

# Higher weight at birth is related to decreased maternal amino acid oxidation during pregnancy<sup>1-3</sup>

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## ABSTRACT

**Background:** Small size at birth is associated with cardiovascular disease in adult life. Decreased fetal growth may result from a limitation in the nutrient supply to the fetus. Net tissue deposition in the mother and fetus increases the demand for nitrogen, but because maternal consumption of protein does not increase, there must be a change in the partitioning of amino acids, away from oxidation and toward deposition.

**Objective:** Our objective was to characterize amino acid oxidation in pregnancy and to investigate whether the relative partitioning of amino acids was related to fetal growth.

**Design:** We determined amino acid oxidation as urea production in 25 women during mid (17–19 wk) and late (26–29 wk) gestation. Urea production was measured from urinary [<sup>15</sup>N-<sup>15</sup>N]urea excretion over 48 h after a single oral dose of [<sup>15</sup>N-<sup>15</sup>N]urea. We measured the infant's size at birth.

**Results:** For the group as a whole, urea excretion decreased and amino acid oxidation remained similar between mid and late pregnancy, but there was wide variation between the women. Heavier infants were born to the mothers in whom amino acid oxidation decreased the most during pregnancy (slope of regression line:  $-80 \text{ g} \cdot \text{g N}^{-1} \cdot \text{d}^{-1}$ ; 95% CI:  $-129, -31$ ;  $P = 0.003$ ). After adjustment for length of gestation and the infant's sex, the change in maternal amino acid oxidation explained 34% of the variation in birth weight.

**Conclusions:** Amino acid oxidation varied widely between the women during pregnancy. Understanding the ability of a pregnant woman to adapt metabolically may have implications for establishing dietary recommendations in pregnancy. *Am J Clin Nutr* 2002;76:852–7.

**KEY WORDS** Nutrition, pregnancy, amino acid oxidation, urea production, birth weight, fetal origins, white women

## INTRODUCTION

A significant body of epidemiologic evidence shows that growth and development during fetal life may have important effects on health in adult life. Many studies have shown that smaller size at birth is associated with a higher risk of developing cardiovascular disease, type 2 diabetes, and hypertension later in life (1–3). Graded associations are seen across the normal range of birth weight and are not explained by differences in gestational age or by socioeconomic influences in childhood or adult life (4).

During pregnancy, net protein synthesis leads to tissue deposition in the mother and fetus, increasing the demand for nitrogen

and amino acids (5). It is estimated that 925 g protein is accumulated, and recommendations suggest that protein intake should be increased by 6–10 g/d throughout pregnancy (6–8). However, women do not appear to increase their protein consumption as pregnancy advances (9), and therefore to enable net protein deposition, there must be a change in the partitioning of amino acids away from oxidation and toward deposition.

In the present study, we measured urea production as an index of amino acid oxidation. Previous researchers assessed amino acid oxidation from measurements of urinary nitrogen excretion (10). It is now recognized that not all the urea produced is excreted in urine: a portion passes into the colon where it is hydrolyzed by the colonic microflora. Hence, urea excretion underestimates the extent of amino acid oxidation (11, 12).

We studied women twice during pregnancy to characterize changes in amino acid oxidation and to investigate whether the relative partitioning of amino acids was related to the growth of the fetus. Because body mass index (BMI; in  $\text{kg}/\text{m}^2$ ) has been shown to influence aspects of protein metabolism (10, 13), we studied women with a BMI of 21.0–28.9.

## SUBJECTS AND METHODS

### Subjects

We recruited 30 healthy, nonsmoking, multiparous white women from 2 hospitals and a health center in the Southampton area when the women attended a midwives' antenatal booking clinic. To be eligible for the study, subjects had to be multiparous, aged between 24 and 33 y, and nonsmokers and had to have a height of 1.58–1.68 m and a BMI of 21.0–28.9. Length of gestation was estimated from the first day of the last menstrual period. The study was approved by the local ethics committee, and informed, written consent was obtained from each subject. The

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women were studied in their own homes with the use of noninvasive methods suitable for free-living individuals (14).

### Study design

Each subject was studied twice during pregnancy: at 17–19 wk gestation (mid pregnancy) and at 26–29 wk gestation (late pregnancy). On both occasions, measurements of amino acid oxidation and anthropometric measurements were carried out. In mid pregnancy each woman completed a 3-d food diary. When the subject delivered, the infant's body size was measured within 48 h.

