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The Power of Programming 2016

Developmental Origins of Adiposity and Long-Term Health

October 13–15, 2016, Munich

Abstracts

Guest Editor
B. Koletzko, Munich

Plenary Sessions

II

Complementary Feeding: An Opportunity for Programming Later Health?

Friday 14th October, 09.30–10.15

Mary Fewtrell

UCL Institute of Child Health, London, UK

Complementary feeding (CF) practices could potentially influence later health outcomes by a number of mechanisms including programming effects, but also by lasting influences on food preferences, appetite and eating behaviour. Evidence for an effect of nutritional aspects of CF – timing or content – on later health outcomes still comes largely from observational studies. These suggest, for example, that delaying the introduction of CF until 4 months may protect against later obesity; whilst a high protein intake during CF may be associated with increased later obesity risk, perhaps dependent on the type of protein. However, new data from randomised trials have provided important information on the impact of the age at introduction of allergenic foods and gluten on the development of food allergy and coeliac disease respectively; leading to changes in CF advice.

There is increasing focus on the role of CF practices in shaping taste/flavour preferences and subsequent food choices. For example, exposure to a more varied CF diet, including bitter-tasting vegetables, may be associated with more optimal dietary patterns later in childhood. Parenting behaviour can also influence infant feeding practices and growth; a recent systematic review concluded that the most promising interventions for reducing obesity are those focussing on diet and responsive feeding.

There is thus some evidence that the CF period represents an opportunity for influencing later health outcomes. However CF practices vary markedly between and within countries. Given the complex interplay between nutrition, feeding behaviour and psychological factors during this period, a holistic approach is required.

III

What Is Normal Pre- and Postnatal Growth

Thursday 13th October, 15.30–16.15

Michelle Lampl

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As the body is constructed from the cellular level to the organs that make-up the units of structure and function, tissue composition and metabolic processes unfold. The nuances that determine these outcomes involve cross-talk amongst systems that negotiate energy demands and availability of constituents. This progression is a fundamental embodiment of growth, and programming is a central aspect of the complex network interactions. Details of the processes of normal pre- and postnatal growth are beginning to be described. Mechanisms remain to be discovered. Clarifying the process of normal growth is essential to supporting health, as documented by the substantial evidence of lifetime sequelae following growth perturbations in early life. It is important to clarify that what we think we know is both evidence-based and methodologically sound. Concepts of normal growth drive pediatric interventions, supplementary food formularies, recommendations and feeding decisions. The common image of growth that emerges from growth curve reference charts of size-for-age has been adopted as the representation of normal growth trajectories to be expected among children across development. These smoothed population-level statistical distributions are, problematically, not accurate representations of how growth as a biological process occurs within individuals. Normal pre- and postnatal growth occurs by discontinuous saltatory bursts against a background of stasis. These discrete critical periods organize complex cellular alternatives influencing growth rate, hence size, and the biology of body composition.

IV

Pregnancy Interventions – What Should Our Recommendations Be into the Future?

Saturday 15th October, 12.05–12.50

Dodd J.M.

Discipline of Obstetrics & Gynaecology, and the Robinson Research Institute, The University of Adelaide, Adelaide, South Australia

Overweight and obesity during pregnancy is common. There is an extensive observational literature highlighting the effects of excessive gestational weight gain among overweight and obese

pregnant women and an increased risk of adverse pregnancy and birth outcomes. However, evidence from large-scale randomised trials which have utilised robust methodology indicate that provision of an antenatal dietary and lifestyle intervention during pregnancy is ineffective in limiting gestational weight gain to the extent required to potentially impact clinical pregnancy and birth outcomes.

This presentation will focus on the evidence available to date for antenatal interventions for pregnant women who are overweight or obese, in addition to challenging the current focus on gestational weight gain as a relevant clinical outcome, and considering alternative approaches which may be required if outcomes for women and their infants are to be improved.

Parallel Sessions

A.1 Pregnancy and Programming of Obesity and Related Disorders

Thursday 13th October, 11.50–13.25

The Role of Epigenetic Processes in the Developmental Programming of Obesity

Karen Lillycrop

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There is now substantial evidence from both human epidemiological studies and animal models that an adverse intrauterine environment induced by a variety of environmental and maternal factors such as diet, body composition or endocrine factors can induce a phenotype in the offspring that is characterized by an increased risk of developing chronic non-communicable diseases in later life. The mechanism by which cues about nutrient availability in the postnatal environment are transmitted to the fetus and the process by which different, stable phenotypes are induced are beginning to be understood and involve the epigenetic regulation of specific genes. Epigenetic processes induce heritable change in gene expression without altering gene sequence. The major epigenetic mechanisms include DNA methylation, histone modification and non coding RNAs. The epigenetic changes induced in response to nutritional cues from the mother may allow the fetus to adjust its developmental programme in order to be better adapted to the future environment, while inappropriate adaptations may predispose an individual to increased risk of a range of non-communicable diseases. Some studies have also suggested that environmental challenges in early life can induce an altered phenotype which can be transmitted between generations.

This talk will describe how early life environment influences later susceptibility to non communicable diseases through the altered epigenetic regulation of genes, and how epigenetic changes

in early life may be used as predictive markers of future disease risk as well as providing valuable insights into the development of complex diseases.

Obesity, Related Metabolic Conditions and DNA Methylation – What Can Genome Wide DNA Methylation Studies in Cohort Studies Show Us?

Rae-Chi Huang

Telethon Kids Institute, University of Western Australia, Perth, Australia

Experimental studies have suggested that the early life environment makes a significant contribution to an individual's risk of developing obesity, through the altered epigenetic regulation of genes. Currently we have multiple gaps in knowledge as to how this occurs in humans. Despite genome wide DNA methylation arrays and large datasets being increasingly available in many birth cohorts around the world, and the progressive identification of loci associated with obesity and related phenotypes, a complete understanding of the role of the methylome in mediating early life environmental exposures on obesity and related diseases remains challenging. Uncertainty exists as to whether variability of the methylome after birth determines or is a consequence of obesogenic postnatal environmental influences.

