

Amnionic Fluid

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Obstetrics and Gynecology
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Outline

- NORMAL AMNIOTIC FLUID VOLUME
- PHYSIOLOGY
- SONOGRAPHIC ASSESSMENT
- HYDRAMNIOS
- OLIGOHYDRAMNIOS
- BORDERLINE OLIGOHYDRAMNIOS
- TESTS OF FETAL LUNG MATURITY

NORMAL AMNIOTIC FLUID VOLUME

- Amniotic fluid volume increases from approximately 30 mL at 10 weeks to 200 mL by 16 weeks and reaches 800 mL by the mid-third trimester
- The fluid is approximately 98-percent water.
- A full-term fetus contains roughly 2800 mL of water and the placenta another 400 mL, such that the term uterus holds nearly 4 liters of water
- Abnormally decreased fluid volume is termed oligohydramnios, whereas abnormally increased fluid volume is termed hydramnios or polyhydramnios.

Physiology

- Early in pregnancy, the amniotic cavity is filled with fluid that is similar in composition to extracellular fluid.
- 1st half of pregnancy, transfer of water and other small molecules takes place:
 - transmembranous flow: across the amnion
 - intramembranous flow: across the fetal vessels on placental surface
 - transcutaneous flow: across fetal skin.
- Fetal urine production begins between 8 and 11 weeks' gestation
 - After the first trimester, **fetal urine is the major contributor to the amniotic fluid volume.** At the time that fetal urine production occurs, fetal swallowing of the amniotic fluid begins and regulates the increase in fluid from the fetal urine.
 - Water transport across the fetal skin continues until keratinization occurs at 22 to 25 weeks (explains why extremely preterm neonates can experience significant fluid loss across their skin)

Physiology

- 4 pathways play a major role in amniotic fluid volume regulation:
 1. **Fetal urination** is the primary source of amniotic fluid in the second half of pregnancy.
 - By term, fetal urine production may exceed 1 liter per day, and the entire amniotic fluid volume is recirculated on a daily basis.
 2. The hypotonicity of amniotic fluid accounts for significant **intramembranous fluid transfer** across and into fetal vessels on the placental surface.
 - This transfer reaches 400 mL/ day and is a second regulator of fluid volume
 - In the setting of maternal dehydration, the resultant increase in maternal osmolality favors fluid transfer from the fetus to the mother, and then from the amniotic fluid compartment into the fetus

TABLE 11-1. Amniotic Fluid Volume Regulation in Late Pregnancy

Pathway	Effect on Volume	Approximate Daily Volume (mL)
Fetal urination	Production	1000
Fetal swallowing	Resorption	750
Fetal lung fluid secretion	Production	350
Intramembranous flow across fetal vessels on the placental surface	Resorption	400
Transmembranous flow across amniotic membrane	Resorption	Minimal

Data from Magann, 2011; Modena, 2004; Moore, 2010.

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Physiology

3. Through fetal lung fluid secretion (respiratory tract).
 - Approximately 350 mL of lung fluid is produced daily during late gestation, and half of this is immediately swallowed.
4. fetal swallowing is the primary mechanism for amniotic fluid resorption and averages 500 to 1000 mL per day
 - Impaired swallowing, secondary to either a central nervous system abnormality or GI tract obstruction, can result in an impressive degree of hydramnios.
5. MINOR pathways are transmembranous and transcutaneous flow, which together account for a far smaller proportion of fluid transport in the second half of pregnancy.

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Physiology

- Failure of the fetus to begin swallowing results in excessive accumulation of amniotic fluid (***polyhydramnios***) and is an indication of fetal distress, often associated with neural tube disorders.
- Polyhydramnios may be secondarily associated with fetal structural anomalies, cardiac arrhythmias, congenital infections, or chromosomal abnormalities.
- Increased fetal swallowing, urinary tract deformities, and membrane leakage are possible causes of decreased amniotic fluid (***oligohydramnios***).
- Oligohydramnios may be associated with umbilical cord compression, resulting in decelerated heart rate and fetal death.