### Amino acid oxidation

The rate of urea production is a direct measure of the rate of amino acid oxidation. Urea production was measured from the recovery in urine of a single oral dose of [<sup>15</sup>N-<sup>15</sup>N]urea. A sample of urine was collected before administration of the isotope to determine the background abundance of <sup>15</sup>N. Each subject ingested 100 mg [<sup>15</sup>N-<sup>15</sup>N]urea (99 atom%; Bioquote Ltd, York, United Kingdom) dissolved in 10 mL tap water between 0730 and 0930 (14). Urine was collected in 4 samples of 12 h each for the next 48 h. The urine was collected directly onto 98% (by wt) thymol (Sigma-Aldrich Co Ltd, Dorset, United Kingdom), the total urine volume was measured, and a 60-mL sample was acidified with 100 μL of a 6-mol HCl/L solution and frozen at –20°C until analysis. Isotopic enrichment was measured in each 12-h sample.

### Biochemical analysis

The urinary urea concentration was measured by the Berthelot method (15). Urea was chromatographically isolated from urine for mass spectrometry with the use of a short ion-exchange column (16). Nitrogen gas was liberated from urea by reaction with alkaline hypobromite in a monomolecular reaction: 2 nitrogen atoms from urea yield a single molecule of nitrogen gas. Thus, the relative proportions of [<sup>15</sup>N-<sup>15</sup>N]urea, [<sup>15</sup>N-<sup>14</sup>N]urea, and [<sup>14</sup>N-<sup>14</sup>N]urea can be determined. Measurements of enrichment were carried out in a triple collector isotope ratio mass spectrometer (SIRA 10; VG Isogas Instruments, Cheshire, United Kingdom).

### Calculations

Urea production was measured with the use of a stochastic model in which the recovery in urine of a single oral dose of [<sup>15</sup>N-<sup>15</sup>N]urea is determined, assuming a biological quasi-steady state (14). In this method it is presupposed that the oral dose of [<sup>15</sup>N-<sup>15</sup>N]urea is absorbed from the upper bowel and mixes freely with the urea pool within the body and hence that the fate of the [<sup>15</sup>N-<sup>15</sup>N]urea effectively traces the fate of unlabeled urea. There are 2 possible fates for the urea within the body pool. It can be excreted in urine as [<sup>15</sup>N-<sup>15</sup>N]urea and lost from the body, or it can pass to the lower bowel, where it is hydrolyzed by the microflora, in which case the nitrogen is returned to the body pool for further potential metabolic interaction. Hence the proportion of the dose recovered in urine is presumed to be the same as the proportion of endogenously produced urea that passes into urine. If the amount of urea excreted in urine over a defined period is known, it is possible to derive the production of urea over an equivalent period. Thus, if labeled urea reliably traces endogenous urea, production can be derived from

$$eu_{30}/d_{30} = Eu/P \quad (1)$$

where eu<sub>30</sub> is the excretion of [<sup>15</sup>N-<sup>15</sup>N]urea in mg, d<sub>30</sub> is the dose of [<sup>15</sup>N-<sup>15</sup>N]urea in mg, Eu is the total urea excretion in mg/48 h,

and P is urea production in mg/48 h. On the assumption that urea that is not recovered in urine has been hydrolyzed in the colon, it is possible to derive colonic hydrolysis (T) from

$$T = P - Eu \quad (2)$$

The nitrogen derived from the hydrolysis of urea is retained within the body and has 1 of 2 fates (17). A small proportion returns to urea formation, whereas a greater proportion is incorporated into the metabolically active pool of nitrogen within the body.

The period over which urine is collected must be long enough to ensure that all of the [<sup>15</sup>N-<sup>15</sup>N]urea has been cleared from the body pool and recovered in the urine. Earlier experience has suggested that 48 h is sufficient (14).

In 2 women, amino acid oxidation was very high (>3 SDs greater than the mean value for the group), raising the possibility that they were infected with *Helicobacter pylori*, in which case the dose of [<sup>15</sup>N-<sup>15</sup>N]urea would have been hydrolyzed before absorption, thus producing erroneous results. Excessive hydrolysis of an oral dose of labeled urea is used as a diagnostic test for infection with *H. pylori*. There was no increase, however, in the excretion of [<sup>15</sup>N-<sup>14</sup>N]urea in either subject, making it unlikely that they had an active, ongoing infection; therefore these 2 women were not excluded from the analyses (18).

