Title: Amniotic Fluid Volume: Rapid MR based assessment at 28-32 weeks gestation

Keywords:

Magnetic Resonance Imaging, Ultrasonography, Comparative Study, Pregnancy, Amniotic Fluid

Key points:

- MR projection hydrography can be used to estimate amniotic fluid volume.
- This technique relies on the T2w signal from amniotic fluid.
- Amniotic fluid volume is more accurately assessed than with ultrasound
- More accurate depiction in high risk pregnancies could influence clinical management

Abbreviations and acronyms:

PH – Projection hydrography

AFV – Amniotic fluid volume

SDVP – Single deepest vertical pocket

AFI – Amniotic fluid index

MSP – Multi-section planimetry

SSFSE – Single-shot fast spin echo

bSSFP – Balanced steady-state free precession

Abstract

Objectives: This work evaluates rapid magnetic resonance projection hydrography (PH) based amniotic fluid volume (AFV) estimates against established routine ultrasound single deepest vertical pocket (SDVP) and amniotic fluid index (AFI) measurements, in utero, at 28-32 weeks gestation. Manual multi-section planimetry (MSP) based measurement of AFV is used as a proxy reference standard.

Methods: 35 women with a healthy singleton pregnancy (20-41 years) attending routine antenatal ultrasound were recruited. SDVP and AFI were measured using ultrasound, with same day MRI assessing AFV with PH and MSP. The relationships between the respective techniques were assessed using linear regression analysis and Bland-Altman method comparison statistics.

Results: When comparing estimated AFV, a highly significant relationship was observed between PH and the reference standard MSP (R^2=0.802, p<0.001). For the US measurements, SDVP measurement related most closely to amniotic fluid volume, (R^2=0.470, p<0.001), with AFI demonstrating a weaker relationship (R^2=0.208, p=0.007).

Conclusion: This study shows that rapid MRI based PH measurement is a better predictor of AFV, relating more closely to our proxy standard than established US techniques. Although larger validation studies across a range of gestational ages are required this approach could form part of MR fetal assessment, particularly where poly or oligohydramnios is suspected.

Introduction

Amniotic fluid volume (AFV) is subjectively assessed during routine ultrasound scans in the second and third trimesters and may be measured in more specific circumstances where there is a concern over fetal condition. Poly and oligohydramnios are associated with a wide range of disorders reflecting fetal compromise or developmental abnormalities, such as; diabetes, neuromuscular conditions, fetal anaemia and upper GI abnormality in the case of polyhydramnios, or renal abnormalities and fetal growth restriction in the case of oligohydramnios. As such, abnormal AFV quantification is widely used as an indicator that further materno-fetal assessment is required[1,2].

Current established methods of assessing AFV use real-time 2D ultrasound. An initial qualitative assessment is used in the majority of pregnancies, and two semi-quantitative techniques; amniotic fluid index (AFI) and single deepest vertical pocket (SDVP), in the case of high risk pregnancies or where qualitative assessment is abnormal. These semi-quantitative surrogate assessments are relatively easily obtained linear measurements which correlate with, but do not measure true AFV. Research over several decades has provided some validation of these measurements in respect of fetal development and outcomes, though recent evidence suggests SDVP is a more accurate predictor of clinical outcome than AFI[3]. Dye dilution methods have been used previously to obtain more accurate estimates of AFV[16] but these are invasive and difficult to implement in clinical practice or to justify for research purposes.

There has been controversy regarding the accuracy of these US based estimates of AFV in clinical practice, as over- or under-estimation of fluid volume may lead to inappropriate

management decisions[2,3,4]. As a result, the role of AFV measurements and their contribution to antenatal care varies widely world-wide.

MRI is increasingly used for visualising the fetus in utero. Although primarily used for neurological assessment[5] it has been shown to be useful in the management of diaphragmatic[6] and renal anomalies[7]. As fetal MRI is performed more regularly and becomes more widely available, there is also the potential to provide the standard biometric data normally obtained with ultrasound, including AFV estimation.