Measurement

- the average fluid volume is approximately 400 mL between 22 and 30 weeks, doubling thereafter to a mean of 800 mL.
- The volume remain at this level until 40 weeks and then decline by approximately 8 percent per week.

Sonographic Assessment

- Amnionic fluid volume may be measured using either of two semi-quantitative techniques:
 - single deepest pocket of fluid
 - amnionic fluid index (AFI)
- Using either technique, a fluid pocket must be at least 1 cm in width to be considered adequate.
- Fetal parts or loops of umbilical cord may be visible in the pocket, but they are not included in the measurement.
- Color Doppler is generally used to verify that umbilical cord is not within the measurement.

Sonographic Assessment: Single Deepest Pocket (SDP)

- also called the maximum vertical pocket [MVP] or the largest vertical pocket
- This the vertical dimension in centimeters of the largest pocket of amniotic fluid not persistently containing umbilical cord (on gray-scale examination) or fetal extremities and measured at a right angle to the uterine contour.
- The ultrasound transducer is held perpendicular to the floor and parallel to the long axis of the woman. Then, while scanning in the sagittal plane, the largest vertical pocket of fluid is identified and measured.
- normal value: $2 \text{ cm} \leq x < 8 \text{ cm}$
- When evaluating twin pregnancies and other multifetal gestations, a single deepest pocket of amniotic fluid is assessed in each gestational sac, again using a normal range of more than 2 cm to less than 8 cm
- The fetal biophysical profile similarly uses a single deepest vertical pocket threshold of more than 2 cm to indicate normal amniotic fluid volume.

Sonographic Assessment: Amnionic Fluid Index

- the ultrasound transducer is held perpendicular to the floor and parallel to the long axis of the woman.
- The uterus is divided into four equal quadrants—the right and left upper and lower quadrants, respectively.
- The AFI is the sum of the single deepest pocket from each quadrant.
- A useful guideline is that the AFI approximates three times the single deepest pocket of fluid
- The AFI is generally considered normal if greater than 5 cm and below 24 or 25 cm. Values outside these ranges indicate oligohydramnios and hydramnios, respectively.
 - The upper threshold of 24 cm is used in consensus documents (American College of Obstetricians and Gynecologists, 2016

Criteria for mild, moderate, and severe polyhydramnios

	Mild	Moderate	Severe
Single deepest pocket	8.0 to 11.9 cm	12.0 to 15.9 cm	≥16.0 cm
Amniotic fluid index	24.0 to 29.9 cm	30.0 to 34.9 cm	>35.0 cm

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Single deepest vertical pocket or amniotic fluid index as evaluation test for predicting adverse pregnancy outcome (SAFE trial): a multicenter, open-label, randomized controlled trial

Conclusion: Use of the AFI method increased the rate of diagnosis of oligohydramnios and labor induction for oligohydramnios without improving perinatal outcome. The SDP method is therefore the favorable method to estimate amniotic fluid volume, especially in a population with many low-risk pregnancies.

single deepest pocket

HYDRAMNIOS

- This is an abnormally increased amniotic fluid volume, and it complicates 1 to 2 percent of singleton pregnancies
- more frequently noted in multifetal gestations
- Hydramnios may be suspected if the uterine size exceeds that expected for gestational age.
- The uterus may feel tense, and palpating fetal small parts or auscultating fetal heart tones may be difficult.

HYDRAMNIOS

- Hydramnios may be further categorized according to degree.
 - mild if the AFI is 25 to 29.9 cm;
 - moderate, if 30 to 34.9 cm;
 - severe, if 35 cm or more
- Using the single deepest pocket of amniotic fluid:
 - mild hydramnios is defined as 8 to 9.9 cm
 - moderate as 10 to 11.9 cm
 - severe hydramnios as 12 cm or more
- In general, severe hydramnios is far more likely to have an underlying etiology and to have consequences for the pregnancy than mild hydramnios, which is frequently idiopathic and benign.