In this talk, we discuss within the context of the Western Australia Pregnancy Cohort (RAINE), studies to assess epigenetic loci associated with obesity-related phenotypes. We also discuss approaches to understand how early life environmental exposures (maternal BMI, gestational weight gain, maternal smoking in pregnancy, maternal stress, breast feeding) and metabolomic endophenotypes mediate the association between the methylome and adiposity.

The Western Australia Pregnancy Cohort (RAINE) study (www.rainestudy.org.au) recruited 2900 pregnant women between 1989 and 1992. The offspring born to these women have longitudinal exposure data, and adiposity phenotypes. Follow-up of the offspring has been undertaken at birth, 1, 2, 3, 5, 8, 10, 14, 17 and 24 years. During adolescence, all subjects have datasets derived from Illumina Infinium HumanMethylation450 BeadChips and mass spectrometry based metabolomics.

Maternal Obesity, the Gut Microbiome, and Long-Term Disease Risk

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Farncombe Family Digestive Diseases Research Institute,
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Since the advent of the Developmental Origins of Health and Disease (DOHaD) hypothesis, many models have consistently shown that the early life environment plays a central role in offspring disease risk. Recent studies have identified new connections between the mother and its developing fetus through the maternal intestinal microbiome, suggestive of novel links between the maternal-fetal unit and new avenues of investigation into postnatal metabolic development and function. Although still controversial, it has been suggested that the developing fetal gut may be colonized before birth. Bacteria associated with the maternal gut have been isolated from meconium, fetal membranes and cord blood of healthy neonates. Experiments in mice support the notion that the fetus may be exposed to maternal gut-derived bacteria which may prime fetal gut development. There is currently no direct evidence of a fetal-microbial interaction in the context of maternal obesity – although maternal obesity results in offspring that have metabolic defects associated with intestinal dysbiosis, inflammation, and altered immune responses. Studies have shown that maternal microbial priming of neonatal intestinal development likely occurs during lactation through breast milk, and that bacterial populations are modifiable by diet and obesity. Whether early life exposure to maternal microbial/inflammatory triggers in the context of maternal obesity impairs offspring development and predisposes to postnatal obesity, is unclear, but is an exciting new avenue of obesity related programming research.

B.1 Breastfeeding and Later Health

Thursday 13th October, 11.50–13.25

Breastfeeding and Celiac Disease, Allergy and Type 1 Diabetes

M. Luisa Mearin

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Leiden, The Netherlands

Breastfeeding (BF) protection against coeliac disease (CD), allergy and type1 diabetes (T1D) has been suggested. Most of the studies were retrospective and easily biased.

A systematic review of prospective observational studies up until February 2015, showed that BF had no preventive effect for CD. The ESPGHAN position paper 2016 states that although BF should be promoted for its other well established health benefits, neither any BF nor BF during gluten introduction has been shown to reduce the risk of CD.

A systematic review and meta-analysis on BF and asthma and allergies (2015) showed low grade quality evidence of a reduced risk of asthma, allergic rhinitis and eczema in children by longer duration of BF. No association was found between BF and food allergy. Later, a big population-based study in 1 year old Australian showed that duration of exclusive BF and use of partially hydrolyzed formula were not associated with food allergy. The results of the big prospective population study Generation Rotterdam, showed that BF may influence wheezing and asthma in childhood, which seems to be partly explained not by allergy, but by infectious mechanisms.

Some retrospective studies showed a small reduction in the risk of T1D with BF, but almost all the prospective birth cohort studies failed to find a protective effect. Studies showing that children who were still breastfed at the time of introduction to cereals had a reduced risk of T1D suggest that BF might play a protective role in the relationship between dietary factors and T1D.

Persistent Environmental Toxicant and Nutrient Content in Human Breast Milk and Infant Growth

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Background: Perinatal exposure to certain environmental toxicants is associated with rapid infant growth, a risk factor for obesity. Certain plasma lipid profiles in adolescents have been associated with obesity and insulin resistance, and perinatal n-6 and n-3 long chain polyunsaturated fatty acid (PUFA) status may influence long-term health. No studies have examined potential additive or interactive effects of these compounds in human milk.

Methods: We utilized the Norwegian HUMIS study, a multi-center cohort of 2,606 mothers and newborns enrolled between 2002–2008. Milk samples from a subset of 800 women were analyzed for triglyceride fatty acids, phospholipids, and 172 environmental toxicants, including persistent organic toxicants, heavy metals, and pesticides. Obese women were oversampled. Growth was defined as change in weight-for-age z-score between 0–6 months. We will use Bayesian model averaging, which deals with highly correlated mixtures, to determine significant exposures. The resulting frequently selected model will be refit using a multivariable logistic regression.

Results: Median age, pre-pregnancy BMI, and cumulative breastfeeding of mothers was 30 years, 24.0 kg/m², and 5.78 months, respectively. Of infants, 18.2% (n = 143) displayed rapid growth. The toxicant with the highest levels was *p,p'*-dichlorodiphenyldichloroethylene. Also present in high levels were polychlorinated biphenyls 153 and 138. Median concentrations of arachidonic and docosahexaenoic acids, which indicate levels of n-6 and n-3 PUFAs, respectively, were similar at 0.08 g/l (IQR 0.06–0.11 for arachidonic and 0.05–0.12 for docosahexaenoic acid).

Analysis is ongoing. Anticipated results include triglyceride fatty acids, lipid species, and toxicants associated with rapid growth and their potential interactions.