### Protein intake and test diet

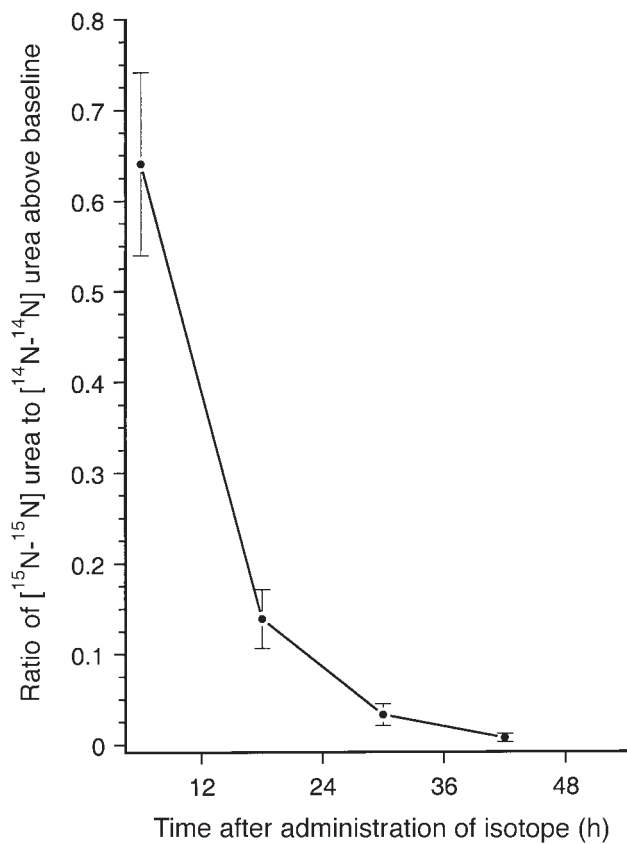
Current energy and protein intakes were assessed with the use of the 3-d food diary (19). To enable accurate assessment of intake during the study period, food comprising items such as cereal, milk, bread, ready-prepared meals (eg, lasagna), vegetables, and fruit was supplied for the duration of the 48-h study period; this food provided an amount of energy and protein similar to that of the food reported in the food diary. Additional food was available at the woman's request.

### Anthropometry

Maternal height was measured with a Harpenden pocket stadiometer (Chasmors Ltd, London) to the nearest 0.1 cm, and weight was measured with Seca portable digital scales (Seca, Hamburg, Germany) to the nearest 100 g. At birth, each infant was weighed to the nearest 5 g with the use of digital scales. Using a Harpenden neonatometer (Holtain, Crymych, United Kingdom), 2 trained research nurses measured each infant's crown-to-heel length 3 times to the nearest 0.1 cm, and the mean value was used in the analysis. We calculated the ponderal index at birth as birth weight/length<sup>3</sup> (in kg/m<sup>3</sup>). The placenta was weighed on digital scales after removing the umbilical cord and membranes (9).

### Statistical analysis

Results are expressed as means ± SDs. Paired *t* tests were used to detect statistically significant differences between measurements in mid and late pregnancy. Linear regression analysis was used to explore the associations between maternal amino acid oxidation and the infant's size at birth. In late pregnancy, there was a significant positive association between urea production and the length of gestation at the time of the measurement and between urea excretion and the length of gestation at the time of the measurement. Therefore, both were adjusted to the mean length of gestation at the time of measurement (28.3 wk). Measurements of urea hydrolysis were positively skewed and were



**FIGURE 1.** Mean ( $\pm$ SD) ratio of [ $^{15}\text{N}$ - $^{15}\text{N}$ ]urea to [ $^{14}\text{N}$ - $^{14}\text{N}$ ]urea above baseline, representing urinary excretion over 48 h of a dose of [ $^{15}\text{N}$ - $^{15}\text{N}$ ]urea in 25 women in mid pregnancy.

therefore log transformed for analysis. The value for urea production includes a contribution from nitrogen that was derived from urea that was hydrolyzed in the colon and subsequently returned to urea production. Thus, the value for urea production may be greater by this amount ( $\approx 5\%$  of the total). All analyses were repeated by using a value for urea production that did not include this contribution from recycled urea nitrogen. Applying this correction did not alter any of the associations reported, and thus these data are not shown.

**TABLE 1**

Characteristics of the 27 women

	All women ( <i>n</i> = 27)	Mid pregnancy ( <i>n</i> = 25)	Late pregnancy ( <i>n</i> = 26)	Paired difference <sup>1</sup> ( <i>n</i> = 25)	<i>P</i>
Age (y)	30.6 $\pm$ 2.3 <sup>2</sup>				
Height (m)	1.63 $\pm$ 0.03				
Weight (kg)	—	67.9 $\pm$ 6.8	73.0 $\pm$ 7.2	5.0 $\pm$ 1.8 (4.3, 5.8)	<0.001
BMI (kg/m <sup>2</sup> )	—	25.7 $\pm$ 2.4	27.6 $\pm$ 2.5	1.9 $\pm$ 0.7 (1.6, 2.2)	<0.001
Protein intake (g N/d)	—	13.9 $\pm$ 2.8	14.3 $\pm$ 2.4	0.4 $\pm$ 1.7 (−0.3, 1.1)	0.2
Energy intake (MJ/d)	—	10.8 $\pm$ 2.9	11.0 $\pm$ 2.0	0.27 $\pm$ 1.65 (−0.41, 0.95)	0.4
Urea excretion (g N/d)	—	8.4 $\pm$ 1.4	7.7 $\pm$ 1.4	−0.7 $\pm$ 1.3 (−1.3, −0.2)	0.01
Amino acid oxidation (g N/d)	—	12.0 $\pm$ 2.7	11.8 $\pm$ 2.6	−0.3 $\pm$ 2.6 (−1.3, 0.8)	0.6
Urea hydrolysis (g N/d) <sup>3</sup>	—	3.3 $\pm$ 1.5	3.7 $\pm$ 1.5	—	0.2

<sup>1</sup>95% CI in parentheses.

