Cysteine is the only sulfhydryl-containing amino acid in proteins and is the thiol residue in glutathione (1). In addition to its importance in the storage and transport of cysteine, glutathione plays a pivotal role in detoxification by the action of glutathione S-transferases and in scavenging free (oxygen) radicals by the action of glutathione peroxidases (1).

Thiol metabolism may be altered during pregnancy. In healthy pregnant women, plasma concentrations of cysteine, glutathione, and homocysteine are decreased, whereas increased homocysteine and cysteine concentrations are seen in pathologic conditions, such as pre-eclampsia, in which oxidative stress may play an important role (2–4).

Thiols may have important physiological functions in fetal metabolism. Although protein and amino acid turnover in the human placenta has been studied extensively (5–7), few data concerning fetal concentrations of thiols and placent maternal-fetal thiol interactions are currently available (8). During normal pregnancy, fetal growth depends on the supply of nutrients from the mother, and a clear correlation between maternal and fetal amino acid and homocysteine concentrations has been shown (6–8). Decreased concentrations of amino acids in the umbilical artery, as compared with the umbilical vein, have been interpreted as an uptake of amino acids into fetal tissues where they may be used in protein biosynthesis or as sources of energy (5). We studied fetal and maternal thiol plasma concentrations in normal pregnancies to determine reference concentrations for cysteine, homocysteine, and cysteinylglycine in arterial and venous umbilical cord plasma to gain insight into maternal-fetal thiol interactions.

Arterial and venous umbilical cord blood samples from 320 consecutive neonates were drawn in heparinized tubes immediately after birth (cat. no. 260545; Kemper Medical) from March 1997 to January 1998 at the Department of Obstetrics/Gynecology of the ‘Nij Smellinghe’ Hospital. Approval for this study was granted by the Institutional Review Board of the hospital. Blood gas values were assessed on an ABL-330 analyzer (Radiometer Nederland). Samples with a pH difference between arterial and venous umbilical cord blood <0.02 pH units and samples from neonates with a gestational age <37 weeks, an umbilical artery pH <7.20, a birth weight below the 10th percentile according to Kloosterman (9), or who were the offspring of women with a diastolic blood pressure >90 mmHg during gestation were excluded. Antecubital maternal venous blood of 35 women was collected in parallel with the umbilical cord samples after informed consent. Samples were taken in heparinized tubes (cat. no. 367684; Becton and Dickinson) 4 h before elective caesarian delivery or <15 min after vaginal birth. Blood was centrifuged within 10 min at 1200 × g for 10 min at room temperature. Plasma samples were stored at −30 °C for ~2 years until analysis. Plasma concentrations of total cysteine (tCys), total homocysteine (tHcy), and total cysteinylglycine (tCysGly) in 195 umbilical cord (102 males and 92 females; no gender was recorded for 1 neonate) and 35 maternal samples were determined by HPLC as described previously (2).

After log transformation to approach normalization, we analyzed the data by the paired t-test to assess statistical differences between maternal venous blood and arterial
Cysteinylglycine 33 (20–50) 35 (20–51) 0.005
Homocysteine 9.6 (4.8–17.4) 8.8 (4.9–20.4) 0.009
Cysteine 207 (146–299) 203 (134–303) 0.0002

The median (central 0.95 interval) characteristics for the study group (n = 195) were as follows: maternal age, 29 years (20–41 years); gestational age, 40 weeks, 3 days (37 weeks, 3 days to 42 weeks, 3 days); maternal blood pressure, 80 mmHg, phase IV Korothoff (K4; 60–90 mmHg); birth weight, 3510 g (2800–4570 g); and placental weight, 695 g (500–980 g). These characteristics were representative of the population as admitted for term deliveries in the Drachten Hospital. The maternal characteristics of the subgroup (n = 35) were not different from the total study group.

In arterial umbilical cord plasma, median (central 0.95 interval) values for the following were significantly lower than in venous umbilical cord plasma: PO₂, 17 kPa (9–34 kPa) for arterial vs 28 kPa (16–43 kPa) for venous (P < 0.0001); HCO₃⁻, 19.2 mmol/L (14.7–24.5 mmol/L) for arterial vs 20.2 mmol/L (16.7–23.0 mmol/L) for venous (P < 0.0001); pH 7.27 (7.20–7.37) for arterial vs 7.34 (7.26–7.43) for venous (P < 0.0001); and Hb 138 g/L (117–161 g/L) for venous (P < 0.0001). Apgar at 1 min was 8.5 (7.0–9.5) for arterial vs 8.1 (7.0–8.5) for venous (P = 0.0001).

Concentrations of tCys and tHcy were significantly lower in arterial compared with venous umbilical cord plasma (P = 0.0022 and P = 0.009, respectively; see Table 1), whereas concentrations of tCysGly were significantly higher in arterial compared with venous umbilical cord plasma (P = 0.005).

