

Measurement of changes in body composition of piglets from birth to 4 kg using quantitative magnetic resonance (QMR)*

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Abstract

The purpose of this study was to use quantitative magnetic resonance (QMR) to measure changes in the body composition of piglets during growth from birth to 4 kg body weight. Using QMR, 60 pigs were scanned an average of 5 times starting at 2.7 ± 1.3 days of age (1.95 kg) and finally at 13.1 ± 4.3 days (4.14 kg). Regression analysis revealed that the rates of total body growth and fat and lean deposition were linear throughout this period. Subsequently, a second group of 235 pigs (109 males and 126 females) were scanned twice, first at 2.7 ± 1.2 days of age and then at 13.4 ± 3.1 days of age. The mean (\pm SD) rate of total body growth was 230 ± 57 g/day. The rates of fat and lean deposition were 40 ± 13 g/day and 191 ± 52 g/day, respectively. The rates of both fat and lean deposition were highly correlated ($P < 0.001$) with total body growth rate ($R^2 = 0.81$ and 0.93 , respectively) and the coefficient of determination between the rates of fat and lean deposition was 0.71 ($P < 0.001$). The results of this study demonstrate that QMR is a useful method for measuring changes in body composition in neonatal pigs. Furthermore, the results indicate that during the period of growth from birth to 4 kg, the rates of both fat and lean deposition are linear and highly correlated with total body growth.

Keywords: quantitative magnetic resonance, piglets, body composition

Introduction

The survival of low birth weight pigs in particular may depend on energy stores in the body (Girard 1981). The growth and composition of the neonatal pig is also of interest because of potential impact on subsequent growth and composition (Young & Sharma 1973, Mahan & Lepine 1991). Studies have found that factors such as birth weight (Powell & Aberle 1980, Poore & Fowden 2004) and nutrition during the neonatal period (Pond *et al.* 1965, Campbell & Dunkin 1983) can affect both growth and composition. Most studies have relied on chemical analysis or dissection of the carcass to measure composition (fat and lean) of the neonatal pig (Kuhn *et al.* 2001, Birkenfeld *et al.* 2006, Charneca *et al.* 2010), thus the impact on subsequent composition can only be inferred.

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It is difficult to accurately measure *in vivo* body composition of the young piglet. While many methods have been developed for measuring body composition, most have serious deficiencies for use in longitudinal studies. There are a number of criteria to be considered when selecting the most suitable method for measuring the body composition of a particular group of subjects. In all cases, accuracy and precision are very important. Previous results (Andres *et al.* 2010, Kovner *et al.* 2010) indicate that with proper calibration, QMR can provide accurate measurements of the water and lipid content of piglets in the 2 to 4 kg body weight range. Two additional major advantages of the QMR method over other methods are that QMR does not expose the subject to X-radiation and that QMR does not require immobilization (i.e., restraint or anaesthesia) of the subject.

The QMR method is a branch of nuclear magnetic resonance (NMR), for whole body measurement of fat, lean tissues, free water (not bound in various tissues), and total body water (TBW, water contained in all the liquids and in tissues) of live animals including humans. QMR differs from magnetic resonance imaging (MRI) in that the processed signal is obtained from the whole body at once (without spatial encoding) and it differs from magnetic resonance spectroscopy (MRS) in that the time domain signal (rather than spectrum) is processed directly. Principles of QMR are described elsewhere (Taicher *et al.* 2003, Tinsley *et al.* 2004, Kovner *et al.* 2010). Briefly, this system generates a signal that modifies the spin patterns of hydrogen atoms within the subject, and uses an algorithm to evaluate the resulting T1 and T2 relaxation curves specific to each of the four components measured: fat mass, lean muscle mass equivalent, total body water, and free water. Each component is estimated based on an individually derived T1/T2 relaxation curve fractionated from the total returned signal. This study used QMR to measure changes in the body composition of piglets during growth from birth to 4 kg body weight.

Material and methods

Using a QMR instrument (EchoMRI, Echo Medical Systems, Houston, TX, USA), 60 pigs were scanned an average of 5 times starting at 1.95 ± 0.42 kg (2.7 ± 1.3 days of age) and finally at 4.14 ± 0.52 kg (13.1 ± 4.3 days of age). The EchoMRI-Infants device is based on a permanent magnet with constant field of ≈ 0.021 T (Larmor frequency ≈ 880 kHz) and is designed for live subjects in mass range from 1 kg to 5 kg. The rates of total body growth and fat and lean deposition were calculated for each individual pig by linear regression analysis

$$Y = a + b \times X \quad (1)$$

where Y was the grams of fat or lean and X was age in days. For each analysis, the deposition rate was represented by the slope (b) of the regression equation. The mean coefficient of determination (R^2) was calculated for the entire group.

