Metabolomics Application in Maternal-Fetal Medicine

Vassilios Fanos, Luigi Atzori, Karina Makarenko, Gian Benedetto Melis, and Enrico Ferrazzi

1 NICU, Puericulture Institute and Neonatal Section, AOU and University of Cagliari, 09124 Cagliari, Italy
2 Department of Biomedical Science, University of Cagliari, 09124 Cagliari, Italy
3 Department of Woman, Mother and Neonate, Buzzi Hospital, Biomedical and Clinical Sciences School of Medicine, University of Milan, 20122 Milan, Italy
4 Department of Obstetrics and Gynecology and Human Reproduction Physiopathology, University of Cagliari, 09124 Cagliari, Italy

Correspondence should be addressed to Enrico Ferrazzi; enrico.ferrazzi@unimi.it

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1. Introduction

Metabolomics (or metabonomics) is the youngest and possibly the most promising among “omics technologies.” It represents a holistic approach to the metabolites that constitute a cell, tissue, or organism [1]. It is able to provide a phenotypic fingerprinting of a cell, tissue, or organism by virtue of its ability to measure multiple metabolites directly from complex biological systems. Any molecule less than 1 kDa in mass can be sorted out by metabolomics technology as a single final product of active/inactive genes in a given condition (genome), its activation (mRNA transcriptome), the setup of enzymatic machineries (proteome), and their actual biological processes.

Metabolomic studies in pregnancy are not a novel concept. Since the 1960s, the role of specific single metabolites in the dynamic interactions among fetus, placenta, and pregnant woman had been reported in various studies [2–6]. However, metabolomics in pregnancy, under present high-throughput technologies, is still an “embryonic” science: in 2009, the first review on this topic had been published based on limited data [1], the state of art situation has not changed much since then except for the fact that recent studies prove again and again the immense possibilities of this technology in the complex field of pregnancy where two partner organisms are interacting for mutual benefit [5, 7–11].

2. The Metabolomic Approach

The metabolomic approach consists of two sequential phases. The analytical phase is designed to profile all low molecular weight metabolites in a given biological specimen to generate an all-inclusive spectrum. Possible sources of biological tissues that can be exploited in pregnancy are both from the mother (plasma, urine, vaginal fluids, milk), the fetus (amniotic fluid, umbilical cord blood), and the newborn (plasma, urine, placenta, saliva, other fluids). Different technologies might be generally adopted: nuclear magnetic resonance (NMR) spectroscopy, gas chromatography-mass spectrometry (GC-MS), and liquid chromatography-mass spectrometry
Based on the VIP plot were notable for their differences late versus early-onset preeclampsia groups. Two metabolites on early onset preeclampsia proved a strong separation of prediction. A comparison with database of the previous study 60% sensitivity at 96.6% specificity for late preeclampsia. A parsimonious genetic computing model proved yielded gestation to assess the possible prediction of late preeclampsia only. Unfortunately, the lack of consistent classification of hypertensive disorder in pregnancy as regards the relationship between early onset preeclampsia and late onset preeclampsia and placental vascular damage [17] might impact adversely the correlations observed in the metabolome in cases with and cases without early placental vascular damage and fetal growth restriction (FGR) [9].

Very recently Bahado-Singh et al. [7, 8] published 2 papers focusing on early onset and late onset preeclampsia, selected by the calendar based definition of early and late preeclampsia and by evidence of placental damage in early onset preeclampsia. NMR-based metabolomic analysis was performed on first-trimester maternal serum at 11–13 weeks of gestation to assess the possible prediction of late preeclampsia. A parsimonious genetic computing model proved yielded 60% sensitivity at 96.6% specificity for late preeclampsia prediction. A comparison with database of the previous study on early onset preeclampsia proved a strong separation of late-versus early-onset preeclampsia groups. Two metabolites based on the VIP plot were notable for their differences between preeclamptic and normal pregnant women: glycerol and carnitine. Glycerol and carnitine are both important for lipid metabolism and mitochondrial energy productions based on lipids. More significantly carnitine inhibits oxidative stress. All these metabolic pathways are overexpressed in metabolic syndrome and its acute expression in late preeclampsia.

3. Metabolomics in Plasma of Pregnant Women

To the best of our knowledge, metabolomic analysis performed on plasma of pregnant women had been reported in eight studies [2, 7, 8, 18, 26–29] addressing different pregnancy complications. Plasma samples were analyzed with NMR or LC-MS. Main results of these studies are reported in Table 1.