Only one study has made a direct comparison between MRI measurement of AFV, AFI and SDVP, but this was limited to 80 term fetuses[8], and found that MRI provided good correlation with ultrasound measurements. A single early study attempted 0.5T MRI measurement of amniotic fluid volume in 34 complicated pregnancies from the 24th to 42nd weeks[9]; this group demonstrated a correlation with fetal birth weight, but did not make any comparison with normal ultrasound measures of fluid volume. No study we are aware of has investigated whether MRI estimates of amniotic fluid volume correlate with AFI and SDVP measurements in the early third trimester.

The use of multi-section planimetric (MSP) volume analysis of the feto-placental unit for the calculation of organ volume[10] and fetal weight[11,12] has already been demonstrated using MRI, and found to be more accurate than ultrasound[12]. This approach was adopted in the MR based measurement of amniotic fluid volume by Zaretsky et al[8]. However these techniques, typically using the sum of manually or semi-automatically defined regions of interest (ROIs) across multiple image sections through the uterus, are typically too time consuming for routine practice.

Rapid MR hydrographic projection techniques for estimating fluid volume have been described for estimating fluid volumes of the adult stomach[13], bladder[14] and pancreatic

secretions[15]. The hydrographic projection technique relies on a single heavily T2w thick-section acquisition, and that the majority of bodily fluids possess a long T2 relaxation time similar to water. In the resulting images the signal is proportional to the volume of fluid present. Comparison between the signal from a reference fluid volume and the volume of interest allows an estimate of fluid volume to be calculated. As the relevant ROIs have only to encompass the whole of the reference and interest volume on a single section this is much less time consuming than a manual multi-section planimetric approach.

This work evaluates, in a population of uniparous women at 28-32 weeks gestational age, rapid MR projection hydrography (PH) based AFV estimates against established routine ultrasound SDVP and AFI measurements. Manual MSP based measurement of AFV is used as a proxy reference standard.

Methods

Study design:

35 women with a singleton pregnancy (age: 20-41 yrs) were recruited from the population attending for routine antenatal ultrasound with a normal healthy pregnancy. Ethical approval was obtained from the local Research Ethics Committee and participants provided written informed consent. Participants with contra-indications for MRI (eg cardiac pacemakers, intra-cranial aneurysm clips) were excluded. The range of gestational age at the time of the examination was 28 to 32 weeks. In all cases ultrasound was performed immediately prior to MRI.

US:

A single experienced sonographer performed all ultrasound examinations with a 7 MHz curvilinear probe using a commercial ultrasound system (E6 Voluson, GEHC, Wisconsin,

USA). Participants were positioned supine. SDVP was recorded as the deepest fluid depth in a single pocket of fluid around the fetus. AFI was calculated as the sum of the deepest fluid depth in the four quadrants of the uterus measured vertically.

MR:

MR examinations were performed using a 1.5 T MRI system (MR 450, GEHC, Wisconsin, USA), using the integrated body coil, an 8 channel cardiac receive coil and left decubitus positioning. Participants were asked to empty their urinary bladder immediately prior to the examination. A 50 ml bag of normal 0.9% saline for infusion (Macopharma, Twickenham, UK) was included as a reference volume in the field of view anterior to the abdominal wall. Breath-hold PH was performed using the integral body coil in the sagittal plane with 20cm thick-slab single-shot fast spin echo (SSFSE) sequences (TE 800msec, TR 10seconds, matrix 384x256, FOV 32cm). This plane was prescribed sagittally relative to the uterus after performing additional axial acquisitions for localisation using balanced steady-state free precession (bSSFP). Five separate breath-hold acquisitions were obtained to reduce the risk of fetal motion causing unwanted signal loss through motion related spin dephasing. Between these acquisitions, a pause of at least 20 seconds was included to ensure full recovery of the longitudinal magnetisation. In addition breath-hold 5mm thick multisection 2D bSSFP sequences for MSP were obtained in the axial and sagittal planes, with no inter slice gap (TE, 1.4msec TR 4.3msec, matrix 384x256, FOV 36cm), through the whole uterus. These acquisitions used the cardiac coil rather than the integral body coil.

Data analysis:

Two independent observers (blinded to US results) analysed the images on a workstation (iMac, Cupertino, California, USA) using OsiriX (version 5.5.2, Pixmeo, SARL).

