The hyperoxygenation test will be considered positive when the fetal PA PI decreases by more than 10% from its baseline (responders), in keeping with a previous study [14]. Where the fetal PA PI does not decrease by at least 10%, cases will be classified as non-responders.

Measurements of the UA, MCA, DA, AoI and DV will also be obtained pre and post MH. After a minimum of three uniform waveforms are obtained the Doppler images from the UA, MCA, DA, AoI, DV and PA will be frozen and saved to the ultrasound machine for future analysis. The measurements will be averaged from the values obtained from the three best cardiac cycles, using a conventional computerised programme linked to the equipment that calculated the PSV, EDV and TAmx velocities, PI (PIPSV-EDV/TAMXV) and RI (RI = PSV-EDV/PSV). These Dopplers will be included to assess the utero-placental response and to evaluate for ductal constriction secondary to MH. The UA Doppler will be obtained from a free loop of cord. The MCA Doppler will be sampled in the area overlying the anterior wing of the sphenoid bone near the base of the skull [15]. The DA waveform will be obtained in either the three-vessel trachea view or the longitudinal ductal arch (LDA) view [16]. The AoI Doppler waveform will be obtained in the longitudinal aortic arch (LAA) view or from the three vessel and tracheal view [17] and the DV Doppler waveform will be obtained at the isthmus, near its origin from the umbilical vein in a mid-sagittal or cross-sectional abdominal plane [18]. All Doppler images will be frozen on the ultrasound machine and stored for future analysis and for quality control purposes. Only images that are deemed satisfactory by the study investigators will be included in the study data.

Amniotic fluid indices, routine fetal biometry and estimated fetal weight measurements based on the Hadlock formula [19] will be obtained as well as fetal heart rate variation. The study participants will not be made aware of the result of the hyperoxygenation test during the study period.

To assess the reproducibility of the measured PA parameters, velocity waveforms from the PA of 10 fetuses not recruited to the study will be recorded. Intraobserver and interobserver variability of PA indices will be assessed using this subset of 10 patients. One reader (A.M) will repeat measurements at a time temporally remote from the initial assessment (approximately 15 minutes). To assess interobserver variability, a second reader (F.B), blinded to the original data, will repeat PA measurements.

5.10. Oxygen therapy

Oxygen will be administered to the patients while in a semi-recumbent position in the hospital ultrasound department. Oxygen will

be administered at a rate of 12L/min for a duration of 10 minutes via a non-rebreather mask. The ultrasound department in our hospital is equipped to administer oxygen to the patients, using a portable oxygen cylinder and disposable plastic non rebreather masks.

5.11. Investigational medicinal product (IMP)

The trade name of the medicinal product is medical oxygen. The name of the active substance is oxygen. The formulation is 100% V/V inhalational gas. The marketing authorisation number in Ireland is PA1357/001/001. The marketing authorisation holder is Industrial Pressure testing Ltd. The dose will be 60% FiO₂ administered via a face mask over 10 minutes. This dosing has been used previously without any adverse effects [20–22]. Oxygen will be stored in appropriate oxygen cylinders.

5.12. Neonatal echocardiogram

The presence of pulmonary hypertension in the neonate will be formally assessed with a neonatal echocardiogram performed within the first 24 hours of life. Evaluations will be performed using the Vivid echocardiography system (GE Medical, Milwaukee) and a cardiology multi-frequency probe. All studies on asymptomatic infants will be performed in a dedicated quiet room on the postnatal ward when the infant is in a quiet state ideally after feeds. No sedation will be used. If the infant is admitted to the neonatal intensive care unit, the echocardiography study will be performed there. All scans will be recorded on the machine's internal hard drive and transferred to the EchoPAC archiving system for offline measurements and validation. All studies will be archived and reviewed later to assess quality and accuracy of data acquisition. Studies will be performed using standard neonatal windows including apical, parasternal, subcostal, and high parasternal windows. The archiving system is also available at the Rotunda Hospital and has enough storage capacity for 15 years with an activity of 300 scans per year. All studies will be performed by a neonatologist with experience in neonatal functional echocardiography. The person performing the neonatal echocardiogram will be blinded to the prenatal ultrasound echocardiogram findings and to the response of the fetus to the hyperoxygenation test. The following echocardiography parameters will be measured using previously described methods [23], left ventricular (LV) length measured at end diastole; mitral valve annular diameter; left ventricular output (LVO, ml/kg/min); Ejection fraction using Simpson's Biplane method (EF, %); mitral valve inflow velocities and velocity time index, PDA diameter (mm) measured in 2D at the pulmonary end; diastolic and systolic flow velocity across the PDA; flow pattern across the duct; pulmonary artery acceleration time (PAAT); and right ventricular (RV) end systolic pressure measured using the tricuspid valve regurgitant jet.