Lower plasma betaine and trimethylamine-N-oxide concentrations [18] were observed in maternal plasma in case of fetal malformation. Additional different significant findings were reported for amino acids involved in gluconeogenesis, for cis-aconitate, acetone, 3-hydroxybutyrylic, and hypoxanthine. These data suggest that the malformed fetuses demand enhanced gluconeogenesis and tricarboxylic acids cycle (Krebs Cycle), possibly due to hypoxic metabolism. Significant differences were observed also between normal pregnancies and cases which would later develop gestational diabetes. Changes in 3-hydroxyisovalerate and 2-hydroxyisobutyrate were observed, suggesting early changes in biotin status and altered aminoacid and/or gut metabolism. Preeclampsia (PE) had been a major focus in metabolomics technologies applied to maternal-fetal medicine. Both Kenny et al. [26] and Odibo et al. [27] focused their studies on predicting preeclampsia. Kenny’s work was mainly focused on late preeclampsia of maternogenic origin, as we can argue from the limited prevalence of small for gestational age (SGA) newborns in his cohort, and early and late PE were not objects of a different analysis. Odibo in a slightly smaller sample observed a lower detection rate for early onset preeclampsia only. Unfortunately, the lack of consistent classification of hypertensive disorder in pregnancy as regards the relationship between early onset preeclampsia and late onset preeclampsia and placental vascular damage [17] might impact adversely the correlations observed in the metabolome in cases with and cases without early placental vascular damage and fetal growth restriction (FGR) [9].

In a recent paper by Diaz and coworkers [18], maternal plasma and urine were collected and analysed with NMR. Metabolic alterations observed in maternal biofluids in pregnancies with fetal malformations, chromosomal disorders, GDM, PPROM, and preterm delivery suggest potential use of metabolomics in identifying women at risk of prenatal and maternal pregnancy-related disorders. The study by Dessi and coworkers on neonatal urine demonstrated the potential of metabolomics in identifying FGR foetuses and contributing to their clinical management [34] with metabolomics analysis of neonatal urine being another promising field for further investigation [19, 20, 35]. In a preliminary study by our group, a metabolomic approach on maternal urine was used for the diagnosis of labor, suggesting that it is possible to anticipate the timing of delivery [36].

4. Metabolomics in Urine of Pregnant Women and Neonates

Table 2 reports the relevant methodology, results, and conclusions of five studies investigation metabolomics findings in urine of pregnant women and neonates [30–34]. In pregnant women with fetuses affected by somatic abnormalities metabolomics findings confirmed enhanced plasmatic concentration of glucogenic amino acids as well as an increase in hypoxanthine concentration and tricarboxylic acid cycle suggesting ATP degradation and fetal stress.

A possible role of methionine, phenylalanine, histidine, and hexose (possibly glucose) changes in preterm delivery has been suggested.

Inositol phosphoglycan P type (P-IPG) has been linked to insulin resistance, thus making its raise an important marker in Gestational Diabetes Mellitus (GDM) evaluation, fetal growth alterations, and preeclampsia prediction.

Generic markers of prenatal disorders, such as preeclampsia, hypoxia, chromosomal disorders, or prediagnostic GDM, are N-methyl-2-pyridine-5-carboxamide and N-methylnicotinamide (products of abnormal nucleotide metabolism) most likely increasing due to the effects of stress.

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5. Metabolomics in Amniotic Fluid

Metabolomic analysis of amniotic fluid, mainly determined with NMR and MS (ULPC and LC), has been reported by several studies addressing different pregnancy complications [3, 30, 37–41]. Main results of these studies are presented in Table 3.

Amniotic fluid biomarkers seemed to have the best predictive value for malformed fetuses; in this case, lower glucose and higher free lactate levels indicate that energy...
Table 1: Methodology, findings, and main conclusions derived from metabolomics studies on maternal plasma.

| Population | Gestational age at examination | Metabolomic analysis | Main differences in abnormal pregnancies | Significance/take-home messages | Reference | Author, year |
|------------|--------------------------------|----------------------|------------------------------------------|---------------------------------|-----------|-------------|
| 20 normal pregnancies | Matched | Metabolomic analysis | Gluconeogenetic amino acids, cis-aconitate, acetone, 3-hydroxyisoburitic, hypoxanthine | Pregnant women with malformed fetus showed enhanced gluconeogenesis and tricarboxylic acids cycle | [18] | Diaz et al., 2011 |
| 27 fetal malformations | At diagnosis | NMR | 3-Hydroxyisovalerate, 2-hydroxyisoburitic acid | Pregnant women who later develop GDM showed early changes in biotin status and altered amino acid and/or gut metabolism | [18] | Diaz et al., 2011 |
| 23 chromosomal disorders | At diagnosis | NMR | Hystidine, tyrosine, phenylalanine | Diagnosis of preeclampsia was possible by metabolomics | [28] | Turner et al., 2008 |
| 14 prediagnostic GDM | Before positive OGTT | UPLC-MS | 40 metabolites (detection rate 71%, 5% false positive) | Metabolomics might predict preeclampsia, early and late PE not analysed separately | [26] | Kenny et al., 2010 |
| 18 pPROM | At diagnosis | NMR | Citrate, glycerol, hydroxyisovalerate, and methionine and uterine Doppler abnormal PI (detection rate of 82.6%, at an FPR of 1.6%) | Metabolomics might predict early onset PE | [27] | Odibo et al., 2011 |
| 11 normotensive pregnancies versus 11 preeclamptic pregnancies | n.r. | NMR | 40 acylcarnitine species and 32 amino acids (AUC-ROC = 0.85) | Metabolomics might predict early onset PE | [27] | Odibo et al., 2011 |
| 60 cases of PE versus matched controls | First trimester | UPLC-MS | 17 metabolites | Metabolomics might predict late onset PE | [28] | Turner et al., 2008 |
| 30 cases of early PE versus 60 controls | First trimester | NMR | 17 metabolites | Metabolomics might predict late onset PE | [7] | Bahado-Singh et al., 2012 |
| 30 cases of late PE versus 119 controls | First trimester | NMR | 17 metabolites | Metabolomics might predict late onset PE | [8] | Bahado-Singh et al., 2013 |
| Pregnant women who subsequently delivered an SGA baby | First trimester | UPLC-MS | 19 metabolites (OR = 44; AUC-ROC = 0.90) | Metabolomics might predict SGA | [29] | Horgan et al., 2011 |