Maternal and venous and arterial umbilical cord plasma in the subgroup of 35 cases are presented in Fig. 1. Maternal tCys concentrations were lower than venous umbilical cord plasma (P = 0.04), whereas there was a tendency for higher tCys concentrations in venous vs arterial umbilical cord plasma (P = 0.06). Arterial umbilical cord tCys concentrations tended to be higher than maternal concentrations (P = 0.1). Associations were found between tCys concentrations in maternal and venous umbilical cord plasma (r = 0.84; P < 0.0001), venous and arterial umbilical cord plasma (r = 0.82; P < 0.0001), and arterial umbilical cord and maternal plasma (r = 0.81; P < 0.0001).

Fig. 1. Maternal and corresponding venous and arterial umbilical cord thiol concentrations (n = 35).

Median (central 0.95 interval) values are given in µmol/L. NS, not significant.

Table 1. Thiol concentrations in umbilical cord (n = 195).

<table>
<thead>
<tr>
<th>Thiol</th>
<th>Venousa</th>
<th>Arteriala</th>
<th>P</th>
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<tr>
<td>Cysteine</td>
<td>207 (146–299)</td>
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a Medians (central 0.95 intervals) are given in µmol/L.
Effect of Storage on Phenylalanine and Tyrosine Measurements in Whole-Blood Samples, Martin Beck, Arend Bökenkamp,* Nicolas Liappis, and Michael J. Lentze (The Children’s Hospital, Medical Center of Bonn University, D-53113 Bonn, Germany; * address correspondence to this author at: Universitätskinderklinik Bonn, Adenauerallee 119, D-53113 Bonn, Germany; fax 49-228-287-3444, e-mail a.boekenkamp@uni-bonn.de)

With an incidence of 1 in 6600 newborns, phenylketonuria (PKU) is among the most common inborn errors of metabolism. PKU is caused by a deficiency of hepatic phenylalanine hydroxylase (1). The increase in the blood Phe concentration leads to permanent structural damage of the central nervous system as a result of disturbed myelination and neurotransmitter deficiency (2). If plasma Phe concentrations are normalized by a low-protein diet with supplementation of essential amino acids before 3 weeks of age, irreversible mental retardation is prevented (2). Still, strict metabolic control is mandatory throughout childhood (2) and perhaps into adult life (3). This is achieved by regular measurement of blood Phe and Tyr concentrations. Tyr is monitored because Phe hydroxylase deficiency renders it an essential amino acid in PKU. Recommendations for the duration and intensity of dietary control are not uniform (2); likewise, there is some variation in the local practices of PKU monitoring. The current guidelines of the German “Arbeitsgemeinschaft für Pädiatrische Stoffwechselkrankungen” (The German Working Group for Metabolic Diseases) set a target range for Phe concentrations of 40–250 μmol/L, at least until age 10 (≥900 μmol/L is acceptable during adolescence) (4). These recommendations were based on data from samples that were analyzed immediately after sampling (Udo Wendel, University Children’s Hospital, Düsseldorf, Germany, personal communication). Pregnant women with even mild hyperphenylalaninemia also require strict dietary control of Phe concentrations to prevent PKU-induced fetopathy (1).

To ensure optimal metabolic control, patients are monitored in specialized clinics where dietary Phe intake recommendations are adjusted according to weekly to monthly Phe and Tyr measurements. To reduce the inconvenience of regular outpatient visits, most German metabolic centers (like centers in other countries) have established methods that allow patients to have capillary samples collected at home and mailed to the laboratory as whole blood (5). Postal transfer of whole-blood samples takes 24–48 h. Storage of whole blood for 6 h at 20 °C has been shown not to significantly affect the recovery of Phe and Tyr (6). To our knowledge, a delay of 48 h until sample preparation, which is introduced by the mailing process, has not been formally evaluated. We therefore studied the effect of delayed sample preparation on Phe and Tyr serum concentrations from whole blood.

Forty-nine blood samples from 35 PKU patients (11.9 ± 10.1 years of age) were obtained by venipuncture during routine monitoring at the metabolic unit of Bonn University Children’s Hospital. Blood was collected into tubes that contained bead-activating coagulation (Serum Monovette; Sarstedt). After informed consent, Phe and Tyr were measured immediately in one aliquot (Pheearly, Tyrearly), whereas the second aliquot was stored as whole blood at room temperature for 48 h until amino acid analysis (Phelate, Tyrlate), thus simulating the mailing process. Serum sample preparation included centrifugation for 10 min at 500g and deproteinization with 50 g/L sulfosalicylic acid (1:1 by volume). Amino acid analysis was performed by ion-exchange chromatography using a

References