Toxicology and Applied Pharmacology 275 (2014) 79–87

Contents lists available at ScienceDirect

Toxicology and Applied Pharmacology

journal homepage: www.elsevier.com/locate/ytaap

Maternal glucocorticoid elevation and associated blood metabonome changes might be involved in metabolic programming of intrauterine growth retardation in rats exposed to caffeine prenatally

Hao Kou [a,1], Yansong Liu [a,1], Gai Liang [a], Jing Huang [a], Jieqiong Hu [a], You-e Yan [a], Xiaojun Li [a], Hong Yu [b], Xiaohua He [b], Baifang Zhang [b], Yuanzhen Zhang [b,d], Jianghua Feng [c,**], Hui Wang [a,b,*]

[a] Department of Pharmacology, Basic Medical School of Wuhan University, Wuhan 430071, China
[b] Hubei Provincial Key Laboratory of Developmentally Originated Diseases, Wuhan 430071, China
[c] Department of Electronic Science, Fujian Provincial Key Laboratory of Plasma and Magnetic Resonance, Xiamen University, Xiamen 361005, China
[d] Center for Reproductive Medicine, Zhongnan Hospital of Wuhan University, Wuhan 430071, China

ARTICLE INFO

Article history:
Received 1 October 2013
Revised 12 January 2014
Accepted 13 January 2014
Available online 23 January 2014

Keywords:
Caffeine
Intrauterine growth retardation
Maternal glucocorticoid
Metabonome
Metabolic programming

ABSTRACT

Our previous studies demonstrated that prenatal caffeine exposure causes intrauterine growth retardation (IUGR), fetuses are over-exposed to high levels of maternal glucocorticoids (GC), and intrauterine metabolic programming and associated metabonome alteration that may be GC-mediated. However, whether maternal metabonomes would be altered and relevant metabolite variations might mediate the development of IUGR remained unknown. In the present studies, we examined the dose- and time-effects of caffeine on maternal metabonome, and tried to clarify the potential roles of maternal GCs and metabonome changes in the metabolic programming of caffeine-induced IUGR. Pregnant rats were treated with caffeine (0, 20, 60 or 180 mg/kg · d) from gestational days (GD) 11 to 20, or 180 mg/kg · d caffeine from GD9. Metabonomes of maternal plasma on GD20 in the dose–effect study and on GD11, 14 and 17 in the time–course study were analyzed by ^{1}H nuclear magnetic resonance spectroscopy, respectively. Caffeine administration reduced maternal weight gains and elevated both maternal and fetal corticosterone (CORT) levels. A negative correlation between maternal/fetal CORT levels and fetal bodyweight was observed. The maternal metabonome alterations included attenuated metabolism of carbohydrates, enhanced lipolysis and protein breakdown, and amino acid accumulation, suggesting GC-associated metabolic effects. GC-associated metabolite variations (α/β-glucoses, high density lipoprotein-cholesterol, β-hydroxybutyrate) were observed early following caffeine administration. In conclusion, prenatal caffeine exposure induced maternal GC elevation and metabonome alteration, and maternal GC and relevant discriminatory metabolites might be involved in the metabolic programming of caffeine-induced IUGR.
© 2014 Elsevier Inc. All rights reserved.

Introduction

Intrauterine growth retardation (IUGR) is defined as the intrauterine events leading to the birth of an infant born weighing 10% or two or more standard deviations less than the mean body weight of normal babies at same gestational age (Resnik, 2002; Valsamakis et al., 2006). The incidence of small for gestational age (SGA) in developed countries (i.e., U.S.) is approximately 10% in which one third are diagnosed as IUGR (Vandenbosche and Kirchner, 1998), while the incidence of IUGR ranges from 6% to 30% in developing countries (Imdad et al., 2011). IUGR is associated with greater risks of developing metabolic syndrome (MS) in adult life (Alisi et al., 2011).

Caffeine is a xanthine alkaloid that is widely present in human environments. Many epidemiological studies have revealed that caffeine ingestion during pregnancy correlates with IUGR (Bakker et al., 2010; Group, 2008), and an excess ingestion of caffeine-contained beverage during childhood is associated with greater susceptibilities to MS (James et al., 2004). Previously, we demonstrated that caffeine exposure before and during gestation leads to reproductive and developmental toxicities in rodents, including IUGR (J. Huang et al., 2012). Meanwhile, IUGR fetuses are exposed to high levels of maternal glucocorticoids (GC), and the intrauterine programming alteration of glucose and lipid

Abbreviations: IUGR, intrauterine growth retardation; MS, metabolic syndrome; GC, glucocorticoid; GD, gestational day; CORT, corticosterone; NMR, nuclear magnetic resonance; CPMG, Carr–Purcell–Meiboom–Gill; PCA, principal component analysis; OPLS-DA, orthogonal projection to latent structure with discriminant analysis; SGA, small for gestation age; HPA, hypothalamus–pituitary–adrenal; VLDL, very low density lipoprotein; HDL-C, high density lipoprotein–cholesterol.