GDM: gestational diabetes mellitus; pPROM: preterm premature rupture of membranes; OGTT: oral glucose tolerance test; NMR: nuclear magnetic resonance spectroscopy; PE: preeclampsia; UPLC-MS: ultra performance liquid chromatography-mass spectrometry; LS/MS: liquid chromatography-mass spectrometry; SGA: small for gestational age.

Production is being conducted preferentially through glycolysis under anaerobic (hypoxic) conditions and a reduced use of mitochondrial respiratory chain pathway (reflected by succinate increase). Due to hypoxia less glucose is available to other tissues and in order to replenish the glucose level an augmented glucose production via gluconeogenesis is observed (with a resulting decrease in gluconeogenic amino acids levels).

Higher glutamine level, glutamine/glutamate ratios, increase in glycoprotein PI, and lower urea level reflect kidney disorders/underdevelopment. Changes in glycine, serine, α oxoisovalerate, leucine, and ascorbate suggest a disturbance in amino acid biosynthesis. Amniotic fluid changes described above are characteristic for the whole group of malformations indicating changes in glycolysis, gluconeogenesis, and kidney underdevelopment.

Data regarding GDM (increase of glucose level consistent with previously reported insulin increase; slight reduction of several amino acids, creatinine, acetate, formate, glycerophosphocholine) suggest changes in amino acids biosynthesis, higher demand for protein, and changes in lipid metabolism, and renal function.
Table 2: Methodology, findings, and main conclusions derived from metabolomics studies on maternal and neonatal urine.

| Population study | Gestational age at examination | Metabolomic analysis | Main results | Significance/take-home messages | Reference | Author, year |
|------------------|--------------------------------|----------------------|--------------|---------------------------------|-----------|--------------|
| 48 GDM cases versus 23 controls | Third trimester | MS | Increased urinary excretion of P-IPG, positive correlation with blood glucose | Metabolomic identification of P-IPG is a potential marker of insulin resistance, may predict fetal growth alterations in GDM patients | [33] | Scioscia et al., 2007 |
| 9 PE cases versus 84 controls | First trimester | Elisa-based assay | Rapid raise of P-IPG (sensitivity and specificity of 88.9% and 62.7%, resp.) | Metabolomics by multiple assessment of samples might predict preeclampsia | [32] | Paine et al., 2010 |
| 3083 pregnancies positive to the screening of Smith-Lemli-Opitz syndrome | Second trimester | GC/MS | 16α-OH-DHEA, urine total estriol. Fetal steroid sulfatase deficiency 98% detection rate, 95% CI 92–99 | Maternal urine steroids is effective in detecting STSD. Possible use for diagnosing ADHD and CGDS (common in association with STSD) | [31] | Marcos et al., 2009 |
| 75 pregnancies (13 healthy) 13 fetal malformations 20 predisposition to GDM 6 preterm delivery | Second trimester | UPLC-MS NMR | Hippurate, amino acids No relevant changes Methionine, phenylalanine, histidine, hexose (possibly glucose) | Potential of the tandem use of MS and NMR for metabolomics studies of urine and amniotic fluid in pregnant women | [30] | Graça et al., 2012 |
| 26 preterm FGR versus 30 preterm appropriate for gestational age | Neonates | H NMR | Myoinositol, sarcosine, creatine, and creatinine | Metabolomics might identify FGR and contribute to the clinical management of FGR neonates | [34] | Dessi et al., 2011 |

GDM: gestational diabetes mellitus; MS: mass spectrometry; P-IPG: inositol phosphoglycan P type; PE: preeclampsia; GC/MS: gas chromatography-mass spectrometry; STSD: steroid sulfatase deficiency; ADHD: attention deficit-hyperactivity disorder; CGDS: contiguous gene deletion syndrome; UPLC-MS: ultra performance liquid chromatography-mass spectrometry; NMR: nuclear magnetic resonance spectroscopy; 1H NMR: proton nuclear magnetic resonance spectroscopy; FGR: fetal growth restriction.