C.1 Programming of NCDs in Preterm Infants

Thursday 13th October, 11.50–13.25

Programming of NCDs in Preterm Infants – Focus on Growth

Ken Ong

University of Cambridge, Cambridge, UK

Preterm infants typically exhibit poor initial weight gain between birth to term gestational age, with subsequent rapid catch-up growth. Rapid early growth is a suggested risk factor for metabolic diseases; conversely poor early growth in preterm infants is associated with adverse neurodevelopmental outcomes.

A recent systematic review summarized the available evidence on postnatal growth in preterm infants in relation to neurodevelopmental and metabolic outcomes. Only few intervention studies were identified, providing little evidence for beneficial effects of faster early growth. Observational studies showed generally consistent positive associations between postnatal weight gain (18 studies) or head growth (15 studies) and neuro-cognitive outcomes at ages ranging from 1 y to adulthood, but no obvious period of weight gain or head growth was more consistently associated with later outcomes. Two of four observational studies reported positive associations between postnatal weight gain and later percent body fat; three of four studies reported positive associations with insulin resistance; and associations with various cardiovascular disease risk factors were more variable. In summary, while observational studies report consistent positive associations between postnatal growth and neuro-cognitive outcomes, there is limited evidence from the few intervention studies. Evidence linking postnatal weight gain to later adiposity and cardiovascular risk factors is also limited. The dissonance between findings of intervention and observational studies raises the possibility of confounding by diseases or factors that affect both growth and cognition. Future nutritional intervention studies in preterm infants should report effects on weight gain and growth, as well as later body composition and neuro-cognitive outcomes.

Glucocorticoid-Programming in Very Preterm Birth

Martijn Finken

VU University Medical Center Amsterdam, Amsterdam, The Netherlands

The adrenal gland is not yet fully functional in newborns in the very preterm (i.e., <32 weeks of gestation) range; a considerable proportion of them manifest clinical signs of adrenal insufficiency, such as refractory hypotension. Conversely, children with a history of preterm birth displayed increased hypothalamus-pituitary-adrenal (HPA) axis activity, though studies have been inconsistent. They were also found to carry the pathophysiological correlates of increased HPA axis activity, such as increased abdominal fat contents, raised blood pressure and glucose intolerance.

The switch in HPA axis activity has repercussions for subsequent growth, body composition, metabolism, neurodevelopment and, ultimately, long-term disease risk. This talk provides an overview of studies that investigated HPA axis activity in very preterm newborns and how the axis changes throughout development, as a possible explanation for the association between prematurity and certain chronic diseases.

A.2 Interventions to Prevent Adverse Fetal Programming

Thursday 13th October, 16.45–18.20

Antenatal Interventions to Improve Maternal and Infant Outcome – Results from the ROLO Study

Fionnuala McAuliffe

University College Dublin, Dublin, Ireland

The ROLO randomised controlled trial is a low glycaemic index dietary intervention aiming to reduce macrosomia (large for dates infants).

800 secundigravida women who had previously given birth to a macrosomic baby (>4 kg) were randomised to receive low GI dietary advice or usual antenatal care, which did not include dietary advice. Low GI dietary advice was given at week 14 of pregnancy and demographic, well-being and lifestyle questionnaires were returned by 28 weeks gestation. 3-day food diaries were completed during each trimester of pregnancy.

Women in the intervention group lowered their glycaemic index and glycaemic load following dietary advice and demonstrated an attenuation in the normal rise in insulin resistance in late pregnancy. Significant maternal benefits were noted with less gestational weight gain and improved maternal glucose tolerance. No impact however was noted on birthweight, though neonatal thigh circumference was slightly less in the intervention groups. At 6 month and 2 years of age no differences were noted in infant anthropometry.

Additional findings from the ROLO study include a positive association between physical activity and wellbeing in pregnancy and relationships between maternal vitamin D status and childhood adiposity.

In summary a low glycaemic index diet was acceptable to pregnant women with good compliance noted and while it did not impact on birthweight significant maternal benefits were noted. This diet to be considered for women at risk of excessive gestational weight gain and those at risk for glucose intolerance in pregnancy.

The UPBEAT Trial; Obesity in Pregnancy and Developmental Programming

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UPBEAT was a randomised controlled trial in 1555 obese pregnant women (BMI >30 kg/m²) of a complex behavioural intervention to encourage change in diet (reduced glycaemic load and saturated fat intake) and increase physical activity. The primary aim, to reduce gestational diabetes and large for gestational age deliveries was not met, but the intervention improved maternal diet, reduced gestational weight gain and lowered maternal adiposity (by anthropometry), all pre-specified secondary outcomes. Neonatal anthropometry assessed at birth was not different between intervention and standard care arms of the trial. 698 infants attended at 6 months of age for a follow up visit. Those in the intervention arm demonstrated lower values for sub-scapular skin fold thickness, an index of central adiposity, (difference -0.26 SD, -0.49 to -0.02; p = 0.031). Maternal dietary glycaemic load and saturated fat intake were reduced in the intervention arm. Causal mediation analysis suggested that lower infant subscapular skinfold thickness was mediated by changes in antenatal maternal diet and gestational weight gain.

The UPBEAT trial has shown that modification of diet in pregnancy can reduce a measure of infant adiposity. The intervention was also associated with lasting improvement in the mother's diet. Follow up of children at 3–4 yrs currently underway will determine whether the maternal intervention has led to persistent change in childhood adiposity.

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The LIMIT Randomised Trial – Longer-Term Effects on Maternal and Child Health

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Overweight and obesity during pregnancy is common. Robust evidence about the effect of antenatal dietary and lifestyle interventions on potential mechanistic pathways and longer-term maternal and childhood health outcomes is lacking.