All neonates will be offered a follow-up appointment in the outpatient's department for review and a repeat neonatal echocardiogram at six weeks of age.

5.13. Data collection and retention

Both a hard copy and electronic versions of the data will be retained. Electronic data will be retained on a research computer database within the Rotunda Hospital. All data will be maintained on password protected computers and no patient names will be recorded. Source documents for this study will include hospital records, ultrasound procedure reports and data collection forms. These documents will be used to enter data onto the case report files (CRF). Data reported on the CRF that are derived from source documents must be consistent with the source documents or the discrepancies must be explained. All documents will be stored safely in confidential conditions. On all study-specific documents other than the signed consent, the

subject will be referred to by a study subject identification number. All fetal ultrasound images will be archived on the ultrasound machine from which they were taken. All records and documents will be maintained by the investigator for a period of at least 2 years.

5.14. Adverse events (AEs)

Probable AEs associated with oxygen administration specifically, which occur during the study will be recorded on the CRF. These will include those observed by the investigator or reported by the subject. These will include ductal constriction, increase in tricuspid regurgitation and any other reported events. If an AE occurs, the following information will be recorded: description, date of onset and end date, severity, assessment of relatedness to the study medication, other suspect medication or device and action taken and outcome. All AEs will be captured up to 6 weeks postpartum.

5.15. Statistics

A power calculation was performed for our study. Using an anticipated incidence of PPHN in a high risk cohort of 0.7% compared to a background incidence in the general population of 0.05% [24], 308 participants will be required for this study (80% confidence and a type I error rate of 0.05).

Descriptive statistics will be used to summarise the findings into two groups responders and non-responders. Descriptive statistics will be used to analyse the different pathologies from group A (i.e CDH, mid-trimester PPRM and skeletal dysplasia) given their heterogeneity. The primary analysis of the primary outcome will be performed using an independent *t*-test (or a Wilcoxon Rank Sum test as appropriate) to compare the presence of PPHN in infants with and without a normal MH test. A chi squared test will be used for the primary analysis of the dichotomous secondary outcomes. For the continuous secondary outcomes, a *t*-test will be used to compare normally distributed data, and Wilcoxon Rank Sum test will be used for skewed data. We will accept a *p* value of <0.05 as significant. We will use SPSS (version 24.0) to perform the statistical analysis. Intraobserver and interobserver variability will be assessed using the intraclass correlation coefficient (ICC).

5.16. Informed consent

Informed consent will be taken at the subject's recruitment visit to the ultrasound department when they attend for the hyperoxygenation test. The subject will have received an information leaflet and the consent form prior to attending the ultrasound department giving them adequate time to make an informed decision on partaking in the study. Prior to any study related screening procedures being performed on the subject, the informed consent statement will be reviewed, signed and dated by the subject and the person who administered the informed consent form.

6. Discussion

We do not foresee any undue risk to the subjects. Oxygen is safe to use in pregnancy and is not known to cause any long-term adverse maternal or fetal effects. It will be administered for a total duration of 10 minutes. The communication of outputs and achievements from this study will be disseminated to the following target audiences - specialist healthcare professionals, the general medical and scientific community, study participants, patient groups, and the wider community. Information about the project and its results will be disseminated at international conferences and seminars.

Despite recent advances in the management of PPHN, the risk of mortality and adverse neurological sequelae remain high. By under-

taking this study, we may be able to identify a way of accurately predicting the outcomes in babies at risk of PPHN. The study may impact our clinical management of these pregnancies in the future. The study may also impact our routine care of babies at risk of PPHN, in that; they may all undergo an echocardiogram and maternal hyperoxygenation test in the future. This may help in identifying those fetuses with a poorer neonatal outcome.