MSP was performed on all the sections of the axial and sagittal bSSFP imaging where amniotic fluid was present. The fetus, umbilical cord and placenta were excluded from each ROI. The volume results were summed for the relevant sections in each plane to provide an AFV measurement. On the PH images separate ROIs were drawn around the entire amniotic sac and the reference saline volume. Examples of MP and PH are provided in **Figure 1**. Through extrapolation from the 50ml reference volume the AFV was estimated. This was calculated from the following formula:

AFV = [(mean uterus signal x uterus area) / (mean reference signal x reference area)] x 50

The maximum AFV estimate from the 5 acquisitions was used as the final AFV, as this was likely to represent the acquisition with the least bulk motion related dephasing effect.

Statistical Analysis:

Normality assumptions were investigated using the formal Shapiro-Wilks-W test, and the resulting distributions were summarised accordingly. Least squares linear regression analysis was used to investigate the relationship between the MRI derived amniotic fluid metrics MSP and PH as well as the ultrasound indices (SDVP and AFI). The strength of the relationships were summarised as R^2. P-values <0.05 were deemed as statistically significant. In addition, Bland-Altman summary statistics were calculated to investigate the relationship between the reference standard MSP and PH. Inter-rater agreement was calculated using the intra-class correlation statistic. All statistical analyses were performed in R (version 3.1.1, The R Foundation for Statistical Computing, Vienna, Austria).

Results

35 participants were enrolled and underwent same day ultrasound and MRI. There was one failure of MR projection hydrography acquisition, due to operator error. The MRI

examination was well tolerated and all the participants completed the examination. 34 participants (gestational range 28-32 weeks, mean 30 weeks) had complete datasets for analysis.

MR calculation of AFV by planimetry and projection hydrography, with distributional statistics for amniotic fluid volumes, are summarised in **Table 1**. The relationship between the reference standard MSP and the rapid MRI PH technique is shown in **Figure 2** and **Table 2**, along with a comparison with conventional ultrasound metrics. A highly significant relationship was observed between PH and MSP ($p<0.001$), the R^2 measurement demonstrates that PH accounts for ~80% of the variability of the MSP measurement. A trend was noted for the PH technique to overestimate the amniotic fluid values at the lower end of the distribution, and conversely, underestimate the amniotic fluid values at the upper end of the distribution (observed slope = 1.45 and intercept = -190.82). This observation is also evident in the general upward trend observed in the Bland-Altman plot **Figure 3**. The Bland-Altman summary statistics comparing manual planimetry and projection hydrography were; bias -9.3mL and 95% limits of agreement 151.9 to -170.5mL.

A Bland-Altman plot comparing both axial and sagittal AFV calculation using MSP is shown in **Figure 4**. This shows only a small systematic difference between the axial and sagittal measurements (bias 20.5ml), with good agreement between the two planes (95% limits of agreement 35.8 to -76.8 ml).

A weak but significant relationship was demonstrated between MSP and the ultrasound derived AFI. The AFI accounts for only 20.8% of the variation in MSP ($R^2=0.208$, $p=0.007$). The SDVP measurement correlated more strongly with MSP, accounting for 47% of the variation in the measurement ($R^2=0.407$, $p<0.001$).

Inter-observer agreement is reported for the MRI derived amniotic fluid metrics in **Table 1**. Inter-rater agreement was excellent for all MRI techniques, but highest in the PH technique (ICC=0.997).

Discussion

The demonstration of an abnormal AFV during pregnancy usually has a diagnostic impact, although the clinical management can vary, as the accuracy of US assessment of AFV is controversial[2,3]. Most commonly, detection of oligohydramnios in the third trimester will prompt detailed ultrasound of the fetus and may lead to elective induction of labour at term, though oligohydramnios as currently assessed may be a poor predictor of fetal outcome[4]. Current guidance favours SDVP as the more reliable measurement for monitoring and management decisions[3], and our study also found that SDVP is the US technique that most accurately reflects AFV (as measured using MSP). Ultrasound measurements can also be influenced by operator technique, maternal position during the ultrasound examination and fetal position, and provide only an indirect assessment of fluid volume. This study demonstrates the utility of a rapid MR based technique (PH) for measurement of AFV, which has the potential to estimate AFV more accurately than current US methods.