The LIMIT randomised trial recruited and randomised 2,212 women during pregnancy with BMI >25 kg/m². Women randomised to Lifestyle Advice participated in a comprehensive dietary and lifestyle intervention over pregnancy, delivered by research staff. Women randomised to Standard Care received pregnancy care according to local guidelines, which did not include such information. Women who received the antenatal lifestyle intervention demonstrated improvements in their diet quality and physical activity, while significantly reducing the risk of infant birth weight above 4 kg.

We have conducted follow-up of women and their children who participated in the LIMIT randomised trial at 6 months, 18 months, and 3–5 years of age. This presentation will discuss some of the findings obtained from childhood follow-up assessments.

B.2 GDM – Early Diagnosis of Women at Risk

Thursday 13th October, 16.45–18.20

Studying Fetal Programming in South Asian Populations: The GIFTS Study

Sarah Finer

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The concept of fetal programming has developed from the study of extreme environments and phenotypes, often in historical contexts. To study fetal programming in current populations, and its effect on cardiometabolic disease, a contemporary view must be taken to understand a complex interplay of genetic, epigenetic, environmental, social, cultural and demographic factors. In this EU-funded study, we have sought to understand the complex environmental and molecular drivers of type 2 diabetes and obesity in the offspring of a global population of South Asian women, and develop novel insights to develop targeted interventions to mitigate their risk.

The GIFTS study used a multi-level experimental approach to study fetal programming of cardiometabolic disease in South Asian populations, incorporating epidemiological, molecular, clinical and qualitative techniques.

Data will be presented from selected GIFTS work packages, including epidemiological and clinical studies. These data will highlight the need to take a global perspective and to understand the complex socio-cultural and demographic setting in which fetal programming occurs and to develop effective strategies to prevent cardiometabolic disease. Preliminary evidence of an effective population-level intervention aimed at women of reproductive age, and its potential to break the intergenerational transmission of cardiometabolic disease will be presented.

GDM: Early Identification of Obese Women at Risk

Sara White¹, Debbie Lawlor², Annette Briley¹, Scott Nelson³, Naveed Sattar³, Paul Seed¹, Matias Vieira¹, Lucilla Poston¹, Dharmendra Pasupathy¹, UPBEAT Consortium

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Background: Increasing obesity amongst pregnant women worldwide has been associated with a parallel increase in gestational diabetes (GDM). In clinical practice all obese women are categorised as being of equally high risk of GDM whereas the majority do not develop the disorder. Our aim was to develop an early prediction tool for identification of obese women at high risk of GDM.

Methods: Clinical and maternal anthropometric data and non-fasting blood samples were obtained at 15⁺⁰–18⁺⁶ weeks' gestation in 1303 obese pregnant women from UPBEAT, a randomised controlled trial of a behavioural intervention (diet and physical activity). Twenty one candidate biomarkers associated with insulin resistance, and a targeted nuclear magnetic resonance (NMR) metabolome were measured. Prediction models were constructed using stepwise logistic regression.

Results: 26% of women (n = 337) developed GDM (International Association of Diabetes and Pregnancy Study Group criteria, IADPSG). A model based on clinical and anthropometric variables provided an AUC of 0.71 (95% CI 0.68–0.74). This increased to 0.77 (95% CI 0.73–0.80) with addition of candidate biomarkers but was not improved by addition of NMR metabolites (0.77; 95% CI 0.74–0.81).

Conclusions: It is feasible to identify obese women at high risk of developing GDM later in pregnancy using clinical characteristics and biomarkers measured at early gestation.

Women with Early Diagnosed GDM in DALI: Characteristics and Outcomes

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Introduction: Controversies exist on the use of IADPSG/WHO 2013 GDM criteria for the diagnosis of gestational diabetes (GDM) in early pregnancy.

Research Design and Methods: Pregnant women with a body mass index (BMI) ≥ 29.0 kg/m² enrolled into the DALI pan-European multicentre trials, attended an oral glucose tolerance test in early pregnancy. Demographic, anthropometric and metabolic information were obtained at enrolment. GDM was diagnosed using IADPSG/WHO 2013 criteria. Birth outcomes were obtained from medical dossiers.

Results: GDM prevalence was high (23%) in early pregnancy. Women with early GDM were significantly older, with higher pre-pregnancy BMI, waist circumference, sum of skinfold callipers, blood pressure, triglycerides and free fatty acids. Histories of GDM, macrosomia and pregnancy induced hypertension (PIH) occurred more often in women with early GDM. Women with early GDM had significantly higher insulin resistance and secretion. Among women with early GDM, PIH, spontaneous abortion, caesarean delivery, and delivery of a large-for-gestational age baby occurred more often. Gestational age at birth was reduced, but without increased risk for preterm birth. No differences in birth weight, birth length or Apgar score were found.

Conclusion: The IADPSG/WHO 2013 GDM criteria identify a substantial subgroup of women in early pregnancy with marked insulin resistance and many features of the metabolic syndrome. Pregnancy and birth outcomes were less favourable. Such women might benefit from early treatment. Criteria for GDM in early pregnancy, supported by robust evidence of the benefits of early and late treatment are urgently needed to guide modern European GDM screening and treatment strategies.