Trial insurance

Royal College of Surgeons in Ireland.

Data monitor

Ms Mandy Jackson. RCSI. E-mail: mandyjackson@rcsi.ie. Tel: 01 8093863 fax: 01 809 3809.

Ethics

Approved by the National Maternity Hospital Ethics Committee September 2016, Dublin, Ireland. This is a national ethics committee which provides ethical approval for studies being performed in other regional maternity hospitals.

Availability of data and material

Full trial protocol available at request of Principal Investigator Prof Fionnuala Breathnach, fbreathnach@rcsi.ie. Trial data will be made available on request once the trial commences and data is generated.

Funding

The Rotunda Foundation, Pillar Room, Rotunda Hospital, Parnell Street, Dublin 2. Registered. Charity (CHY20091). Tel: (01) 872 2377.

Author's contributions

Conception of this work by AM and FB.

Trial sponsor

Royal College of Surgeons in Ireland, 123 St Stephen's Green, Dublin 2.

Role of Sponsor and Funders in data collection and analysis.

Neither the sponsor nor the funder had any role in trial design. Neither the sponsor nor the funder will have any role in: data collection; data analysis; data interpretation; report writing; or the decision to submit the manuscript for publication.

Trial registration

EnduraCT Number 2016-003181-12.

Declaration of competing interest

The authors declare that they have no conflict of interest.

Acknowledgements

The author would like to acknowledge the financial support from the Rotunda Foundation.