<sup>2</sup> $\bar{x} \pm$  SD.

<sup>3</sup>Geometric  $\bar{x} \pm$  SD. The paired difference as a percentage was 13.1  $\pm$  65.1% (95% CI: −8.0%, 39.2%).

Results are expressed as g nitrogen/d. It was assumed that 1 g nitrogen is equivalent to 6.25 g protein. The statistical package SPSS version 7.5 (SPSS Inc, Chicago) was used for statistical analyses.

## RESULTS

Of the 30 women who agreed to take part in the study, 1 miscarried and 2 dropped out for medical reasons. Of the remaining 27 women, 25 and 26 completed the 48-h urine collection in mid pregnancy (mean gestation: 18 wk) and late pregnancy (mean gestation: 28 wk), respectively. The maximum number of subjects possible was included in each analysis.

The pattern of isotope excretion over the 48 h of measurement for the studies completed in mid pregnancy is shown in **Figure 1**. In this time the dose of [ $^{15}\text{N}$ - $^{15}\text{N}$ ]urea had been cleared from the body pool and recovered in the urine.

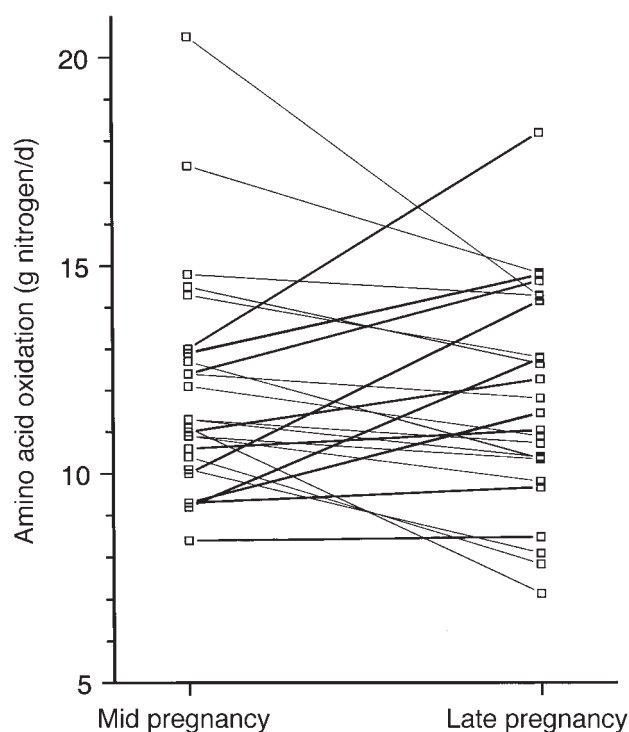
## Anthropometry and amino acid oxidation

The anthropometric characteristics, dietary protein intake, and amino acid oxidation of the women over the 48-h study period are shown in **Table 1**. Dietary protein intake and amino acid oxidation in late pregnancy were not significantly different from those in mid pregnancy, but urea excretion decreased significantly from mid to late pregnancy. In both mid and late pregnancy, amino acid oxidation was greater than urea excretion. The amount of urea hydrolyzed in the colon was not significantly different between mid and late pregnancy. For the male and female infants, respectively, birth weight was 3.90  $\pm$  0.42 and 3.52  $\pm$  0.43 kg, birth length was 52.3  $\pm$  1.6 and 50.3  $\pm$  1.5 cm, the ponderal index (in kg/m<sup>3</sup>) was 27.3  $\pm$  1.9 and 27.7  $\pm$  2.4, and placental weight (in 26 women) was 543  $\pm$  50 and 497  $\pm$  100 g.