C.2 Metabolomics and Obesity in Humans

Thursday 13th October, 16.45–18.20

Opportunities of Metabolomic Studies in the Context of Early Programming

Christian Hellmuth

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Intra-uterine or early life exposures to environmental factors are bearing risks for several metabolic diseases, like diabetes or the metabolic syndrome. Pre-pregnancy BMI, maternal stress and diet, gestational weight gain, placental function, and duration of breast-feeding are considered to have effects on infant growth and development, but underlying mechanisms remain unclear. Thus,

it is worthwhile to investigate metabolic alterations of the mother, the fetus and the newborn, to unravel the link between early programming factors and later disease risk. The scientific study of a large set of small molecules in a given biological matrix such as blood plasma is called 'Metabolomics' and provides a tool to investigate metabolic changes in respect to environmental exposures or identify potential biomarkers of, for instance, obesity or insulin resistance. Beyond, Metabolomics will set the route for new interventions in pregnancy and infancy tackling these non-communicable diseases. The inclusion of several studies – even though not originally set up for mechanistic metabolomic studies – with various primary outcomes and diversity in the data as well as in the studied collectives offers the possibility to answer manifold questions and hypothesis. Recent findings identified a few metabolites, like dihomogamma linoleic acid or branched-chain amino acids, potentially involved in early programming pathways but evidence and causality are still missing.

However, in the absence of an appropriate animal model, evidence for programming mechanisms can only be achieved with repeated, standardized analysis in several longitudinal studies.

Cord Blood Metabolomics and Weight Development Later in Life: Methods and Results from the GINIplus and LISAplus Studies

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Exposure to maternal factors, like obesity or diabetes in-utero, may affect offspring metabolism, growth trajectories and later-life obesity, via effects potentially observable in cord blood metabolites. We aimed to characterize associations of cord blood metabolites with birth weight, postnatal weight gain, and BMI in adolescence. Cord blood samples from 753 LISAplus study participants were analysed. Glycerophospholipid fatty acids (GPL-FA) were measured by gas chromatography. Polar lipids, non-esterified fatty acids (NEFA) and amino acids were quantified by a mass-spectrometry based metabolomic platform. Associations were modelled using linear regression, adjusted for potential confounding factors. Of 581 metabolites, 209 passed quality control. Birth weight was associated with significant alterations in 14 analytes. Lysophosphatidylcholines C16:1, C18:1, C20:3, C18:2, C20:4, C14:0, C16:0, C18:3, GPL-FA C20:3n9, and GPL-FAC22:5 n6 were positively related to birth weight, while higher cord blood concentrations of NEFA C22:6, NEFA C20:5, GPL-FA C18:3n3 and PCe C38:0 were associated with lower birth weight. Postnatal weight gain and BMI z-scores in adolescents were not significantly associated with cord blood metabolites after adjustment for multiple

testing. However, metabolites positively associated with birth weight appeared to be (non-significantly) associated with lower postnatal weight gain. In contrast, metabolites negatively associated with birth weight tended to be positively associated with both postnatal weight gain and BMI z-scores at age 15 years. Although cord blood metabolites were highly associated with birth weight, long-term programming effects of the intra-uterine environment and metabolism on later health cannot be predicted with profiling of the cord blood metabolome.

Analysis of Metabolic Profiles in Adolescents from the RAINE Cohort by Clinical Targeted Metabolomics: Metabolic Consequences of Early Programming

Sebastian Rauschert

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The Western Australian Pregnancy Cohort (Raine) Study offers a huge possibility to study the effect of early programming factors like breast-feeding on early adulthood health and risk for non-communicable diseases, with a special focus on underlying metabolic mechanisms. Sphingomyelin (SM), and phosphatidylcholine (PC) patterns have been identified as potential markers for waist circumference, while lyso-phosphatidylcholine C14:0 was associated with HOMA-IR.

The results suggest weight status-dependent mechanisms for the development of IR with the respective lyso-phosphatidylcholine C14:0 as a key metabolite in non-obese IR.

Further, an analysis according to sex showed significant differences, not only in general, but also disease specific. More than 100 metabolites have been significantly differently associated with sex and hormonal contraceptive use, 43 were significantly different in their effect size on disease markers for the metabolic syndrome and highlighted the importance and need for sex specific analysis in metabolomics and interventional studies that also take hormonal contraceptive-usage in females into account.

However, besides a non-significant tendency for NEFA and Carn to be lower with breast-feeding duration, a general breast-feeding (duration and cut-offs) influence on obesity and the metabolome at 20 years could not be shown. For this, principal components analysis have been performed for general differences based on different breast-feeding cut-offs in months, and multiple regression models were used to look more specifically, if there are metabolites associated with breast-feeding duration.

In conclusion, metabolomics seems to be valid for cross sectional analyses, but the early programming effect might be overlapped by the 20 year factors.

A.3 Postnatal Nutrition

Friday 14th October, 10.45–12.20

Infant Nutrition and Its Effect on the Metabolome

Franca Kirchberg

Ludwig-Maximilians-University, Munich, Germany

With the increasing prevalence of non-communicable diseases such as diabetes or obesity, it becomes more important to investigate and understand the triggering mechanisms. A high protein intake during infancy is suspected to stimulate the secretion of insulin and IGF-1 which in turn enhances the infant's growth ('early protein hypothesis'). The underlying metabolic mechanisms, however, still need to be clarified.

Metabolomics is the study of small-molecule metabolites. Reflecting the interaction of the genome, epigenome, transcriptome, proteome, and the environment, metabolomics facilitates the characterization of pathological conditions.

As part of the double-blind, randomized, multicenter intervention CHOP trial, we examined the metabolic responses to infant formula milk with higher or lower protein content. We obtained 691 plasma samples of 6 months old infants and determined 163 metabolites. BCAA seemed to play a pivotal role in the effect of a high protein diet on the fatty acid oxidation and fat storage in this analysis. Furthermore, the infants were clustered based on their metabolic profile with the aim to unravel underlying metabolic phenotypes. However, we could not identify stable clusters containing infants with similar metabolic profiles.

In summary, a high protein intake resulted in increased BCAA concentrations that might negatively affect fat storage and fatty acids oxidation. Moreover, as we could not identify clusters that the children can be grouped into, we have highlighted the broad diversity of the infants metabolism and their response to the formula milk.