A second group of 235 pigs (109 males and 126 females) were scanned twice, first at 2.7 ± 1.2 days of age (2.04 ± 0.40 kg) and then at 13.4 ± 3.1 days of age (4.45 ± 0.86 kg). Fat, lean and water growth were calculated based on differences between the two measurements. Protein deposition was estimated as 0.25 water deposition (Campbell & Dunkin 1983). An additional, 74 piglets (3.2 ± 2.5 days of age, 1.75 ± 0.69 kg) were scanned only once; thus, the total number of scans for this group was 544, ($235 \times 2 + 74 = 544$).

The pigs used in these studies were of a Large White × Poland China cross. At the time of scanning the entire litter was removed from the sow and each pig was weighed individually using an electronic scale (± 0.02 kg). The piglets were scanned individually then the entire litter was returned to the sow. Each scan consisted of triplicate measurements, with each measurement lasting approximately 1.5 min. The accuracy of the QMR instrument for measuring total body fat and water had been previously calibrated and validated based on measurements and chemical analysis of 50 piglets weighing 1.2 to 4.3 kg (Kovner *et al.* 2010). Experimental animal protocols used in this study were approved by the Beltsville Area Institutional Animal Care and Use Committee, Beltsville, USA.

Statistical analysis was performed using Statgraphics Plus 5.1 procedures (Statistical Graphics Corporation 2001). Coefficients of determination and prediction equations were generated by linear and polynomial regression analysis. Differences in the mean values for body composition components were evaluated by the GLM procedure followed by a multiple range test that uses the Fisher's least significant difference (LSD) procedure to discriminate among the means at the 5% level.

Results and discussion

During studies of the growth of neonatal piglets it is important to be able to accurately assess changes in body composition. Body composition analysis results can be used to monitor and evaluate growth patterns, genetic improvement, dietary treatments, progression of chronic disease, and efficacy of medical interventions. Data reported by Manners & McCrea (1963) indicated that as pigs grow from 2 to 28 days of age there is a linear increase total body weight as well as in the fat and protein content in the body. This was based on the serial slaughter and chemical analysis of three litters of pig at five times between birth and 28 days of age. The first objective of the present study was to confirm those linear relationships using QMR to measure serial changes in body composition of live pigs.

From the 1st study, Table 1 shows the rates of fat and lean deposition that were observed when pigs were measured by QMR an average of five times during growth from approximately 2 to 4 kg body weight. The mean (\pm SD) rate of total body growth was 236 ± 76 g-day⁻¹ and was linear throughout the study period ($R^2 = 0.98 \pm 0.04$). This is close to the 250 to 270 g-day⁻¹ reported as the growth rate for modern genotypes during the 2- to 4-week suckling period (King *et al.* 1999). The rate is variable depending on the availability of milk which is also a function of litter size. There was a wide range in the rates of both fat and lean deposition and both were linear throughout the study period with coefficients of determination (R^2) of > 0.95 . Consistent with the results by Noblet & Etienne (1987), the rates of both fat and lean deposition were highly correlated ($P < 0.001$) with total body growth rate ($r = 0.88$ and 0.94 , respectively). The correlation (r) between the rates of fat and lean deposition was 0.74 ($P < 0.001$).

Having confirmed the linearity of growth for the total body, fat and lean; the next objective (2nd study) was to make additional measurements based on two time points. Table 2 shows the rates of fat and lean deposition that were observed when pigs were measured by QMR only twice during growth from approximately 2 to 4 kg body weight. Similar to the first group, the mean (\pm SD) rate of total body growth was 230 ± 57 g-day⁻¹. Likewise, measurements of the rates of fat and lean deposition were similar to the first study, confirming that these rates

were linear and that only the two measurements are needed for this growth period. By comparison, using dual energy X-ray absorptiometry (DXA) Dunshea *et al.* (2003) measured a fat deposition rate of 52.7 g/d and a lean deposition rate of 211 g/d for pigs with a growth rate of 258 g/d between 1 d and 21 d of age. That study was based on a single DXA measurement at 21 d of age and estimates of initial fat and lean content at 1 d of age.

Table 1
Rates of fat and lean deposition based on linear regression analysis of individual measurements (n=60)¹

Measurement	Mean, g/day±SD	Range, g/day	R ² (mean±SD)
Fat deposition rate	32±13	10.6-64.9	0.97±0.04
Lean deposition rate	188±60	39.1-353.6	0.95±0.10

¹Pigs were scanned by QMR an average of 5 times starting at 2.7±1.3 days of age (1.95±0.42 kg) and finally at 13.1±4.3 days (4.14±0.52 kg).