Our results show that MR based PH is highly predictive of amniotic fluid volume, as determined using the sagittal MSP method. A strength of the PH technique is that it requires much less post hoc analysis than MSP in providing an AFV (approximately 5 minutes for PH versus 60-90 minutes for MSP), reflecting the many fewer ROIs that need to be drawn. An advantage of PH over ultrasound is that it provides a less subjective volume estimate, in comparison to the semi-quantitative ultrasound AFV assessment techniques.

There was good reproducibility of the results between the two observers. The slightly lower ICC noted for MSP (~0.96) likely reflects the subjectivity in demarcating amniotic fluid

regions in utero on multiple MRI sections. PH is available on most clinical MR systems as a similar sequence is widely used for MR cholangiopancreatography. A 50mls bag of normal saline as a reference volume within the field of view is the only additional material required. The use of an internal reference is likely to make the technique easier to reproduce with different MR systems, although further work is required to demonstrate this. These are important factors for a measurement technique that could be widely used in clinical practice.

We also incidentally demonstrated that SDVP is a more accurate predictor of AFV than AFI, which is in keeping with published studies correlating ultrasound based AFV measurements with outcomes[3]. Neither SDVP nor AFI is a true fluid volume measurement, limiting direct comparison with MR, but when comparing with MSP we considered the use of correlations acceptable.

Zaretsky el al[8] examined AFV in term pregnancies, comparing MSP MR AFV measurement, SDVP and AFI with the volume of amniotic fluid extracted at caesarean section. They specifically focused on the predictability of oligohydramnios using each method, and demonstrated the three techniques were comparable, though MR tended to overestimate AFV when compared with the volume found at section. A weakness of their study was that they did not exclude the umbilical cord from their ROIs, possibly contributing to the overestimation. In our study we did not include the cord in our MSP measurements, however this approach was time consuming (see **Figure 5**).

There are some limitations of the PH technique. Examination of the Bland-Altman plots (**Figure 3**) indicates there is a systematic bias that varies with fluid volume levels. At lower AFV there is a tendency for PH to overestimate AFV, whereas at higher AFVs PH tends to underestimate AFV. We chose to specifically use sagittal slices for the PH calculation, to allow exclusion of the maternal bladder and to ensure the whole uterus was encompassed in

the slice thickness. Integral to the PH technique is summation of the entire T2w fluid signal from the amniotic sac. This includes signal from the fetal bladder, gastrointestinal tract and cerebrospinal fluid. Intuitively these will give a greater contribution to the volume estimates at lower AFV levels, which may explain the overestimation bias. This could be exacerbated by conditions that increased the fluid volumes of these other fetal structures, for example hydrocephalus.

We speculate that possible reasons for the underestimation of AFV at higher fluid volume levels include greater fetal motion induced spin dephasing allowed by the larger space within the amniotic sac, and/or a reduction of fluid signal related to spatial variation of excitation flip angle – a recognised problem using a projection approach exacerbated by the increasing size of the subject being imaged, i.e. a larger amniotic fluid sac. Despite similar limitations, when a projection hydrographic method for measuring adult bladder urine volume was compared with true post-micturition urine volume there was no significant difference[14].

MR imaging of the fetus is often complicated by undesired fetal motion, with rapid sequences favoured for this reason. Although each PH acquisition takes approximately one second, fluid motion during the data readout period may lead to spin dephasing and signal loss. We intentionally repeated the sequence five times, and chose the largest AFV estimate obtained in order to obtain signal during the period of least fetal motion. Examples of motion induced spin dephasing are shown in **Figure 6**. Clearly if a fetus moved during every PH acquisition this would lead to an inevitable underestimate of AFV.