HYDRAMNIOS: Etiology

- Underlying causes of hydramnios include:
 - fetal anomalies (structural abnormalities/genetic syndromes): 15%
 - Diabetes: 15-20%
 - Congenital infection, red blood cell alloimmunization, and placental chorioangioma are less frequent etiologies
 - Infections: cytomegalovirus, toxoplasmosis, syphilis, and parvovirus.
 - hydrops fetalis, severe fetal anemia

HYDRAMNIOS: Congenital Anomalies

- Because of this association, targeted sonography is indicated whenever hydramnios is identified.
 - If a fetal abnormality is encountered concurrent with hydramnios, amniocentesis with chromosomal microarray analysis should be offered, because the aneuploidy risk is significantly elevated
- the degree of hydramnios correlates with the likelihood of an anomalous infant
- The anomaly risk is particularly high with hydramnios coexistent with fetal-growth restriction

HYDRAMNIOS: Selected anomalies and mechanism for hydramnios

Mechanism	Anomaly Examples
Impaired swallowing (CNS)	Anencephaly Hydranencephaly Holoprosencephaly
Impaired swallowing (craniofacial)	Cleft lip/palate Micrognathia
Tracheal compression or obstruction	Neck venolymphatic abnormality CHAOS ^a
Thoracic etiology (mediastinal shift)	Diaphragmatic hernia ^a Cystic adenomatoid malformation ^a Pulmonary sequestration ^a
High-output cardiac state	Ebstein anomaly ^a Tetralogy of Fallot with absent pulmonary valve ^a Thyrotoxicosis ^a
Functional cardiac etiology	Cardiomyopathy, myocarditis ^a
Cardiac arrhythmia	Tachyarrhythmia ^a : atrial flutter, atrial fibrillation, supraventricular tachycardia Bradyarrhythmia ^a : heart block
GI obstruction	Esophageal atresia Duodenal atresia
Renal-Urinary	Ureteropelvic junction obstruction ("paradoxical hydramnios") Baastrup syndrome
Neurological or muscular etiology	Arthrogryposis, akinesia sequence Myotonic dystrophy
Neoplastic etiology	Sacrococcygeal teratoma ^a Mesoblastic nephroma ^a Placental chorioangioma ^a

HYDRAMNIOS: Diabetes Mellitus

- The amniotic fluid glucose concentration is higher in diabetic women than in those without diabetes, and the AFI may correlate with the amniotic fluid glucose concentration
 - Such findings support the hypothesis that maternal hyperglycemia causes fetal hyperglycemia with resulting fetal osmotic diuresis into the amniotic fluid compartment.
 - rescreening for gestational diabetes in pregnancies with hydramnios does not appear to be beneficial, provided that the second-trimester glucose tolerance test result is normal

HYDRAMNIOS: Multifetal Gestation

- Hydramnios is generally defined in multifetal gestations as a single deepest amnionic fluid pocket measuring 8 cm or more.
- It may be further characterized as moderate if the single deepest pocket is at least 10 cm and severe if this pocket is at least 12 cm.
- severe hydramnios is more strongly associated with fetal abnormalities.
- In monochorionic gestations, hydramnios of one sac and oligohydramnios of the other are diagnostic criteria for twin-twin transfusion syndrome (TTTS)
- In the absence of TTTS, hydramnios does not generally raise pregnancy risks in non-anomalous twins

HYDRAMNIOS: Idiopathic Hydranmios

- This accounts for up to 70% of cases of hydranmios.
- Idiopathic hydranmios is rarely identified during midtrimester sonography and is often an incidental finding later in gestation.
- The gestational age at sonographic detection usually lies between 32 and 35 weeks
- Although it is a diagnosis of exclusion, an underlying fetal abnormality may subsequently become apparent with advancing gestation, particularly if the degree of hydranmios becomes severe.
- Mild, idiopathic hydranmios is most commonly a benign finding, and associated pregnancy outcomes are usually good.