A Global Perspective on the Importance of Dietary ARA and DHA Intakes from Birth to Age 3 Years

Stewart Forsyth

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For international recommendations on ARA and DHA dietary intakes in early childhood to be valid, there needs to be a greater understanding of dietary patterns across both the developed and developing world. The aim of our studies was to provide estimates of intake of ARA and DHA in infants and young children, with a particular focus on developing countries.

Food Balance Sheets from the Food and Agriculture Organisation and fatty acid composition data from Australian food composition tables were utilised to generate median per capita intake estimates for ARA and DHA in 47 developed and 128 developing

countries. Median daily intake of ARA and DHA in children age 6–36 months was determined by combining the estimated fatty acid intake from breast milk and complementary foods during this period. There was a direct relationship between per capita median dietary ARA and DHA intake and the per capita gross national income of the country. Regional analysis showed lowest ARA and DHA dietary intakes per capita were located in Sub-Saharan Africa and Central and Southern Asian populations.

The estimated median daily dietary intake of ARA and DHA in infants and young children aged 6–36 months across 76 developing countries was 64 mg/day and 49 mg/day, respectively, with the major source being breast milk. The lowest ARA and DHA intakes from complementary foods were present in low income countries with the highest birth rates. There are many populations worldwide that have ARA and DHA intakes that do not reflect current international recommendations.

Allergy Prevention by Early Nutrition: 15 Years of Follow-Up in the GINI Study

Sibylle Koletzko, Andrea V. Berg, Birgit Filipiak-Pittroff, Holger Schulz, Ute Hoffmann, Elke Link, Maike Sußmann, Markus Schnappinger, Irene Brüske, Marie Standl, Ursula Krämer, Barbara Hoffmann, Heinrich Joachim, Carl-P. Bauer, Dietrich Berdel, for the GINIplus study group

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Aim: To assess the association between early feeding with hydrolyzed formulas in high-risk infants and allergic outcomes in adolescence.

Methods: Between 1995–1998 newborns at allergic risk were randomly allocated to receive double blind partial whey hydrolyzate (pHF-W), extensive whey hydrolyzate (eHF-W), extensive casein hydrolyzate (eHF-C) or cow's milk formula (CMF) during the first four months of life as breast milk substitute if necessary. Children were followed at 1, 2, 3, 4, 6, 10 and 15 years. Outcomes included cumulative incidence and period prevalence of physician-diagnosed allergies and sensitization to common allergens. Logistic and Log-binomial regression models using generalized estimation equation for repeated measures were performed.

Results: Of 2252 infants (ITT), 637 were exclusively breastfed, of the remaining 988 consumed study formula per protocol (PP). At 15 year 61% (66%) of the ITT (PP) population were followed. Between 11 and 15 years, asthma prevalence was reduced in the eHF-C group (OR 0.49, 95% CI 0.26–0.89). The cumulative incidence of allergic rhinitis was lower in eHF-C (RR 0.77, 95% CI 0.59–0.99) and the prevalence in pHF-W (OR 0.67, 95% CI 0.47–0.95) and eHF-C (OR 0.59, 95% CI 0.41–0.84). The cumulative incidence of eczema was reduced in pHF-W (RR 0.75, 95% CI 0.59–0.96) and eHF-C (RR 0.60, 95% CI 0.46–0.77), as was eczema prevalence between 11 and 15 years in eHF-C (OR 0.42, 95% CI 0.23–0.79).

Conclusion: In high-risk children, early feeding with certain hydrolyzed formulas compared with CMF has long-term health effect on allergic diseases.

B.3 Psychosocial Stress, Mental Health and Biological Impact

Friday 14th October, 10.45–12.20

Interaction of Prenatal Psychosocial Stress and Nutrition: Implications for Maternal and Infant Metabolic Outcomes

Karen L. Lindsay, Christian Hellmuth, Olaf Uhl, Claudia Buss, Pathik D. Wadhwa, Berthold Koletzko, Sonja Entringer

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Two of the most independently studied prenatal factors to influence fetal development and pregnancy/offspring outcome are maternal nutrition and psychosocial stress, yet their combined or interactive effects are considerably understudied. Evidence from non-pregnant animal and human studies suggests that psychosocial stress influences eating behaviour by inducing increased consumption of palatable foods high in fat and sugar. On a physiological level, the hormonal cascade response to stress influences metabolism by suppressing insulin action and promoting gluconeogenesis and lipolysis, in order to raise plasma glucose and free fatty acids. In pregnancy, endocrine function and glucose and lipid metabolism are substantially altered from the non-pregnant state and also naturally change across gestation to support the developing fetus. Thus, prenatal stress exposure has significant potential to alter maternal nutrition on both a behavioural and physiological level. Given the public health urgency of the current childhood obesity epidemic, the potential implications of prenatal stress-nutrition interactions on the programming of infant adiposity and subsequent offspring obesity warrant investigation.

The UC Irvine Ecological Momentary Assessment (EMA) study prospectively assessed prenatal behavioural, psychological and physiological exposures among healthy pregnant women. Metabolomic analysis of maternal plasma samples facilitated detailed analysis of whole body metabolism at each trimester. In this cohort, we have identified metabolomic profiles significantly associated with raised maternal pre-pregnancy BMI that may contribute to 'fetal programming' of adiposity in the neonate. This presentation will further describe interactions between maternal psychosocial and biological stress profiles with nutritional intake and metabolism, and the implications for infant adiposity.

Prenatal Stress and Fetal Programming of Obesity Risk: Association of Maternal Cortisol during Gestation with Infant Body Composition

Pathik D. Wadhwa¹, Sonja Entringer^{1,2}, Claudia Buss^{1,2}, Jerod Rasmussen¹, Dan M. Cooper¹

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Background: Glucocorticoids play a key role during intrauterine development in cellular growth and differentiation. Evidence supports the premise that exposure to excessive concentrations of glucocorticoids produce alterations in fetal physiological systems that may increase the risk for obesity in later life. However, this question has not yet been systematically addressed in humans.