## Intraindividual variability

Although for the group as a whole there was no significant difference in either mean protein intake or amino acid oxidation between mid and late pregnancy, the variation between individuals was substantial at each time point. The CVs for protein intake in mid and late pregnancy were 20.1% and 16.8%, respectively, and the CVs for amino acid oxidation in mid and late pregnancy were 22.5% and 22.0%, respectively. Furthermore, the response to pregnancy also varied between individuals. The change ( $\Delta$ ) in amino acid oxidation between the 2 time





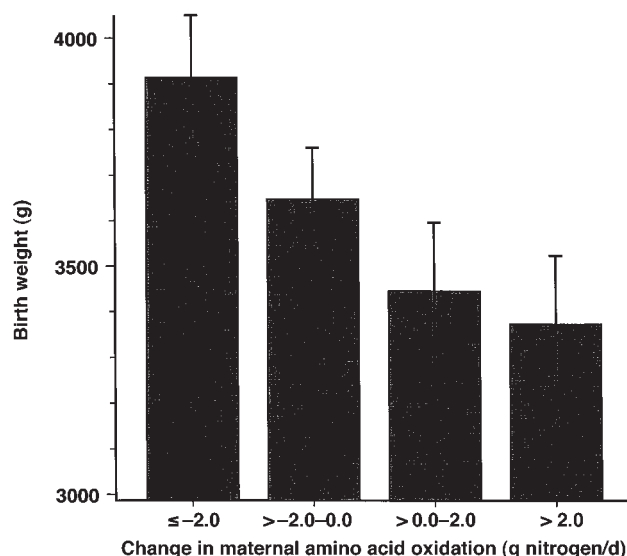
**FIGURE 2.** Maternal amino acid oxidation in mid and late pregnancy. Each square at each time point represents one subject. Lines connect paired data. (Values in late pregnancy were adjusted for the length of gestation at the time of the measurement.)

points was calculated and ranged from  $-6.2$  to  $5.2$  g N/d in the 25 subjects for whom paired data were available (**Figure 2**). With the use of linear regression analysis, 23% of the variation in  $\Delta$ amino acid oxidation was accounted for by  $\Delta$ dietary protein intake. As dietary protein intake increased relative to that during mid pregnancy, amino acid oxidation also increased relative to that during mid pregnancy (the slope of the regression line indicated a  $0.73$ -g N/d increase per gram  $\Delta$ protein intake; 95% CI:  $0.16, 1.31$ ;  $P = 0.02$ ). There was no relation between amino acid oxidation in mid or late pregnancy and the mother's height, weight, or BMI.

### Fetal growth

The difference between the women in the metabolic response to pregnancy was explored in relation to the size of their infants at birth. All of the infants (21 females and 6 males) were delivered after 38.7 wk gestation (271 d). Birth weight, length, and placental weight were higher in the infants who had a greater gestational age at birth and were higher in the males than in the females. We therefore adjusted these 3 variables for length of gestation and the infant's sex in the following analyses. The ponderal index was higher in the infants who had a greater gestational age at birth and was therefore adjusted accordingly.

There was no relation between either maternal BMI in early pregnancy or weight change between early and late pregnancy and birth size. There was also no relation between amino acid oxidation at either time point in pregnancy and the infant's length, ponderal index, or placental weight. The women who had lower amino acid oxidation in late pregnancy, but not during mid pregnancy,



**FIGURE 3.** Relation between mean ( $\pm$  SEM) birth weight, adjusted for length of gestation and the infant's sex, and change in maternal amino acid oxidation from mid to late pregnancy. The 25 women were divided into 4 groups on the basis of the change in amino acid oxidation:  $\le -2.0$  g N/d ( $n = 6$ ),  $> -2.0-0.0$  g N/d ( $n = 9$ ),  $> 0.0-2.0$  g N/d ( $n = 5$ ), and  $> 2.0$  g N/d ( $n = 5$ ).

tended to have heavier babies. As amino acid oxidation in late pregnancy decreased by 1 g N/d, there was a trend for the infant's birth weight to increase by 49 g (95% CI:  $-7, 104$  g  $\cdot$  g  $N^{-1} \cdot d^{-1}$ ;  $P = 0.08$ ). However, when  $\Delta$ amino acid oxidation from mid to late pregnancy was considered, the association with birth weight was strongly significant. The mothers in whom amino acid oxidation decreased more between mid and late pregnancy had heavier babies (slope of the regression line:  $-80$  g  $\cdot$  g  $N^{-1} \cdot d^{-1}$ ; 95% CI:  $-129, -31$  g  $\cdot$  g  $N^{-1} \cdot d^{-1}$ ;  $P = 0.003$ ). This relation is shown in **Figure 3**, in which the women are divided into 4 groups on the basis of  $\Delta$ amino acid oxidation for clarity of presentation. The women in whom amino acid oxidation decreased more between mid and late pregnancy also had fatter babies, as measured by the ponderal index (slope of the regression line:  $-0.41$  kg/m $^3 \cdot$  g  $N^{-1} \cdot d^{-1}$ ; 95% CI:  $-0.72, -0.10$ ;  $P = 0.01$ ). There was no relation between  $\Delta$ amino acid oxidation and birth length or placental weight.