HYDRAMNIOS: Complications

- Unless hydramnios is severe or develops rapidly, maternal symptoms are infrequent.
- With chronic hydramnios, fluid accumulates gradually, and a woman may tolerate excessive abdominal distention with relatively little discomfort.
- Acute hydramnios, however, tends to develop earlier in pregnancy.
 - It may result in preterm labor before 28 weeks or in symptoms that become so debilitating as to necessitate intervention.

HYDRAMNIOS: Complications

- Symptoms may arise from pressure exerted within the overdistended uterus and upon adjacent organs.
- When distention is excessive, the mother may suffer dyspnea and orthopnea.
- Edema may develop as a consequence of major venous system compression by the enlarged uterus, and it tends to be most pronounced in the lower extremities, vulva, and abdominal wall.
- Rarely, oliguria may result from ureteral obstruction by the enlarged uterus
- Maternal complications associated with hydramnios include placental abruption, uterine dysfunction during labor, and postpartum hemorrhage.
 - Placental abruption may result from the rapid decompression of an overdistended uterus that follows fetal-membrane rupture or therapeutic amnioreduction.
 - With prematurely ruptured membranes, a placental abruption occasionally occurs days or weeks after amniorrhexis.
 - Uterine dysfunction consequent to overdistention may lead to postpartum atony and, in turn, postpartum hemorrhage.

HYDRAMNIOS: Pregnancy Outcomes

- Some outcomes more common with hydramnios include birthweight >4000 g, cesarean delivery, and perinatal mortality.
- Risks appear to be compounded when a growth-restricted fetus is identified with hydramnios
 - The combination also has a recognized association with trisomy 18.
 - When an underlying cause is identified, degree of hydramnios has been associated with likelihood of preterm delivery, small-for-gestational age newborn, and perinatal mortality.

HYDRAMNIOS: Management

- treatment is directed to the underlying cause.
- If severe hydramnios result in early preterm labor or the development of maternal respiratory compromise: amnioreduction may be needed.
 - Approximately 1000 to 2000 mL of fluid is slowly withdrawn over 20 to 30 minutes, depending on the severity of hydramnios and gestational age.
 - The goal is to restore amniotic fluid volume to the upper normal range.
 - Hydramnios severe enough to necessitate amnioreduction almost invariably has an underlying cause, and subsequent amnioreduction procedures may be required as often as weekly or even semiweekly.

OLIGOHYDRAMNIOS

- This is an abnormally decreased amount of amniotic fluid.
- When no measurable pocket of amniotic fluid is identified, the term anhydramnios may be used.
- Unlike hydramnios, which is often mild and often confers a benign prognosis in the absence of an underlying etiology, oligohydramnios is always a cause for concern.
- The sonographic diagnosis of oligohydramnios is usually based on an AFI less than 5 cm or a single deepest pocket of amniotic fluid below 2 cm.
- use of AFI rather than single deepest pocket will identify more pregnancies as having oligohydramnios, without evidence of improvement in pregnancy outcomes
- When evaluating multifetal pregnancies for TTTS, a single deepest pocket below 2 cm is used to define oligohydramnios

OLIGOHYDRAMNIOS: etiology

- Early-Onset Oligohydramnios
 - When amnionic fluid volume is abnormally decreased from the early second trimester, it may reflect a fetal abnormality that precludes normal urination, or it may represent a placental abnormality sufficiently severe to impair perfusion.
 - In either circumstance, the prognosis is poor.
 - Ruptured membranes should be excluded, and targeted sonography is performed to assess for fetal and placental abnormalities.