In agreement with the 1st study, the correlation between the rates of fat and lean deposition in the 2nd study was 0.83 ($P<0.001$). In the 2nd study, the ratio of total body growth rate to the rate of fat deposition was 5.6, compared to 6.9 reported by Noblet & Etienne (1987). The ratio of protein gain (estimated) to lipid gain was 0.93 in this study, compared to 1.06 reported by Noblet & Etienne (1987). This ratio is influenced by the energy content of the diet (Campbell & Dunkin 1983). In the study by Campbell & Dunkin (1983) for pigs growing from 1.8 to 6.5 kg while receiving adequate protein, as the energy intake increased there was a curvilinear increase in fat deposition – resulting in a curvilinear decrease in the ratio of protein gain to lipid gain, dropping from 3.31 to 0.90. These results suggest that QMR could be a useful tool for measuring energy deposition in pigs.

Table 2
Measurement of growth and composition based on two observations (mean±SD, n=235)¹

Scans	Age, days	Body Wt, g	Fat, g	Fat, %	Lean, g	Water, g
1st Scan	2.7±1.2	2 044±400	105±63	4.9±2.4	1 942±352	1 637±279
2nd Scan	13.4±3.1	4 451±859	521±156	11.5±2.0	3 930±765	3 216±622
Growth	Fat (g)	Fat (g/day)	Lean (g)	Lean (g/day)	Water (g)	Water (g/day)
Mean	416±147	40±13	1 987±623	191±52	1 579±505	152±41
Range	7-860	0.5-70	506-3 607	39-310	413-2 999	32-253

¹Pigs were scanned twice by QMR, first at 2.7±1.2 days of age (2.04±0.40 kg) and again at 13.4±3.1 days of age (4.45±0.86 kg).

Table 3 shows the coefficients of determination (R² values based on linear regression analysis) among growth and composition measurements of neonatal piglets from the 2nd study. The composition (fat, lean or water content) was more highly correlated with body weight than with age of the piglet. As indicated in Table 1, for individual pigs the deposition of both fat and lean tissue was linear with mean regression coefficients of >0.95. Thus, the reason for the lower relationship between composition and age for the 2nd study (Table 3) is likely due to the large variation in growth rate. The quantity and rates of fat and lean growth were highly correlated with the amount and rate of total body growth. However, the ratio of fat to lean growth showed a low relationship with the rate of total body growth.

The relationship between body weight and the percentage of total body fat is shown in Figure 1 which indicates that there is a curvilinear relationship between body weight and the percentage of body fat for neonatal pigs. This is consistent with the data reported by Manners & McCrea (1963) where the percentage of total body fat increased in a similar curvilinear pattern, peaking at approximately 18% fat as the pigs grew from 2 d to 28 d of age (1.5 to 9.9 kg).

Table 3
Coefficients of determination (R^2) for growth and composition measurements of neonatal piglets

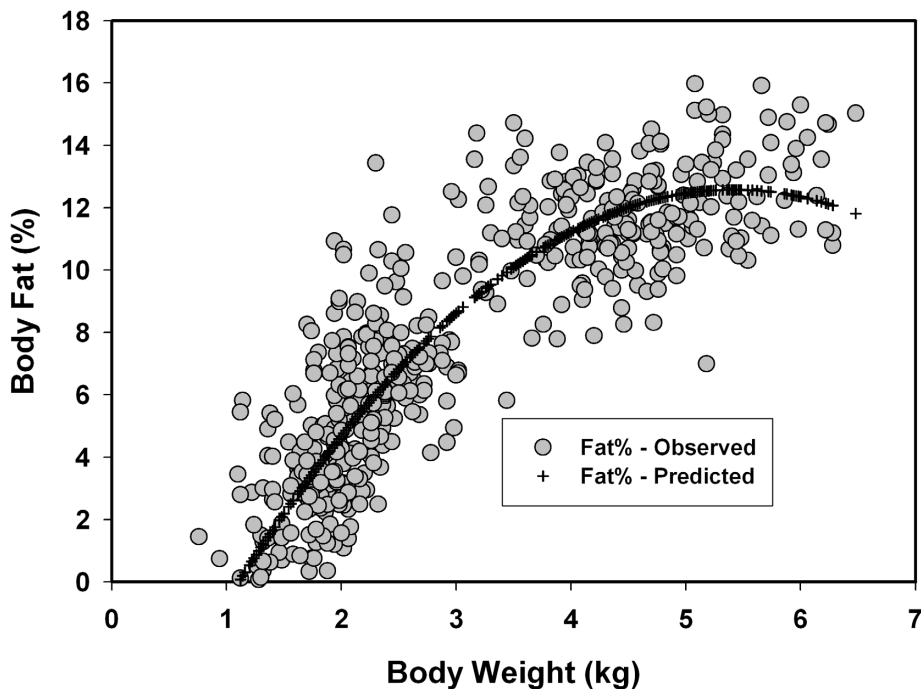
Dependent variables (Y)	Independent variables (X)	
Composition (n=544) ¹	Body weight, g	Age, days
Fat, g	0.94 ($P<0.001$)	0.74 ($P<0.001$)
Lean, g	0.99 ($P<0.001$)	0.71 ($P<0.001$)
Water, g	0.99 ($P<0.001$)	0.71 ($P<0.001$)
Body weight, g	--	0.74 ($P<0.001$)
Gain of fat or lean (n=235) ²	Body weight gain, g	
Fat gain, g	0.83 ($P<0.001$)	--
Lean gain, g	0.96 ($P<0.001$)	--
Rate of growth (n=235) ²	Rate of body weight growth, g/day	Rate of lean growth, g/day
Fat, g/day	0.81 ($P<0.001$)	0.71 ($P<0.001$)
Lean, g/day	0.93 ($P<0.001$)	--
Fat/lean	0.10 ($P<0.001$)	--
Rate of growth (n=235) ²	Birth weight, g	
Body weight, g/day	0.125 ($P<0.001$)	--
Lean, g/day	0.153 ($P<0.001$)	--
Fat, g/day	0.076 ($P<0.001$)	--
Fat/lean	0.003 ($P>0.01$, ns)	--