The predictive value of amino acid oxidation late in pregnancy and  $\Delta$ amino acid oxidation was investigated in the following way. Because the late pregnancy value of amino acid oxidation is correlated with  $\Delta$ amino acid oxidation, it is statistically inappropriate to include both these variables in a multivariate analysis. Therefore, the mean amino acid oxidation for each subject was calculated and used in the multivariate analysis. The association between  $\Delta$ amino acid oxidation and birth weight remained and was independent of the absolute amino acid oxidation (**Table 2**). For this group of women, the range of  $\Delta$ amino acid oxidation (calculated as the mean  $\pm$  2SD) predicted a difference in birth weight of 832 g. After adjustment for length of gestation and the infant's sex,  $\Delta$ amino acid oxidation explained 34% of the variation in birth weight.



**TABLE 2**

Multiple linear regression of infant weight (in g) on mean and change ( $\Delta$ ) in maternal amino acid oxidation and on changes in maternal amino acid oxidation and protein intake in 25 women<sup>1</sup>

Independent variable	$\beta^2$	<i>P</i>
$\Delta$ Amino acid oxidation (g N/d)	-80 (-130, -30)	0.003
Mean amino acid oxidation (g N/d)	-12 (-68, 44)	0.7
$\Delta$ Amino acid oxidation (g N/d)	-69 (-125, -13)	0.02
$\Delta$ Protein intake (g N/d)	-37 (-122, 49)	0.4

<sup>1</sup>Adjusted for the infant's sex and length of gestation.

<sup>2</sup>Gram change in birth weight per unit increase in the independent variable. 95% CI in parentheses.

Because  $\Delta$ protein intake predicted  $\Delta$ amino acid oxidation, the association between  $\Delta$ protein intake and birth weight was investigated. As  $\Delta$ protein intake decreased by 1 g N/d, the infant's birth weight increased by 86 g (95% CI: 0.7, 171 g  $\cdot$  g N<sup>-1</sup>  $\cdot$  d<sup>-1</sup>; *P* = 0.05). In a simultaneous analysis that included both  $\Delta$ amino acid oxidation and  $\Delta$ protein intake,  $\Delta$ amino acid oxidation, but not  $\Delta$ protein intake, remained a significant predictor of birth weight (Table 2). In this analysis, a total of 65% of the variation in birth weight was explained by the predictor variables. Length of gestation and the infant's sex explained 43% of the variation in birth weight. When  $\Delta$ amino acid oxidation was added, an additional 20% of the variation was explained, and  $\Delta$ protein intake explained an additional 2%.

## DISCUSSION

We measured amino acid oxidation in 25 pregnant women at 17–19 and 26–29 wk gestation. There was wide variation between the women in amino acid oxidation at both times and in the change over the 10-wk period. In previous studies substantial variation between women has been found, the basis of which is not clear (20–23). The present study is the first one in which the possible implications of this variation were explored in relation to the growth of the fetus. We showed that mothers in whom amino acid oxidation decreased more between mid and late pregnancy had heavier babies. This finding was independent of any change in protein intake. The change in amino acid oxidation in the mother, length of gestation, and the infant's sex explained 63% of the variation in birth weight. There are few other markers of metabolism in normal pregnancy that can account for such a large proportion of the variation in birth weight; Catalano et al (24) showed that measures of insulin sensitivity are strongly associated with birth weight ( $R^2 = 0.28$ ). The results of the present study suggest that the relative partitioning of amino acids between oxidation and deposition during this period may be crucial with respect to the nutrient supply to the fetus.

During pregnancy complex metabolic changes take place at different times to support the changing needs of the developing fetus. We chose to study the period between 18 and 28 wk gestation. At the beginning of this time, the velocity of fetal growth in length is at its greatest (25). The period of maximum increase in absolute weight, however, occurs later, after 30 wk gestation. In agreement with other evidence (20), our data show that there are important changes over this period and suggest that changes in maternal nitrogen metabolism may anticipate the needs of the fetus because


those changes occur in advance of any increase in fetal demand. This may explain why we found an association between the mother's amino acid oxidation and the infant's weight, but not length. A progressive decrease in amino acid oxidation from mid to late pregnancy would reflect effective conservation of amino acids and nitrogen, with preferential partitioning toward net deposition. In some women in our study, amino acid oxidation increased, implying a net degradation of protein. The babies born to these women were lighter. We did not have measures of the infant's fat or lean body mass, but we used the ponderal index as a measure of thinness. Because the infant's length was not associated with amino acid oxidation, these infants were also thinner. It will be important to understand how amino acid oxidation relates to the availability of amino acids to the fetoplacental unit and to the growth and body composition of the fetus.

The values obtained for amino acid oxidation in the present study were similar to the values we obtained previously with the use of the same method (22, 23). Kalhan (20) has reported a significant decrease in urea synthesis expressed relative to body weight between the first and last trimesters, and others have shown that ureogenesis is lower in the last trimester than it is post partum (21, 26). Our data suggest that this apparent decrease is more likely to reflect changes in maternal body weight than an absolute decrease in urea synthesis. When we expressed our data per kilogram weight, we also found a significant decrease in amino acid oxidation. Expressing the data in this way, however, did not remove the variation between individuals.