Another acknowledged limitation of our study is that a true AFV was not determined. However, our range of amniotic fluid volumes compares well to those taken by invasive methods, such as dye-dilution[16]. We also found good agreement between the volume established in both axial and sagittal MSP. As described above additional fluid volume

contributions from fetal organs were inevitably incorporated and RF excitation variation may have reduced signal values in larger amniotic fluid volumes. Additionally the T2 value of normal saline is unlikely to match exactly that of amniotic fluid. These variations could be taken into account in several ways. The data could be corrected by measuring the relevant organ fluid volumes, using planimetry or estimating using diameters and shape algorithms, as well as taking into account the T2 value difference between amniotic fluid and reference volume. Alternatively an algorithmic approach could be used to fit the MR based AFV to known reference volumes, avoiding the need for further measurements. Either approach would require a larger study with fetal and AFV measurements, using a wider range of gestational ages. In this regard, the linear regression relationship shown here should be used with caution. As with any linear regression equation, it is unwise to extrapolate the observation beyond the dataset limits presented, as it is unknown if the reported relationship holds beyond the extreme ends of our report observations. Amniotic fluid is relatively well auto regulated, but the osmolality can vary both with gestational age and, to a lesser extent, with maternal hydration[17], which could influence the T2 value of the fluid. Despite these factors we observed that the PH technique is highly predictive of amniotic fluid as measured using MSP in our cohort of healthy pregnancies.

Future work could explore measuring the T2 value of amniotic fluid non-invasively with MRI, and variations in the relative signal intensity of saline versus amniotic fluid could be addressed. A larger population study using MR PH based AFV estimation involving a wider range of gestational ages could provide useful reference data and demonstrate the relationship with AFV at other gestational ages.

Ultrasound estimation of AFV is recognised to be relatively inaccurate[18], particularly at higher and lower AFV. However these techniques are routinely used world-wide to

determine management and predict fetal outcome[3] because they are practical and relatively easy to perform. We have demonstrated that rapid MRI based projection hydrography measurement of AFV correlates better than US measurements with a reference AFV based on MSP calculation. If this MR approach can be further optimised, it may provide a substantially more accurate measure than ultrasound, with analysis rapid enough to be useful clinically. Accurate quantification of amniotic fluid volume is most likely to be of use where it has a large impact on management, such as in oligohydramnios and intra-uterine growth retardation. An improved non-invasive method of estimating AFV may also allow for more robust research on the influence of AFV on management and fetal outcome, an area where debate and controversy persists. This could allow the development of a more accepted standard for evaluating AFV during pregnancy which can be used selectively where there is clinical uncertainty caused by the current US methods.

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Figure Legends

Figure 1. (a) Example of MSP measurement of AFV on a single central 5mm slice in the sagittal plane with several regions of interest demonstrated on the image. (b) Example of PH measurement of AFV in the sagittal plane, in the same participant, with two regions of interest: one around the amniotic sac and the second around the 50ml saline reference volume.

Figure 2. Linear regression plots illustrating the ability of the rapid MRI PH technique (a), and the conventional ultra-sound metrics: AFI (b), and SDVP (c) to predict amniotic fluid as defined by the reference standard using MSP. The dashed lines represent the 95th confidence interval for the regression line and the dotted lines represents the 95th prediction intervals.

Figure 3. Bland-Altman plot comparing manual planimetry and projection hydrography. The dashed and dotted lines respectively represent the bias and 95% limit of agreement. Note the trend for a negative bias at lower AFV and positive bias at higher AFV

Figure 4. Bland-Altman plot comparing axial and sagittal manual planimetry. The dashed and dotted lines respectively represent the bias and 95% limit of agreement.

Figure 5. Illustrates the challenge of excluding the cord (white arrow) from each ROI, and also the time consuming process of defining such ROIs across the approximately 35-40slices required for every MP measurement.

Figure 6. The sequential PH acquisitions in the same subject with the same display parameters. Both (a) and (c) demonstrate fetal motion causing signal loss, whereas (b), (d) and (e) show reduced fetal motion and correspondingly increased fluid signal. The final PH figure was obtained from (b). Note the fetal structures are slightly blurred in (a) and (c) confirming motion has occurred.

Table Legends

Table 1. Distribution statistics and inter-rater agreement for the MR derived amniotic fluid metrics. (MSP – multisection planimetry, PH – projection hydrography, ICC – intra class correlation coefficient, CI – confidence interval**)**

Table 2. Linear regression summary statistics assessing the relationship between the rapid MRI technique (PH) and the conventional ultrasound metrics (AFI and SDVP) against the reference amniotic fluid metric (Sagittal MSP)