A derangement of amino acid metabolism due to observed increase in succinic acid and glutamine concentration and significantly lower concentrations of creatine and creatinine has been suggested for the spina bifida cases analyzed with metabolomics techniques.

The changes observed in the amniotic fluid related to preterm delivery include increase of allantoine (a marker of oxidative stress) and a decrease of myoinositol that promotes fetal lung maturation.

Two recent cross-sectional metabolomics studies reported the importance of the amniotic fluid metabolic signature for preterm labor (PTL) with or without intra-amniotic infection or inflammation (IAI) [3, 41]. Amniotic fluid biomarkers were able to discriminate 3 groups based on the pregnancy outcome [41]: (1) patients with PTL who delivered at term, (2) patients with PTL without IAI who delivered preterm, and (3) patients PTL with IAI who delivered preterm (characterized by an increase in amino acid metabolites). A reduction of carbohydrates in the amniotic fluid was associated with preterm delivery (with or without IAI). Methyladenine and diaminopimelic acid, components of bacterial processes and bacterial wall, respectively, were the most significant biomarkers in this clinical condition.

These studies show the potential of human amniotic fluid metabolome analysis as the basis for the development of rapid tests to differentiate between patients at risk of impending preterm birth (with or without infection/inflammation) and patients who will deliver at term.

6. Metabolomics in Placenta

Metabolic analysis of placenta has been reported in several scientific papers [4–6, 10, 11]. The results of these studies are presented in Table 4.
| Population study | Gestational age at examination | Metabolomic analysis | Main results | Significance/take-home messages | Reference | Author, year |
|------------------|--------------------------------|----------------------|--------------|---------------------------------|-----------|-------------|
| 14 pregnancies with spina bifida fetuses versus 18 controls | Second and third trimester (at amniocentesis and cesarean section) | $^1$H NMR | Alterations in the concentrations of succinic acid, glutamine, creatine, creatinine | Metabolomics can point out derangements in amino acids metabolism in fetuses with spina bifida and help to diagnose it | [40] | Groenen et al., 2004 |
| Healthy pregnancies | Second trimester (16-17 ws) | RP-LC, MS $^1$H NMR | 60 metabolites | Extended metabolite database for detecting biomarkers of pregnancy disorders | [39] | Graça et al., 2008 |
| 51 healthy pregnancies versus 12 fetal malformations | Second trimester (15–24 ws) | NMR | Glucose, free lactate levels, succinate, gluconeogenic amino acids, glutamine, glycine, urea, glutamine/glutamate ratios | Metabolomics may help detecting malformed fetuses (due to the changes in glycolysis and gluconeogenesis, and their kidney underdevelopment) | [38] | Graça et al., 2009 |
| 82 normal pregnancies 27 fetal malformation | | | | | |
| 27 prediagnostic GDM | Second trimester | $^1$H NMR | 22 metabolites, inclusion of new metabolites: ascorbate, α oxoisovalerate, creatinine, isoleucine, serine, threonine Glucose, amino acids, organic acids, creatinine, glycerophosphocholine (GPC) Allantoin, alanine, citrate, myo-inositol Minor metabolic profile changes no relevant changes | Metabolomics might be able to identify biomarkers of prenatal disorders | [37] | Graça et al., 2010 |
| 12 preterm delivery | | | | | |
| 34 PROM 10 chromosomopathies | | | | | |
| 2 studies including 16 + 40 PTL → at term 19 + 33 PTL without IAI 20 + 40 PTL with IAI | Second and third trimester (22–35 ws) | LC/MS GC/MS | Carbohydrates, amino acids, presence of xenobiotic compounds (salicylamide, bacterial products) Classification of patients at risk for PTL with 88.5% accuracy | Metabolic profiling of amniotic fluid can be used to assess the risk of preterm delivery with or without IAI | [41] | Romero et al., 2010 |
| 36 controls 29 fetal malformations 37 prediagnostic GDM 34 preterm delivery | Second trimester | UPLC-MS NMR | New results: changes in pyroglutamate, hippurate No relevant changes | Potential of the tandem use of MS and NMR for metabolomics studies of urine and amniotic fluid in pregnant women | [30] | Graça et al., 2012 |