The measurement of amino acid oxidation in the present study does not differentiate between the contribution from the maternal tissues and that from the fetal tissues, but it is unlikely that the relation between  $\Delta$ amino acid oxidation and birth weight can be explained on the basis of changes in the fetal compartment alone. Fetal urea production has been estimated at 540 mg urea  $\cdot$  kg<sup>-1</sup>  $\cdot$  d<sup>-1</sup> at term (250 mg N  $\cdot$  kg<sup>-1</sup>  $\cdot$  d<sup>-1</sup>) (27), similar to values measured in newborn infants (28). In the present study birth weight was 3.6 kg, and therefore urea production in the fetus at term would have been 1.94 g urea/d (0.91 g N/d). This is <8% of the total. At 28 wk gestation, when the fetus would have weighed about 1 kg, the total fetal contribution would have been <3%. This is insufficient to account for either the variability between the women or the change between mid and late pregnancy. It appears that the change in the partitioning of amino acids takes place predominantly in the mother in anticipation of increased demands by the fetus.

It is not clear what drives the overall partitioning of amino acids in the mother between deposition and oxidation. The fetus utilizes amino acids both as precursors for the synthesis of protein and other metabolically active compounds, such as DNA, RNA, creatine, and neurotransmitters, and as a source of energy. It is possible therefore that amino acid partitioning may be affected by the needs of the fetus for energy (29). We did not have any information on the mother's glucose or insulin physiology, but it is possible that the mothers in whom urea production decreased during pregnancy were those who had higher plasma glucose concentrations. In nondiabetic women, increased plasma glucose concentrations within a normal range are associated with greater infant size (30, 31). A second explanation for the observed relation between amino acid oxidation and birth weight may be that oxidation reflects the goodness of fit between the pattern of amino acids provided in the diet and the needs of the fetus. Although the fetus requires an adequate supply of essential amino acids, the

greatest demand quantitatively is for certain nonessential amino acids (32). Those amino acids in excess of requirements for synthetic processes must be catabolized. This may underlie our observation that the women whose protein intake increased more during pregnancy had smaller infants, because higher intakes of protein were associated with higher amino acid oxidation and because increased oxidation may result when the fit is poor (33). Epidemiologic studies have shown that a high energy intake from protein is associated with low birth weight (34).

In summary, we found that amino acid oxidation varied widely during pregnancy in a group of well-nourished women. These metabolic differences may reflect the various abilities of individual women to adapt to pregnancy and conserve nitrogen. Understanding the ability of a woman to adapt metabolically during pregnancy and how this interacts with her body composition and diet may have implications for establishing dietary recommendations in pregnancy. 