OLIGOHYDRAMNIOS: etiology

- Oligohydramnios after Midpregnancy
 - When amniotic fluid volume becomes abnormally decreased in the late second or in the third trimester, it is very often associated with fetal-growth restriction, with a placental abnormality, or with a maternal complication such as preeclampsia or vascular disease
 - The underlying cause in such cases is frequently uteroplacental insufficiency, which can impair fetal growth and reduce fetal urine output.
 - Exposure to selected medications has also been linked with oligohydramnios
 - Investigation of third-trimester oligohydramnios generally includes evaluation for ruptured membranes and sonography to assess fetal growth.
 - Umbilical artery Doppler studies are recommended if growth restriction is identified

OLIGOHYDRAMNIOS: etiology

Congenital Anomalies

- By approximately 18 weeks, the fetal kidneys are the main contributor to amniotic fluid volume.
- Selected renal abnormalities that lead to absent fetal urine production include:
 - bilateral renal agenesis
 - bilateral multicystic dysplastic kidney
 - unilateral renal agenesis with contralateral multicystic dysplastic kidney
 - infantile form of autosomal recessive polycystic kidney disease.
- Urinary abnormalities may also result in oligohydramnios because of fetal bladder outlet obstruction. Examples of this are:
 - posterior urethral valves
 - urethral atresia or stenosis
 - megacystis microcolon intestinal hypoperistalsis syndrome.
- Complex fetal genitourinary abnormalities such as persistent cloaca and sirenomelia similarly may result in a lack of amniotic fluid

OLIGOHYDRAMNIOS: etiology

Medication

- Oligohydramnios has been associated with exposure to drugs that block the renin- angiotensin system. These include:
 - angiotensin-converting enzyme (ACE) inhibitors
 - angiotensin-receptor blockers
 - nonsteroidal antiinflammatory drugs (NSAIDs)
- When taken in the second or third trimester, ACE inhibitors and angiotensin-receptor blockers may create fetal hypotension, renal hypoperfusion, and renal ischemia, with subsequent anuric renal failure
- NSAIDs can be associated with fetal ductus arteriosus constriction and with lower fetal urine production.
 - In neonates, their use may result in acute and chronic renal insufficiency

OLIGOHYDRAMNIOS: Pregnancy Outcomes

- Oligohydramnios is associated with adverse pregnancy outcomes.
- AFI ≤ 5 cm identified between 24 and 34 weeks was associated with increased risks for stillbirth, spontaneous or medically indicated preterm birth, heart rate pattern abnormalities, and growth restriction
- women with oligohydramnios had a twofold greater risk for cesarean delivery for fetal distress and a fivefold higher risk for an Apgar score <7 at 5 minutes compared with pregnancies with a normal AFI

OLIGOHYDRAMNIOS:Pulmonary Hypoplasia

- When diminished amniotic fluid is first identified before the mid-second trimester, particularly before 20 to 22 weeks, pulmonary hypoplasia is a significant concern.
- Severe oligohydramnios secondary to a renal abnormality generally has a lethal prognosis.
- If a placental hematoma or chronic abruption is severe enough to result in oligohydramnios—the chronic abruption-oligohydramnios sequence (CAOS)— it commonly also causes growth restriction
- The prognosis for this condition is poor.