$^1$H NMR: proton nuclear magnetic resonance spectroscopy; RP-LC: reverse phase liquid chromatography; MS: mass spectrometry; GDM: gestational diabetes mellitus; PROM: premature rupture of membranes; PTL: preterm labor; IAI: intraamniotic infection/inflammation; LC/MS: liquid chromatography/mass spectrometry; GC/MS: gas chromatography/mass spectrometry; UPLC-MS: ultra performance liquid chromatography-mass spectrometry; NMR: nuclear magnetic resonance spectroscopy.
| Population study         | Material                  | Metabolomic analysis | Main results                                                                                                                                                                                                                                                                                                                                 | Significance/take-home messages                                                                 | Reference            | Author, year |
|-------------------------|---------------------------|----------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------|----------------------|--------------|
| 9 SGA cases versus 8 controls | Placenta villous explants | UPLC-MS              | 574 metabolites were significantly different between SGA and controls; 49% of metabolites of interest were the same for SGA explant cultured under hypoxic conditions and controls cultured under normoxic conditions. Changes in phospholipids, essential amino acids (tryptophan, methionine, and phenylalanine) concentrations. | Metabolomics might predict SGA                                                                | [11] Horgan et al., 2010 |              |
| 11 uncomplicated term pregnancies | Placenta villous explants | GC-MS                | Cultured in 1%, 6%, and 20% oxygen. Differences in 2-deoxyribose, threitol/erythritol, hexadecanoic acid.                                                                                                           | Metabolomics can be applied to placenta studies and could help in detecting hypoxia          | [10] Heazell et al., 2008 |              |
| 6 cases of preeclampsia versus 6 controls | Villous trophoblast | UPLC-MS              | 47 metabolites in preeclampsia-derived media cultured under normoxic conditions showed similarities to that of uncomplicated pregnancies cultured under hypoxic conditions. Alterations in glutamate and glutamine, leukotrienes and prostaglandins, kynurenine metabolism. | Metabolomics might predict preeclampsia of placental origin developed due to hypoxia         | [4] Dunn et al., 2009 |              |
| 8 cases of labor/Cesarean section at 3100 m versus 8 controls with labor/Cesarean section delivery at sea level | Placenta              | $^1$H NMR, $^{31}$P NMR | At sea level: metabolic markers of oxidative stress, increased glycolysis, elevated cholesterol, and free amino acids. At 3100 m: metabolic profiles with adaptation to chronic hypoxia, decreased reliance on anaerobic glycolysis; presence of concentrations of stored energy potential (phosphocreatine), antioxidants (taurine, inositol), and low free amino acid concentrations. | Metabolomics might help identify subjects under hypoxic stress (chronic hypoxic preconditioning state versus acute ischemic/hypoxic insult) | [6] Tissot van Patot et al., 2010 |              |

SGA: small for gestational age; UPLC-MS: ultra performance liquid chromatography-mass spectrometry; GC-MS: gas chromatography-mass SPECTROMETRY; $^1$H NMR: proton nuclear magnetic resonance spectroscopy; $^{31}$P NMR: phosphorus nuclear magnetic resonance spectroscopy.

Dunn and coworkers [4] studied the metabolomic response of the normal and preeclamptic placental tissue to different oxygen levels. Placental tissue from uncomplicated pregnancies cultured in 1% oxygen (hypoxia) showed metabolic similarities to explants from late-onset preeclampsia pregnancies cultured at 6% oxygen (normoxia) thus suggesting an important role of hypoxia in preeclampsia development. Specific metabolic alterations included lipids, glutamate, glutamine, and metabolites related to tryptophan, leukotrienes, and prostaglandins. These metabolic changes were confirmed by Horgan and coworkers [11] that in a similar study compared placental features seen in small for gestational age (SGA) cases versus controls. Placental tissue from both groups was cultured in 1%, 6%, and 20% oxygen. There was a significant difference in more than 500 metabolites between SGA and normal pregnancies at different oxygen tensions, while 49% of metabolites of interest were similar under normoxic conditions suggesting in vivo adjustment to hypoxia by SGA fetus, with a further consideration that normal oxygen level could worsen metabolic dysfunction of the placenta of a SGA fetus. This study reinforces the role of hypoxia and oxidative stress in preeclampsia and SGA. The role of chronic hypoxia was also studied by Tissot van Patot and colleagues [6], who reported the placenta at 3100 m sea level, naturally present higher concentrations of stored energy potential, antioxidants, and lower free amino acid concentrations.
concentrations, with better resistance to hypoxia and less evident metabolic stress response, and their results suggesting adaptation to chronic hypoxia in placenta at high altitude.

7. Metabolomics in Vaginal Secretion

A pilot mass spectrometry metabolic study of women who had a spontaneous preterm birth compared to controls was conducted using the uterine cervical length as a selective factor. Noninvasive collection of cervicovaginal secretions from 15 women (women with a short cervix who had a spontaneous preterm birth compared to a control cohort with a short cervix and with a long cervix) was performed, and 17 markers of interest were identified. This particular metabolomic approach may help in the management of women at high-risk for spontaneous preterm birth [16].