¹Based on QMR single scan data. ²Pigs were scanned twice by QMR, first at 2.7 ± 1.2 days of age (2.04 ± 0.40 kg) and again at 13.4 ± 3.1 days of age (4.45 ± 0.86 kg).

Table 3 also shows the coefficients of determination (R^2) between birth weight and the growth of neonatal piglets. Birth weight in pigs has been linked to differences in both growth rate and body composition. Low birth weight pigs exhibit a 15% to 30% lower growth rate (compared to medium and high birth weight pigs) in the 1st month of postnatal life (Campbell & Dunkin 1982, Wolter *et al.* 2002, Poore & Fowden 2004, Gondret *et al.* 2005 and 2006, Škorjanc *et al.* 2007). Based on chemical analysis of the whole body at birth, Rehfeldt & Kuhn (2006) reported that low birth weight pigs have less fat and protein and more water than their littermates. The birth weight of piglets included in the comparisons shown in Table 3 ranged from 0.84 to 2.42 kg with a mean \pm SD of 1.59 ± 0.08 kg. Only 12 piglets had a birth weight of ≤ 1.0 kg and 29 piglets had a birth weight of ≥ 2.0 kg. There was a low but significant correlation between birth weight and the growth rates of the total body, lean, and fat. However, birth weight was not significantly correlated with the fat:lean ratio.

It has been reported that the effect of birth weight on postnatal growth pattern is sex-specific with females being more dependent on originating birth weight than males (Poore & Fowden 2004). In the present study there was no difference between males and females with regard to birth weight, weight gain, lean gain, or fat gain ($P>0.05$). Likewise, the coefficient of determination (R^2) between birth weight and weight gain was quite similar for males and

females (0.12 vs. 0.13, respectively). The apparent discrepancy between the two studies may lie in the fact that the present study included all pigs (the entire spectrum of birth weights), whereas the study by Poore & Fowden (2004) contrasted low and high birth weight pigs and excluded all pigs that fell within the 95 % confidence interval of the mean.



Predicted values are based on polynomial regression analysis ($\text{fat \%} = -7.28402 + 7.34449 \times \text{Wt} - 0.679082 \times \text{Wt}^2$, $n=544$, $R^2=0.80$; standard error of estimate=1.99), Wt: body weight

Figure 1

Relationship between body weight and percentage of total body fat in pigs as measured by quantitative magnetic resonance (QMR).

In conclusion, the results of the study reported here indicate that during the period of growth from birth to 4 kg, the rates of both fat and lean deposition are linear and highly correlated with total body growth. The correlation between body composition and age was lower than with body weight and the correlation between body composition and birth weight was much lower. The rates of fat and lean deposition and their relationship to total body growth measured here using QMR were in agreement with other studies that relied on slaughter and chemical analysis. The unique feature of this study is that the results were obtained with live non-anesthetized pigs. These results demonstrate that QMR is a useful method for measuring changes in body composition in neonatal pigs that offers the advantage of being an accurate method that results in minimal stress to pig, permitting frequent serial measurements and thus avoids some of the assumptions associated with the serial or comparative slaughter approach.

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