OLIGOHYDRAMNIOS:Management

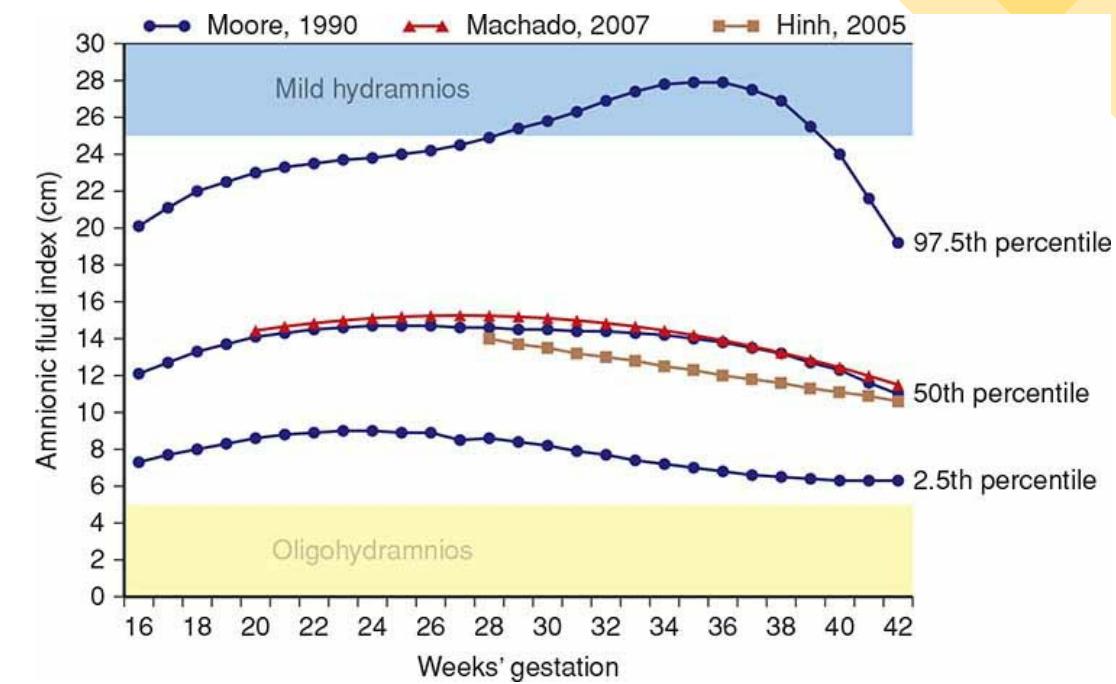
- An evaluation for fetal anomalies and growth is essential.
- In a pregnancy complicated by oligohydramnios and fetal-growth restriction, close fetal surveillance is important because of associated morbidity and mortality
- Oligohydramnios detected before 36 weeks gestation in the presence of normal fetal anatomy and growth is generally managed expectantly in conjunction with enhanced fetal surveillance.

OLIGOHYDRAMNIOS:Management

- Antepartum management of oligohydramnios may include maternal hydration.
 - oral or intravenous hydration was associated with significant improvement in the AFI. However, it was not clear whether this translated into better pregnancy outcomes
- Amnioinfusion may be used intrapartum to help resolve variable fetal heart rate decelerations.
 - It is not considered treatment for oligohydramnios per se, although the decelerations are presumed secondary to umbilical cord compression resulting from lack of amniotic fluid.
 - Amnioinfusion is not the standard of care for other etiologies of oligohydramnios and is not generally recommended.

“Borderline” OLIGOHYDRAMNIOS

- usually refers to an AFI between 5 and 8 cm
- Through the mid-third trimester, an AFI value of 8 cm is below the 5th percentile on the Moore nomogram
- Associated with higher rates of preterm delivery, cesarean delivery for a nonreassuring fetal heart rate pattern, and fetal- growth restriction
- Studies have concluded that evidence is insufficient to support fetal testing or delivery in this setting.



Tests for fetal lung maturity

Lecithin-Sphingomyelin (L/S) Ratio

- The reference method to which tests of FLM are compared is the *lecithin-sphingomyelin (L/S) ratio*.
- Lecithin is the primary component of the **surfactants** (phospholipids, neutral lipids, and proteins) that make up the alveolar lining and account for alveolar stability.
- Lecithin is produced at a relatively low and constant rate until the 35th week of gestation, and thereafter increases in its production rate occurs, resulting in the stabilization of the fetal lung alveoli.
- Sphingomyelin is a lipid that is produced at a constant rate after about 26 weeks' gestation; therefore, it can serve as a control on which to base the rise in lecithin.

Lecithin-Sphingomyelin (L/S) Ratio

- Both lecithin and sphingomyelin appear in the amniotic fluid in amounts proportional to their concentrations in the fetus.
- Prior to 35 weeks' gestation, the L/S ratio is usually less than 1.6 because large amounts of lecithin are not being produced at this time.
- It will rise to 2.0 or higher when lecithin production increases to prevent alveolar collapse.
- Therefore, when the L/S ratio reaches 2.0, a preterm delivery is usually considered to be a relatively safe procedure.
- Falsely elevated results are encountered in fluid contaminated with blood or meconium because both these substances contain lecithin and sphingomyelin.