8. Metabolomics in Cord Blood

Metabolic analysis of cord blood has been carried out on samples collected from low birth weight (LBW) newborns, very low birth weight (VLBW) newborns, newborns diagnosed with FGR, SGA during pregnancy, newborns with asphyxia, and hypoxic ischemic encephalopathy. The results were reported in five studies [29, 42–45]. In the paper by Ivorra and coworkers [42] seven metabolites were able to discriminate a specific LBW metabolome, with a significant positive correlation between birth weight and proline, glutamine, free choline and negative correlation with citrulline and phenylalanine levels. These findings demonstrate the differences in the anabolic rate of the LBW newborns and the possible consequences that these metabolites’ modifications can exercise on the newborn’s health (e.g., maternal choline and its metabolite betaine supply modifying fetal histone and DNA methylation). It is also noteworthy that these changes were observed in the newborns only and not in their mothers, suggesting possible involvement of impaired placental transport of amino acids. Metabolic findings described above reveal the potential of metabolomics in identifying the population at risk. Tea and coworkers [43], analysing the cord blood from VLBW infants, demonstrated that a number of metabolites (glucose, acetate, lipids, pyruvate, glutamine, valine, threonine,) vary depending on gestational age at delivery. In the study by Favretto and coworkers [44], FGR presented changes in 22 metabolites with the most evident modifications in the following aminoacids composition: phenylalanine, tryptophan, and glutamate with 91–100% sensitivity and 85–89% specificity. Cut-off values of metabolomic analysis for the future of the medicine, in this case producing a robust predictive model for detecting encephalopathy in the newborns at 24 hours of life, in order to be able to provide the best timely treatment for the young patient is that of Walsh and coworkers [45]. This study found 29 metabolites among amino acids, acylcarnitines and glycerophospholipids to be significantly different in neonates with hypoxic ischaemic encephalopathy (HIE) versus controls or asphyxia versus controls groups, with 5 metabolites (2 acylcarnitines, 1 glycerophospholipid and 2 aminoacids) clearly delineating the severity of asphyxia and the degree of HIE.

All these data suggest that metabolomics is a noninvasive promising novel way of investigation, with the goal of understanding better the pathogenesis of different diseases, developing screening tests and helping in clinical management of the patients.

9. Conclusions

Metabolomics appears to be one of the most promising novel tools in obstetrics, among other important “omics”. A recently published paper was entitled “Can biofluids metabolic profiling help us to improve health care during pregnancy?” [46]. The answer is “probably yes” if all these preliminary data on the potential usefulness of biofluids metabolomics in perinatal disorders are further investigated, single diseases are targeted and results are confirmed.

Through the metabolome we can observe the enormous and complex interactions between mother, placenta, and fetus. This high throughput technology might allow us to identify key role nodes for prediction, diagnosis, and monitoring of different obstetrics conditions.

References

[1] R. P. Horgan, O. H. Clancy, J. E. Myers, and P. N. Baker, “An overview of proteomic and metabolomic technologies and their application to pregnancy research,” BJOG, vol. 116, no. 2, pp. 173–181, 2009.
[2] P. C. Woodham, T. O’Connell, J. Grimes et al., “Metabolomics to predict severe preeclampsia in early pregnancy,” American Journal of Obstetrics and Gynecology, vol. 206, no. 1, supplement, p. S348, 2012.
[3] C. W. Beecher, “Metabolomic studies at the start and end of the life cycle,” Clinical Biochemistry, vol. 44, no. 7, pp. 518–519, 2011.
[4] W. B. Dunn, M. Brown, S. A. Worton et al., “Changes in the Metabolic Footprint of Placental Explant-Conditioned Culture Medium Identifies Metabolic Disturbances Related to Hypoxia and Pre-Eclampsia,” Placenta, vol. 30, no. 11, pp. 974–980, 2009.
[5] A. E. P. Heazell, M. Brown, S. A. Worton, and W. B. Dunn, “Review: the effects of oxygen on normal and pre-eclamptic placental tissue—insights from metabolomics,” Placenta, vol. 32, supplement 2, pp. S19–S124, 2011.
[6] M. C. Tissot van Patot, A. J. Murray, V. Beckey et al., “Human placental metabolic adaptation to chronic hypoxia, high altitude: hypoxic preconditioning,” American Journal of Physiology—Regulatory Integrative and Comparative Physiology, vol. 298, no. 1, pp. R166–R172, 2010.
[7] R. O. Bahado-Singh, R. Akolekar, R. Mandal et al., “Metabolomics and first-trimester prediction of early-onset preeclampsia,” Journal of Maternal-Fetal and Neonatal Medicine, vol. 25, no. 10, pp. 1840–1847, 2012.

[8] R. O. Bahado-Singh, R. Akolekar, R. Mandal et al., “First-trimester metabolomic detection of late-onset preeclampsia,” American Journal of Obstetrics and Gynecology, vol. 208, no. 1, pp. 58.e1–58.e7, 2013.

[9] E. Ferrazzi, T. Stampalia, and J. E. Au pont, “The evidence for lateonset preeclampsia as a maternogenic disease of pregnancy,” Fetal and Maternal Medicine Review, vol. 24, no. 1, pp. 18–31, 2013.

[10] A. E. P. Heazell, M. Brown, W. B. Dunn et al., “Analysis of the Metabolic Footprint and Tissue Metabolome of Placental Villous Explants Cultured at Different Oxygen Tensions Reveals Novel Redox Biomarkers,” Placenta, vol. 29, no. 8, pp. 691–698, 2008.