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## REFERENCES

- Huxley RR, Shiell AW, Law CM. The role of size at birth and postnatal catch-up growth in determining systolic blood pressure: a systematic review of the literature. *J Hypertens* 2000;18:815–31.
- Barker DJP. Mothers, babies and health in later life. 2nd ed. London: Churchill Livingstone, 1998.
- Barker DJP. Fetal origins of coronary heart disease. *BMJ* 1995;311:171–4.
- Leon DA, Lithell HO, Vagero D, et al. Reduced fetal growth rate and increased risk of death from ischaemic heart disease: cohort study of 15 000 Swedish men and women born 1915–29. *BMJ* 1998;317:241–5.
- Duggleby SL, Jackson AA. Relationship of maternal protein turnover and lean body mass during pregnancy and birth length. *Clin Sci* 2001;101:65–72.
- Hytten FE, Leitch I. The physiology of human pregnancy. 2nd ed. Oxford, United Kingdom: Blackwell Scientific Publications, 1971.
- Energy and protein requirements. Report of a joint FAO/WHO/UNU Expert Consultation. *World Health Organ Tech Rep Ser* 1985;724:1–206.
- Institute of Medicine (US). Nutrition during pregnancy. Washington, DC: National Academic Press, 1990.
- Godfrey K, Robinson S, Barker DJP, Osmond C, Cox V. Maternal nutrition in early and late pregnancy in relation to placental and fetal growth. *BMJ* 1996;312:410–4.
- Henry CJK. Quantitative relationships between protein and energy metabolism: influence of body composition. In: Scrimshaw NS, Schürch B, International Dietary Energy Consultancy Group, eds. Protein-energy interactions. Lausanne, Switzerland: Nestlé Foundation, 1992:191–200.
- Walser M, Bodenlos LJ. Urea metabolism in man. *J Clin Invest* 1959;38:1617–26.
- Jackson AA. Critique of protein-energy interactions in vivo: urea kinetics. In: Scrimshaw NS, Schürch B, International Dietary Energy Consultancy Group, eds. Protein-energy interactions. Lausanne, Switzerland: Nestlé Foundation, 1992:63–79.
- Child SC, Soares MJ, Reid M, Persaud C, Forrester T, Jackson AA. Urea kinetics varies in Jamaican women and men in relation to adiposity, lean body mass and protein intake. *Eur J Clin Nutr* 1997;51:107–15.
- Jackson AA, Danielsen MS, Boyes S. A noninvasive method for measuring urea kinetics with a single dose of [<sup>15</sup>N<sup>15</sup>N]urea in free living humans. *J Nutr* 1993;123:2129–36.
- Kaplan A. Urea nitrogen and urinary ammonia. In: Meites S, ed. Standard methods in clinical chemistry. New York: Academic Press, 1965:245–56.
- Jackson AA, Golden MHN, Jahoor PF, Landman JP. The isolation of urea nitrogen and ammonia nitrogen from biological samples for mass spectrometry. *Anal Biochem* 1980;105:14–7.
- Jackson AA, Picou D, Landman J. The non-invasive measurement of urea kinetics in normal man by a constant infusion of <sup>15</sup>N<sup>15</sup>N-urea. *Hum Nutr Clin Nutr* 1984;38:339–54.
- Hibbert JM, Forrester T, Jackson AA. Urea kinetics: comparison of oral and intravenous dose regimens. *Eur J Clin Nutr* 1992;46:405–9.
- Robinson S, Godfrey K, Osmond C, Cox V, Barker D. Evaluation of a food frequency questionnaire used to assess nutrient intakes in pregnant women. *Eur J Clin Nutr* 1996;50:302–8.
- Kalhan SC. Protein metabolism in pregnancy. *Am J Clin Nutr* 2000;71(suppl):1249S–55S.
- Kalhan SC, Tserng K-Y, Gilfillan C, Dierker LJ. Metabolism of urea and glucose in normal and diabetic pregnancy. *Metabolism* 1982;31:824–33.
- Forrester T, Badaloo AV, Persaud C, Jackson AA. Urea production and salvage during pregnancy in normal Jamaican women. *Am J Clin Nutr* 1994;60:341–6.
- McClelland ISM, Persaud C, Jackson AA. Urea kinetics in healthy women during normal pregnancy. *Br J Nutr* 1997;77:165–81.
- Catalano PM, Drago NM, Amini SB. Maternal carbohydrate metabolism and its relationship to fetal growth and body composition. *Am J Obstet Gynecol* 1995;172:1464–70.
- Tanner JM. Foetus into man: physical growth from conception to maturity. 2nd ed. Ware (Herts), United Kingdom: Castlemead Publications, 1989.
- Olufemi OS, Whittaker PG, Lind T. Glycine and urea metabolism during normal and diabetic pregnancies. *Proc Nutr Soc* 1991;50:200A (abstr).
- Gresham EL, Simons PS, Battaglia FC. Maternal-fetal urea concentration difference in man: metabolic significance. *J Pediatr* 1971;79:809–11.
- Steinbrecher HA, Griffiths DM, Jackson AA. Urea production in normal breast-fed infants measured with primed/intermittent oral doses of [<sup>15</sup>N, <sup>15</sup>N]urea. *Acta Paediatr* 1996;85:656–62.
- Owens JA, Owens PC, Robinson JS. Experimental fetal growth retardation: metabolic and endocrine aspects. In: Gluckman PD, Johnston BM, Nathanielsz PW, eds. Advances in fetal physiology. New York: Perinatology Press, 1989:263–86.
- Langhoff-Roos J, Wibell L, Gebre-Medhin M, Lindmark G. Maternal glucose metabolism and infant birth weight: a study in healthy pregnant women. *Diabetes Res* 1988;8:165–70.
- Farmer G, Russell G, Hamilton-Nicol DR, et al. The influence of maternal glucose metabolism on fetal growth, development and morbidity in 917 singleton pregnancies in nondiabetic women. *Diabetologia* 1988;31:134–41.
- Widdowson EM. Chemical composition and nutritional needs of the fetus at different stages of gestation. In: Aebi H, Whitehead R, eds. Maternal nutrition during pregnancy and lactation. Bern, Switzerland: Hans Huber, 1980:39–48.
- Jackson AA. Maternal and fetal demands for nutrients; significance of protein metabolism. In: O'Brien PMS, Wheeler T, Barker DJP, eds. Fetal programming. Influences on development and disease in later life. London: RCOG, 1999:246–70.
- Campbell DM, Hall MH, Barker DJP, Cross J, Shiell AW, Godfrey KM. Diet in pregnancy and the offspring's blood pressure 40 years later. *Br J Obstet Gynaecol* 1996;103:273–80.