Lecithin-Sphingomyelin (L/S) Ratio

- Quantitative measurement of lecithin and sphingomyelin is performed using thin-layer chromatography.
 - The procedure is labor intensive and subject to high coefficients of variation.
- Many laboratories have replaced the L/S ratio with the more cost-effective *phosphatidylglycerol* immunoassays, fluorescence polarization, and *lamellar body* density procedures.

Amniostat-FLM

- The presence of another lung surface lipid, phosphatidyl glycerol, is also essential for adequate lung maturity.
- The production of phosphatidyl glycerol normally parallels that of lecithin, but its production is delayed in cases of maternal diabetes.
- In cases of DM, respiratory distress may occur in the presence of an L/S ratio of 2.0. Therefore, a thin-layer chromatography lung profile must include lecithin, sphingomyelin, and phosphatidyl glycerol to provide an accurate measurement.

Amniostat-FLM

- Development of an immunologic agglutination test for phosphatidyl glycerol has provided a more rapid method for assessment of fetal maturity that does not require a laboratory to be equipped to perform thin-layer chromatography.
- The Aminostat-FLM (Irving Scientific, Santa Ana, Calif.) uses anti- sera specific for phosphatidyl glycerol and is not affected by specimen contamination with blood and meconium.
- Studies have shown good correlation with thin-layer chromatography but with a slightly higher incidence of false-negative results that may need to be followed up with further testing.

Foam Stability

- Until the development of biochemical techniques to measure the individual lung-surface lipid concentrations, a mechanical screening test, called the “foam” or “shake” test, was used to determine their presence.
- **FOAM/SHAKE TEST:**
 - Amniotic fluid is mixed with 95% ethanol, shaken for 15 seconds, and allowed to sit undisturbed for 15 minutes.
 - At the end of this time, the surface of the fluid is observed for the presence of a continuous line of bubbles around the outside edge.
 - The presence of bubbles indicates that a sufficient amount of phospholipid is available to reduce the surface tension of the fluid even in the presence of alcohol, an antifoaming agent.

Foam Stability

- A modification of the foam test uses 0.5 mL of amniotic fluid added to increasing amounts of 95% ethanol, providing a gradient of ethanol/fluid ratios ranging from 0.42 mL to 0.55 mL in 0.01-mL increments, which can be used to provide a semiquantitative measure of the amount of surfactant present.
- A value of 47 or higher indicates FLM.
- The Foam Stability Index has shown good correlation with the L/S ratio and tests for phosphatidyl glycerol.
- The test cannot be used with contaminated amniotic fluid because blood and meconium also reduce surface tension.

PROCEDURE

Procedure for Foam Stability Index

1. Add 0.5 mL of amniotic fluid to tubes containing increasing amounts of 95% ethanol ranging from 0.42 to 0.55 mL in 0.01-mL increments.
2. Vigorously shake for 15 seconds.
3. Allow to sit undisturbed for 15 minutes.
4. Observe for the presence of a continuous line of bubbles around the outside edge.
5. Values ≥ 47 indicate fetal lung maturity.

Microviscosity: Fluorescence Polarization Assay

- The presence of phospholipids decreases the microviscosity of the amniotic fluid.
 - This change in microviscosity can be measured using the principle of fluorescence polarization employed by the Abbott TDx analyzer with the TDx/TDxFLx FLM II Assay System (Abbott Laboratories, Abbott Park, Ill.).
- The TDx/TDxFLx Fetal Lung Maturity II (FLMII) assay is a reagent system for the quantitative measurement of the ratio of surfactant to albumin in amniotic fluid for assessment of lung maturity of the fetus.
- This assay measures the polarization of a fluorescent dye that combines with both surfactants and albumin.
- Dye bound to surfactant has a longer fluorescence lifetime and exhibits low polarization, whereas dye bound to albumin has a decreased fluorescence lifetime and has high polarization.
- Albumin is used as an internal standard in the same manner as sphingomyelin because it remains at a constant level throughout gestation