[11] R. P. Horgan, D. I. Broadhurst, W. B. Dunn et al., “Changes in the metabolic footprint of placental explant-conditioned medium cultured in different oxygen tensions from placentas of small for gestational age and normal pregnancies,” Placenta, vol. 31, no. 10, pp. 893–901, 2010.

[12] R. Antonucci, L. Atzori, L. Barberini, and V. Fanos, “Metabolomics: the new clinical chemistry for personalized neonatal medicine,” Minerva pediatrica, vol. 62, no. 3, supplement 1, pp. 145–148, 2010.

[13] T. J. Athersuch, “The role of metabolomics in characterizing the human exposome,” Bioanalysis, vol. 4, no. 18, pp. 2207–2212, 2012.

[14] L. Atzori, R. Antonucci, L. Barberini, J. L. Griffin, and V. Fanos, “Metabolomics: a new tool for the neonatologist,” Journal of Maternal-Fetal and Neonatal Medicine, vol. 22, no. 3, pp. 50–53, 2009.

[15] L. Atzori, L. Barberini, M. L. Santoru, R. Antonucci, and V. Fanos, “Metabolomics explained to perinatologists and pediatricians,” Journal of Maternal-Fetal and Neonatal Medicine, vol. 25, supplement 5, pp. 10–12, 2012.

[16] C. Auray-Blais, E. Raiche, R. Gagnon, M. Berthiaume, and J. C. Pasquier, “Metabolomics and preterm birth: what biomarkers in cervicovaginal secretions are predictive of high-risk pregnant women?” International Journal of Mass Spectrometry, vol. 307, no. 1–3, pp. 33–38, 2011.

[17] G. J. Burton, A. W. Woods, E. Jauniaux, and J. C. P. Kingdom, “Rheological and Physiological Consequences of Conversion of the Maternal Spiral Arteries for Uteroplacental Blood Flow during Human Pregnancy,” Placenta, vol. 30, no. 6, pp. 473–482, 2009.

[18] S. O. Diaz, J. Pinto, G. Graça et al., “Metabolic biomarkers of prenatal disorders: an exploratory NMR metabolomics study of second trimester maternal urine and blood plasma,” Journal of Proteome Research, vol. 10, no. 8, pp. 3732–3742, 2011.

[19] V. Fanos, R. Antonucci, L. Barberini, and L. Atzori, “Urinary metabolomics in the newborn and infants,” Advances in Clinical Chemistry, vol. 58, pp. 193–223, 2012.

[20] V. Fanos, R. Antonucci, L. Barberini, A. Noto, and L. Atzori, “Clinical application of metabolomics in neonatology,” Journal of Maternal-Fetal and Neonatal Medicine, supplement 1, pp. 104–109, 2012.

[21] V. Fanos, R. Antonucci, M. Zaffanello, and M. Mussap, “Neonatal drug induced nephrotoxicity: old and next generation biomarkers for early detection and management of neonatal drug-induced nephrotoxicity, with special emphasis on uNGAL and on metabolomics,” Current Medical Chemistry, vol. 19, no. 27, pp. 4595–4605, 2012.

[22] V. Fanos, L. Atzori, A. Dessi, E. D’Aloja, G. Finco, and G. Faa, “The kidney in post-asphyctic syndrome: state of the art,” in Developmental Nephrology: From Embryology To Metabolomics, V. Fanos, R. L. Chevalier, G. Faa, and L. Cataldi, Eds., Hygeia Press, 2011.

[23] V. Fanos, L. Barberini, R. Antonucci, and L. Atzori, “Metabolomics in neonatology and pediatrics,” Clinical Biochemistry, vol. 44, no. 7, pp. 452–454, 2011.

[24] V. Fanos, L. Barberini, R. Antonucci, and L. Atzori, “Pharmacometabolomics in Neonatology: is it a Dream or a Fact?” Current Pharmaceutical Design, vol. 18, no. 21, pp. 2996–3006, 2012.

[25] V. Fanos, M. Mussap, A. Noto, and L. Atzori, “Metabolomics in perinatology: where are we now?” Acta Medica Portuguesa, supplement 2, pp. 117–120, 2012.

[26] L. C. Kenny, D. I. Broadhurst, W. Dunn et al., “Robust early pregnancy prediction of later preeclampsia using metabolic biomarkers,” Hypertension, vol. 56, no. 4, pp. 741–749, 2010.

[27] A. O. Odibo, K. R. Goetzinger, L. Odibo et al., “First-trimester prediction of preeclampsia using metabolic biomarkers: a discovery phase study,” Prenatal Diagnosis, vol. 31, pp. 990–994, 2011.

[28] E. Turner, J. A. Brewster, N. A. B. Simpson, J. J. Walker, and J. Fisher, “Aromatic amino acid biomarkers of preeclampsia—a nuclear magnetic resonance investigation,” Hypertension in Pregnancy, vol. 27, no. 3, pp. 225–235, 2008.

[29] R. P. Horgan, D. I. Broadhurst, S. K. Walsh et al., “Metabolic profiling uncovers a phenotypic signature of small for gestational age in early pregnancy,” Journal of Proteome Research, vol. 10, no. 8, pp. 3660–3673, 2011.