Objective: To elucidate the magnitude and stage of gestation-specific prospective association in humans of maternal cortisol concentrations during pregnancy with infant adiposity in early postnatal life.

Methods: N = 68 newborns from a prospective-longitudinal pregnancy cohort were followed over the early postnatal growth phase until 6 months age. Maternal diurnal salivary cortisol profiles were assessed at 4 consecutive days in early (@13 wks gestation, T1), mid (@24 wks gestation, T2) and late pregnancy (@30 wks gestation, T3). Infant body composition was assessed in the newborn period and at 6 months postnatal age directly with Dual-energy X-ray absorptiometry (DXA) measures and also indirectly with ponderal index measures.

Results: 1) After adjusting for potential confounding factors, higher maternal cortisol concentrations during the beginning of the third trimester of pregnancy were significantly associated with a higher gain in percent body fat (%BF) from 0–6 months age (partial r (adjusted for covariates) = 0.363, p = 0.009), accounting for approximately 13% of the variance in this measure of childhood obesity risk. 2) Commonly-used weight- and length-based proxy measures of infant adiposity (i.e., ponderal index) were only weakly associated with the direct measures of infant adiposity used in our study and were not useful in elucidating the putative effects of maternal cortisol in pregnancy with childhood obesity risk.

Conclusions: The present findings suggest a stage of gestation-specific effect of maternal cortisol (early third trimester) on subsequent infant adiposity gain in early postnatal life and add evidence in humans to support the role of glucocorticoids in fetal programming of obesity risk.

Newborn Insula Gray Matter Volume Is Prospectively Associated with Early Life Adiposity Gain

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Obese children are at greater risk of being obese as adults and developing obesity-related diseases of greater severity. The use of structural and functional MRI has recently helped elucidate the importance of energy homeostasis brain circuitry in adult obesity, yet the developmental time course of this circuitry is unknown. Here, we investigate the association between newborn gray matter (GM) volume in the insula, a brain region underlying energy homeostasis, and rate of fat accrual in the first six months of postnatal life, an outcome thought to be among the most reliable infant predictors of childhood and adult obesity. 52 neonates were assessed using structural MRI within the first month of life and longitudinal Dual X-Ray Absorptiometry shortly after birth and at six months of age. Insula gray matter (GM) volume was inversely associated with change in body fat percentage from birth to six-months postnatal age ($R^2 = 19\%$; $p = 0.001$). The association between insula volume and infant adiposity gain was driven by regions of the insula relevant for gustation and interoception, the direction of effect was in concordance with observations in adults, and the results remained statistically significant after adjusting for relevant confounding variables. Together, these findings suggest an underlying neural basis of childhood obesity that precedes influence from the postnatal environment, thus facilitating the identification of fetal determinants and the discovery of novel strategies for the primary prevention of childhood obesity.

C.3 Placental Nutrient Transfer

Friday 14th October, 10.45–12.20

Maternal Gestational Diabetes Mellitus and Placental Lipids

Olaf Uhl

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Epidemiological observations have shown a link between maternal obesity, pre-pregnancy diabetes as well as gestational diabetes mellitus (GDM) and higher risk for offspring obesity. An unhealthy nutrient supply in early life which programs the fetal metabolism and stays persistent during infancy and childhood has been suggested as the underlying mechanistic background. Increased birthweight, related to childhood obesity, is a consequence of higher fetal fat accretion which is clearly related to lipid metabolism but not necessarily to enhanced placental fatty acid transfer. The study of a large set of small molecules related to energy and lipid metabolism in mothers during pregnancy, in placentas as well

as in cord blood of large cohorts and intervention studies enables to investigate changes in the maternal, placental and fetal metabolism induced by metabolic abnormalities or diet. Specific questions such as the preferred transfer of long-chain poly unsaturated fatty acids, which is known to be limited in GDM, as well as the ability of the placenta to adapt for profuse nutrient supply e.g. in lipid droplets, are objectives of current research. Recent findings identified decreased glycerophospholipid species with dihomo-gamma-linolenic acid and increased species with arachidonic acid and docosahexaenoic acid in placentas of obese and lean GDM, with unknown consequences for the fetus. Glycerophospholipid species with dihomo-gamma-linolenic acid came under suspicion to be related to the transfer of obesity from mothers to the infant, since maternal plasma level of these species were related to pre-pregnancy BMI and cord blood level were associated with increased birth weight.

Placental Lipids and Fatty Acid Transfer in Maternal Overnutrition

Gernot Desoye

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Fetal development in maternal overnutrition (gestational diabetes mellitus, obesity) is characterized by an excessive fat accretion. Because of its key role in mediating maternal-to-fetal nutrient transfer, the placenta has been regarded as contributing to this fetal/neonatal phenotype. The underlying assumption has been a placenta-mediated augmented maternal-to-fetal nutrient transfer, especially of glucose and fatty acids. However, recent *ex vivo* placental perfusion experiments as well as *in vivo* stable isotope studies failed to provide any evidence for an enhanced transfer of fatty acids making up the bulk of fetal adipose fat (C16:0 palmitic acid). Only docosahexaenoic acid (C22:6) transfer is altered. DHA is most important for brain and retinal development, but represents only a minor portion (<2%) of fetal fat.

Some placental fatty acid transporters (FATP1, FATP3) are up-regulated in maternal obesity and correlate with maternal BMI. This may account for more fatty acids being taken up by the syncytiotrophoblast, the outermost placental tissue. There they enter metabolic pathways, predominantly leading to lipid storage. This is accompanied by changes in some proteins involved in the regulation of syncytiotrophoblast lipid storage. The placenta contains more triglycerides in maternal obesity than in lean pregnancies. Storage capacity is limited and at higher maternal BMI triglyceride mobilization may balance formation of new lipid droplets.