* Correspondence to: H. Wang, Department of Pharmacology, Basic Medical School of Wuhan University, 185 East Lake Road, Wuhan, 430071, Hubei Province, China. Fax: +86 27 68759222.
** Correspondence to: J. Feng, Department of Electronic Science, Fujian Provincial KeyLaboratory of Plasma and Magnetic Resonance, Xiamen University, 422 Si-Ming-Nan-Lu, Xiamen 361005, Fujian Province, China. Fax: +86 592 2189426.
E-mail addresses: wanghui19@whu.edu.cn (H. Wang), jianghua.feng@xmu.edu.cn (J. Feng).
[1] Hao Kou and Yansong Liu have contributed equally to this work.

0041-008X/$ – see front matter © 2014 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.taap.2014.01.007

metabolism that may be GC-mediated (Liu et al., 2012; Xu et al., 2012b). Furthermore, we identified that prenatal caffeine exposure brings about GC-associated blood metabonome alterations in fetal rats (Liu et al., 2012). However, whether the maternal metabonome would be altered and relevant metabolite variations might mediate the development of IUGR remained unknown.

Physiologically, mother and fetus is an integrated biological unit during pregnancy. Fetal growth is a complex process that involves the interaction of mother and fetus by the placenta. An interpenetration of multiple endogenous small molecule metabolites between mother and fetus could be achieved through different placental transport pathways. Metabonomics enables the parallel assessment of the levels of a broad range of metabolites and has been suggested to be particularly valuable for the classification and prediction of pathophysiological states by analyzing the metabolic profiles of biofluids, and for identification of endogenous biomarkers of toxicity (Antonucci et al., 2010). In the present study, we investigated the dose- (0, 20, 60, 180 mg/kg · d on GD20) and time- (GD11, 14, 17 at 180 mg/kg · d) dependent effects of prenatal caffeine exposure on maternal blood metabonome alterations in rats by using ^{1}H nuclear magnetic resonance (NMR)-based metabonomics, and some correlative analyses between maternal/fetal blood corticosterone (CORT) levels and fetal body weights were incorporated into elucidating the potential roles of maternal metabonome on the metabolic programming of IUGR. This study will also contribute to the exploration of candidate biomarker(s) in maternal blood for the early alerting of caffeine-induced IUGR.

Materials and methods

Chemicals and reagents. Caffeine (CAS #58-08-2, >99% purity) and sodium azide (NaN_3, CAS #26628-22-8, >99% purity) were purchased from Sigma-Aldrich Co., Ltd (St Louis, MO, USA). Deuterium oxide (D_2O, >99% purity) was provided by Cambridge Isotope Laboratories Inc. (Andover. MA, USA). Isoflurane was purchased from Baxter Healthcare Co. (Deerfield, IL, USA). Rat CORT enzyme-linked immunosorbent assay (ELISA) kit was obtained from R&D Systems, Inc. (Minneapolis, MN, USA).

Animals and treatments. Specific Pathogen-Free (SPF) Wistar rats weighing 180–220 g (female)/260–300 g (male) were obtained from Experimental Center of Hubei Medical Scientific Academy (No. 2008–0005, Wuhan, China). Animals were allowed to acclimate for at least one week before initiating experimental condition and were fed under controlled conditions (24 ± 2 °C, 12:12-h dark–light cycle), with free access to chow and water. The animal experiments were conducted in the Center for Animal Experiment of Wuhan University (Wuhan, China), which has been accredited by the Association for Assessment and Accreditation of Laboratory Animal Care International (AAALAC International). All experimental procedures are approved by and performed in accordance with the Guidelines for the Care and Use of Laboratory Animals of the Chinese Animal Welfare Committee.