[30] G. Graça, B. J. Goodfellow, A. S. Barros et al., “UPLC-MS metabolic profiling of second trimester amniotic fluid and maternal urine and comparison with NMR spectral profiling for the identification of pregnancy disorder biomarkers,” Molecular Biosystems, vol. 8, no. 4, pp. 1243–1254, 2012.

[31] J. Marcos, W. Y. Craig, G. E. Palomaki et al., “Maternal urine and serum steroid measurements to identify steroid sulfatase deficiency (STSD) in second trimester pregnancies,” Prenatal Diagnosis, vol. 29, no. 8, pp. 771–780, 2009.

[32] M. A. Paine, M. Scioscia, P. J. Williams, K. Gumaa, C. H. Rodeck, and T. W. Rademacher, “Urinary inositol phosphoglycan P-type as a marker for prediction of preeclampsia and novel implications for the pathophysiology of this disorder,” Hypertension in Pregnancy, vol. 29, no. 4, pp. 375–384, 2010.

[33] M. Scioscia, S. Kunjara, K. Gumaa, P. McLean, C. H. Rodeck, and T. W. Rademacher, “Urinary excretion of inositol phosphoglycan P-type in gestational diabetes mellitus,” Diabetic Medicine, vol. 24, no. 11, pp. 1300–1304, 2007.

[34] A. Dessi, L. Atzori, A. Noto, V. Fanos et al., “Metabolomics in newborns with intrauterine growth retardation (IUGR): urine reveals markers of metabolic syndrome,” Journal of Maternal-Fetal and Neonatal Medicine, vol. 24, supplement 2, pp. 35–39, 2011.

[35] V. Fanos, J. van den Anker, A. Noto, M. Mussap, and L. Atzori, “Metabolomics in neonatology: fact or fiction?” Seminars in Fetal & Neonatal Medicine, vol. 18, no. 1, pp. 3–12, 2013.

[36] A. Meloni, P. Caboni, F. Manconi et al., “Metabolic approach to diagnosis of labour,” Abstract International Congress of Obstetrics, vol. 119, supplement 1, pp. 1–250, 2012.
[37] G. Grac¸a, I. F. Duarte, A. S. Barros et al., "Impact of prenatal disorders on the metabolic profile of second trimester amniotic fluid: a nuclear magnetic resonance metabonomic study," Journal of Proteome Research, vol. 9, no. 11, pp. 6016–6024, 2010.

[38] G. Grac¸a, I. F. Duarte, A. S. Barros et al., "1H NMR based metabonomics of human amniotic fluid for the metabolic characterization of fetus malformations," Journal of Proteome Research, vol. 8, no. 8, pp. 4144–4150, 2009.

[39] G. Grac¸a, I. F. Duarte, B. J. Goodfellow et al., "Metabolite profiling of human amniotic fluid by hyphenated nuclear magnetic resonance spectroscopy," Analytical Chemistry, vol. 80, no. 15, pp. 6085–6092, 2008.

[40] P. M. W. Groenen, U. F. Engelke, R. A. Wevers et al., "High-resolution 1H NMR spectroscopy of amniotic fluids from spina bifida fetuses and controls," European Journal of Obstetrics Gynecology and Reproductive Biology, vol. 112, no. 1, pp. 16–23, 2004.

[41] R. Romero, S. Mazaki-Tovi, E. Vaisbuch et al., "Metabolomics in premature labor: a novel approach to identify patients at risk for preterm delivery," Journal of Maternal-Fetal and Neonatal Medicine, vol. 23, no. 12, pp. 1344–1359, 2010.

[42] C. Ivorra, C. García-Vicent, F. J. Chaves, D. Monleón, J. M. Morales, and E. Lurbe, "Metabonomic profiling in blood from umbilical cords of low birth weight newborns," Journal of Translational Medicine, vol. 10, no. 1, article 142, 2012.

[43] I. Tea, G. le Gall, A. Küster et al., "1H-NMR-based metabolic profiling of maternal and umbilical cord blood indicates altered materno-foetal nutrient exchange in preterm infants," PLoS One, vol. 7, no. 1, article e29947, 2012.

[44] D. Favretto, E. Cosmi, E. Ragazzi et al., "Cord blood metabolomic profiling in intrauterine growth restriction," Analytical and Bioanalytical Chemistry, vol. 402, no. 3, pp. 1109–1121, 2012.

[45] B. H. Walsh, D. I. Broadhurst, R. Mandal et al., "The metabonomic profile of umbilical cord blood in neonatal hypoxic ischaemic encephalopathy," PLoS One, vol. 7, no. 12, article e50520, 2012.

[46] G. Grac¸a, S. Diaz, J. Pinto et al., "Can biofluids Metabolic profiling help us to improve health care during pregnancy?" Spectroscopy, vol. 27, no. 5-6, pp. 515–523, 2012.