References

- [1] A.S. van Teeffelen, J. Van Der Heijden, S.G. Oei, M.M. Porath, C. Willekes, B. Opmeer, et al., Accuracy of imaging parameters in the prediction of lethal pulmonary hypoplasia secondary to mid-trimester prelabor rupture of fetal membranes: a systematic review and meta-analysis, *Ultrasound Obstet. Gynecol.* 39 (5) (2012) 495–499.
- [2] H.W. Kilbride, D.W. Thibeault, Neonatal complications of preterm premature rupture of membranes. Pathophysiology and management, *Clin. Perinatol.* 28 (4) (2001) 761–785.
- [3] M.T. Harting, Congenital diaphragmatic hernia-associated pulmonary hypertension, *Semin. Pediatr. Surg.* 26 (3) (2017) 147–153.
- [4] D. Alapati, T.H. Shaffer, Skeletal dysplasia: respiratory management during infancy, *Respir. Med.* 131 (2017) 18–26.
- [5] E.M. Levine, V. Ghai, J.J. Barton, C.M. Strom, Mode of delivery and risk of respiratory diseases in newborns, *Obstet. Gynecol.* 97 (3) (2001) 439–442.
- [6] N. Babooa, W.-J. Shi, C. Chen, Factors relating caesarean section to persistent pulmonary hypertension of the newborn, *World Journal of Pediatrics* 13 (6) (2017) 517–527.
- [7] J. Rasanen, D.C. Wood, R.H. Debbs, J. Cohen, S. Weiner, J.C. Huhta, Reactivity of the human fetal pulmonary circulation to maternal hyperoxygenation increases during the second half of pregnancy: a randomized study, *Circulation* 97 (3) (1998) 257–262.
- [8] W.G. Carlos, M.S. Baker, K.A. McPherson, G.T. Bosslet, R. Sood, A.M. Torke, Smoking-related home oxygen burn injuries: continued cause for alarm, *Respiration* 91 (2) (2016) 151–155.
- [9] C.A. Albuquerque, K.R. Smith, C. Johnson, R. Chao, R. Harding, Influence of maternal tobacco smoking during pregnancy on uterine, umbilical and fetal cerebral artery blood flows, *Early Hum. Dev.* 80 (1) (2004) 31–42.
- [10] P.J. Pringle, M.P. Geary, C.H. Rodeck, J.C. Kingdom, S. Kayamba-Kay's, P.C. Hindmarsh, The influence of cigarette smoking on antenatal growth, birth size, and the insulin-like growth factor axis, *J. Clin. Endocrinol. Metab.* 90 (5) (2005) 2556–2562.
- [11] E.M. Kho, R.A. North, E. Chan, P.R. Stone, G.A. Dekker, L.M. McCowan, Changes in Doppler flow velocity waveforms and fetal size at 20 weeks gestation among cigarette smokers, *Bjog* 116 (10) (2009) 1300–1306.
- [12] P. Presbitero, J. Somerville, S. Stone, E. Aruta, D. Spiegelhalter, F. Rabajoli, Pregnancy in cyanotic congenital heart disease. Outcome of mother and fetus, *Circulation* 89 (6) (1994) 2673–2676.
- [13] P. Khairy, D.W. Ouyang, S.M. Fernandes, A. Lee-Parritz, K.E. Economy, M.J. Landzberg, Pregnancy outcomes in women with congenital heart disease, *Circulation* 113 (4) (2006) 517–524.
- [14] A. Szwast, Z. Tian, M. McCann, D. Donaghue, J. Rychik, Vasoreactive response to maternal hyperoxygenation in the fetus with hypoplastic left heart syndrome, *Circ Cardiovasc Imaging* 3 (2) (2010) 172–178.
- [15] J.W. Wladimiroff, J.A. vd Wijngaard, S. Degani, M.J. Noordam, J. van Eyck, H.M. Tonge, Cerebral and umbilical arterial blood flow velocity waveforms in normal and growth-retarded pregnancies, *Obstet. Gynecol.* 69 (5) (1987) 705–709.
- [16] Z. Gou, J. Zhang, X. Yan, Z. Wang, S. Li, X. Deng, Variations in ductus arteriosus Doppler parameters in different sonographic views during the second half of gestation, *Exp. Ther. Med.* 17 (1) (2019) 502–506.
- [17] M. Del Rio, J.M. Martinez, F. Figueras, M. Bennasar, M. Palacio, O. Gomez, et al., Doppler assessment of fetal aortic isthmus blood flow in two different sonographic planes during the second half of gestation, *Ultrasound Obstet. Gynecol.* 26 (2) (2005) 170–174.
- [18] T.W. Huisman, P.A. Stewart, J.W. Wladimiroff, Ductus venosus blood flow velocity waveforms in the human fetus—a Doppler study, *Ultrasound Med. Biol.* 18 (1) (1992) 33–37.
- [19] F.P. Hadlock, R.B. Harrist, R.J. Carpenter, R.L. Deter, S.K. Park, Sonographic estimation of fetal weight. The value of femur length in addition to head and abdomen measurements, *Radiology* 150 (2) (1984) 535–540.
- [20] R.H. Kallet, M.A. Matthay, Hyperoxic acute lung injury, *Respir. Care* 58 (1) (2013) 123–141.
- [21] N.T. Bateman, R.M. Leach, ABC of oxygen. Acute oxygen therapy, *BMJ (Clinical research ed)* 317 (7161) (1998) 798–801.
- [22] B.R. O'Driscoll, L.S. Howard, A.G. Davison, BTS guideline for emergency oxygen use in adult patients, *Thorax* 63 (Suppl 6) (2008) vi1–vi68.
- [23] A.M. Groves, Y. Singh, E. Dempsey, Z. Molnar, T. Austin, A. El-Khuffash, et al., Introduction to neonatologist-performed echocardiography, *Pediatr. Res.* 84 (Suppl 1) (2018) 1–12.
- [24] A. El-Khuffash, P.J. McNamara, C. Breatnach, N. Bussmann, A. Smith, O. Feeney, et al., The use of milrinone in neonates with persistent pulmonary hypertension of the newborn - a randomised controlled trial pilot study (MINT 1): study protocol and review of literature, *Matern. Health Neonatol. Perinatol.* 4 (2018) 24.
- [25] K.C. Winovitch, L. Padilla, M. Ghamsary, D.C. Lagrew, D.A. Wing, Persistent pulmonary hypertension of the newborn following elective caesarean delivery at term, *J Matern Fetal Neonatal Med* 24 (11) (2011) 1398–402.413.
- [26] A.K. Hansen, K. Wisborg, N. Ulbjerg, T.B. Henriksen, Elective caesarean section and respiratory morbidity in the term and near-term neonate, *Acta Obstet Gynecol Scand* 86 (4) (2007) 389–94.414.