Mating and caffeine administration (i.e., dose and time), as well as sample collections, were performed as described previously (J. Huang et al., 2012; Liu et al., 2012; Xu et al., 2012b). The animal studies were conducted in two independent sections for investigating the dose- and time-dependent effects of prenatal caffeine administration on maternal blood metabonomes. In the dose-effect investigations, the dams were administered 0, 20, 60, or 180 mg/kg · d, intragastrically from GD 11 to 20. Each group comprised 8 dams. According to the dose-conversion factor between humans and rats of 1:6.17, based on body surface area (Reagan-Shaw et al., 2008), the caffeine dose of 20 mg/kg · d in pregnant rats is approximately equivalent to 224 mg/day for a pregnant woman weighing 70 kg. On this basis, the dose of caffeine of 20 mg/kg · d is not unreasonable. We employed higher doses, 60, or 180 mg/kg · d, to establish a model of IUGR for mechanistic studies, despite that they might be up to many times than human daily consumption after the dose conversion. With regard to the dosing time selected for study, the rodents' adrenocortical primordium (the precursor of adrenal cortex) is visible at approximately embryonic day (E) 9 (Bland et al., 2003), and becomes to be an independent tissue for differentiation at around E10.5 (Morohashi, 1997). Hence, GD 9–11 appears to be a critical time window for fetal HPA axis ontogenesis and development, and was therefore selected for initiating prenatal caffeine administration. Just as we described previously (Liu et al., 2012), on GD20, animals were kept in their original cages, and each cage was placed in a separate quiet room for anesthesia (5% isoflurane). After the disappearance of the righting reflex, the animals were sacrificed rapidly by cutting the left carotid artery with a sharp pair of scissors to collect maternal blood. Each feto-placental unit was removed quickly from the uterus. In the time–effects study, two groups ($n = 8$ pregnant rats per group) were set: control and caffeine (180 mg/kg · d) group. Caffeine administrations were performed from GD9 to GD20, and maternal blood samples were collected on GD11, GD14 and GD17 via the *vena caudalis* after local anesthesia with 1% procaine. Blood samples were separated into two parts according to the volume requirement of subsequent detection: one sample was centrifuged to prepare serum for measurements of CORT levels, and the other sample was anticoagulated by addition of heparin, followed by preparation of plasma for NMR analysis.

ELISA assay for serum CORT concentration. Serum (10 μl) was used for detection of serum CORT concentration by ELISA kits following the manufacturer's protocol. The intra-assay and inter-assay coefficients of variation for CORT measurements were 5.0% and 7.2%, respectively.

Sample preparation and ^{1}H NMR spectroscopic analysis. Maternal plasma samples were prepared according to the protocol described in our previous paper (Liu et al., 2012). The plasma samples were analyzed by ^{1}H NMR spectroscopy, and NMR spectra were acquired on a Bruker AVIII-600 spectrometer (Bruker BioSpin, Germany) at 298 K. Standard 1D ^{1}H NMR spectra, including NOESYPR1D, CPMG, and diffusion-edited spectra, were acquired with corresponding pulse sequence described in our previous paper (Liu et al., 2012) with some parameter optimizations. For resonance assignment purposes, standard 2D NMR spectra, including COSY, TOCSY, and J-resolved spectra, also were acquired for selected plasma samples.

Spectral processing and analysis. All free induction decays (FIDs) were multiplied by an exponential function equivalent to a 1 Hz line-broadening factor prior to Fourier transformation. All of the ^{1}H NMR spectra were manually phased and baseline-corrected using Topsin (V3.0, Bruker BioSpin, Germany). Chemical shifts in plasma spectra were referenced to the internal lactate CH_3 resonance at δ1.33.

The processed NMR spectra were automatically data-reduced using AMIX package (V3.9, Bruker BioSpin). The CPMG spectrum over the range of δ9.0–0.5 for each maternal rat plasma sample was reduced to 1600 regions, each 0.005 ppm wide, and the signal intensity in each region was integrated. The regions of water resonance (δ5.2–4.32) and urea resonance (δ6.60–5.40) of CPMG spectra of maternal plasma were removed to eliminate baseline effects of imperfect water and urea signals. For diffusion-edited spectrum of each maternal rat plasma sample, the regions of δ6.0–0.5 excluded water resonance (δ5.17–4.36) were integrated in 0.005 ppm wide. The integrated data were normalized to the total sum of the spectrum before pattern recognition analysis.

Pattern recognition analysis was performed with the software SIMCA-P + (V11.0, Umetric, Sweden) by using a mean-centered approach in order to identify intrinsic trends and obvious outliers (Ciosek et al., 2005). To maximize separation between the samples in pair-wise groups, orthogonal projection to latent structure with discriminant analysis (O-PLS-DA) was further performed by using a unit variance-scaled approach as previously described (Liu et al., 2012). In

Download English Version:

https://daneshyari.com/en/article/2568763

Download Persian Version:

https://daneshyari.com/article/2568763

Daneshyari.com