Microviscosity: Fluorescence Polarization Assay

- The recorded changes in polarization produce a surfactant/albumin ratio expressed in milligrams surfactant to grams albumin that is compared with a fetal lung maturity II assay calibration standard curve that includes phosphatidyl glycerol and ranges from 0 to 160 mg/g.
- Fetal Lung Maturity II Calibrators of known surfactant/albumin ratio are run and the resulting standard curve is stored in memory.
- Sample results are calculated from the stored standard curve using polarization values generated for each sample.
- A value of 55 mg surfactant per gram albumin or greater provides a conservative indicator of FLM and lower values may be considered.
- Immature results with the FLM II assay are less than or equal to 39 mg/g.
- Results between 40 mg/g and 54 mg/g cannot be declared “mature” or “immature” and must be evaluated with caution.

Microviscosity: Fluorescence Polarization Assay

- The TDx–FLM II test value correlates well with a L/S ratio of 2.0 and has few falsely mature results, making it an excellent screening tool.
- Sequential L/S testing is recommended when the results of the TDx–FLM II suggest fetal lung immaturity.
- An accurate gestational age is an important consideration in interpreting the results.
- The test requires at least 1.0 mL of amniotic fluid.
- Fluid should be filtered rather than centrifuged prior to examination to prevent sedimentation of the lipids and reporting a falsely decreased result.
- Specimens contaminated with blood, meconium, suspected maternal urine, and visibly icteric samples should not be used.

Lamellar Bodies and Optical Density

- Lamellar bodies are lamellated phospholipids that represent a storage form of surfactant.
- The surfactants responsible for FLM are produced and secreted by the type II pneumocytes of the fetal lung and stored in the form of structures termed lamellar bodies at about 20 weeks of gestation.
- The lamellar bodies enter the alveolar spaces to provide surfactant and also enter the amniotic fluid at about 26 weeks of gestation.
- As the fetal lung matures, increased lamellar body production is reflected by an increase in amniotic fluid phospholipids and the L /S ratio.
- Therefore, the number of lamellar bodies present in the amniotic fluid correlates with the amount of phospholipid present in the fetal lungs.

Lamellar Bodies and Optical Density

- The presence of lamellar bodies increases the optical density (OD) of the amniotic fluid.
- Specimens are centrifuged at 2000 g for 10 minutes and examined using a wavelength of 650 nm, which rules out interference from hemoglobin but not other contaminants, such as meconium.
- An OD of 0.150 has been shown to correlate well with an L/S ratio of greater than or equal to 2.0 and the presence of phosphatidyl glycerol.
- Lamellar body diameter is similar to that of small platelets → therefore, lamellar body counts (LBCs) can be obtained rapidly with use of the platelet channel of hematology analyzers.

Lamellar Bodies and Optical Density

- Lamellar bodies can be counted using resistance-pulse counting, such as that employed by Coulter cell-counting instruments (Beckman Coulter, Inc., Fullerton, Calif.).
- Lamellar bodies can be counted using the platelet channel.
- Samples must be free of particle contamination such as meconium and blood.
- A count of 32,000 or more particles per microliter represents adequate FLM.

Lamellar Bodies and Optical Density

- The ADVIA 120 hematology system (Siemens Medical Solutions Diagnostics, Diagnostics Division, Tarrytown, N.Y.) measures two light-scatter angles of particles as they pass through a laser beam and identifies particles based on their cell volume indices and refractive index.
- The LBC count is the sum of all platelet-sized particles measured in the platelet channel (calculated LBC).
- A count of 35,400 or more particles per microliter indicates FLM.
- Sysmex XE-2100 (Sysmex, Mundelein, Ill.) technology simultaneously detects direct current and radiofrequency impedance thought to reflect intracellular changes.
- Cell-dyn 3500 (Abbott Laboratories, Abbott Park, Ill.) combines optical scatter and impedance.

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