Thus, the placenta appears to serve as a buffer protecting the fetus from an oversupply with maternal nutrients. Only when the capacity of the buffer is exhausted, nutrients may spill over to other metabolic pathways (supported by FP7-project Early Nutrition, grant agreement n°289346).

A.4 Early Life Gut Microbiome and Long-Term Health

Friday 14th October, 14.30–16.05

Determinants and Duration of Impact of Early Gut Bacterial Colonization

Christine Edwards

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A significant role for the gut microbiota in chronic diseases ranging from obesity and cardiovascular disease to allergy, autoimmune diseases and autism has been proposed based on differences in the gut microbiome in these conditions as well as increasing knowledge of the effects of bacterial metabolites and fragments on several human systems. A key common element is low diversity in bacterial populations.

The gut is colonized in the first months of life and the microbiota matures slowly over the first two years. The early factors which have been established in different cohort studies to determine the diversity and composition of the microbiome include mode of delivery, perinatal antibiotics, infant feeding method, geography and weaning.

It has also been suggested that prenatal events such as placental transfer of bacteria and transmission of bacteria in breast milk are possible additional routes for colonisation of the newborn infant which may be selectively promoted by maternal physiology. The evidence for this is less clear.

As the adult microbiome is believed to be quite stable, it is important to consider ways that the initial colonisation process can be optimised. However, our understanding of the duration of the effects of early life events on the microbiota is uncertain but may be important in identifying the modifiable factors which can be manipulated to promote long term health.

Dysbiosis in Health and Disease – The Microbiome at Early Life Stages

Dirk Haller

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Human cohort studies demonstrated changes in gut microbiota composition and function (microbiome dysbiosis) in a variety of different pathologies. Development of the gut microbiota in infants is a dynamic process suggested to be critically important for health later in life. Since the colonization mechanisms and factors associated with this process are still largely unknown, we designed a randomized, placebo-controlled intervention trial to assess the impact of early-life intervention with bifidobacteria-supplement-

ed infant formula on the development of intestinal bacterial communities. Stool samples from 106 healthy neonates receiving infant formula with or without a mixture of bifidobacteria. High-throughput 16S rRNA amplicon sequencing and high-resolution mass spectrometry (UPLC-MS) were used to analyze fecal samples collected over a period of two years. Species richness and Shannon effective diversity was not significantly affected by the intervention. Distinct clusters of bacterial communities and metabolite profiles were observed between formula- and breast-fed infants at early age but then converged over time. These shifts were accompanied by the presence of lipid-related and unknown metabolites. Interestingly, none of the formula-derived bifidobacteria were detected in feces after two years. Independent of bifidobacteria supplementation, levels of pyruvate and lactate were high in breast-fed infants, while propionate and butyrate were abundant in both formula-fed groups. In conclusion, infant formula compared to breast milk influences the assembly and metabolite profile of the early life microbiome, particularly associated with increased bacterial diversity. Effects of bifidobacteria-supplemented formula disappeared shortly after the neonatal stage.

B.4 Early Nutrition in Low Resource Settings

Friday 14th October, 14.30–16.05

How to Do Interventions in Early Nutrition with Your Bare Hands: An Example in Guatemala

Michele Monroy-Valle¹, Eunice López-Pazos², Maira Ruano³

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Child community-based growth promotion as a Primary Health Care intervention is a policy in Guatemala, but it struggles constantly between lack of resources as money to train and pay the Community Health Workers, authorities trying to shorten the amount and coverage of interventions, and population demanding the services, in a county where minimum is the standard and social determinants of malnutrition as poverty, inequity, discrimination lead almost half of the children stunted. The objective is to resume the decisions and tasks that were done to institutionalize as a 'package' scattered interventions to improve quality of healthcare focusing on 1000 days extending to under 5 by establishing a process where interventions such as immunization, acute malnutrition treatment, breastfeeding promotion, micronutrient supplementation, food fortification, and others were delivered to all in a frequency that would allow the health system to ensure timely action close monitoring cases of acute malnutrition and reduce mortality from this event. Through literature review, interviews and field observation; interventions and its coverage were identified. Strategy was designed as a 'package' where weight length/height measurement and nutritional status is added as key to aware the caregiver, CHW and epidemiological surveillance. A booklet personal-

ize by sex, delivery of services control forms and the online system were designed as inputs to ensure demand – delivery of personalized intervention. Then validated through a pilot field in 2009. Currently the system takes monthly data, generating early warnings of acute malnutrition and treatment of the country's 22 departments, but impact is difficult to measure.

The Triple Burden of Malnutrition in Sub-Saharan Africa: Translating Knowledge into Action

Reginald A. Annan

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Undernutrition, overweight/obesity and associated Non-Communicable Diseases (NCDs), and micronutrient deficiencies persist in many resource-poor countries, including Sub-Saharan Africa (SSA). In spite of progress made over the years, stunting affects 51 million and wasting affects 10.9 million children under 5 years of age. About 14.0% of SSA children are born with low birth weight (LBW), and close to 4 in 10 women (76.7 million) have anaemia, a risk factor for maternal mortality. About half of pre-school children in the region are vitamin A deficient, over twice the global prevalence, and only 11 of 48 countries in the region have optimal iodine nutrition. Levels of overnutrition and associated chronic diseases are also increasing. Over 5% of under-5 children are overweight, 14% and 3.2% of adolescents are overweight and obese respectively, while 40% of adults are overweight or obese. The situation in the region confirms the understanding of foetal origins of adult diseases, implying that many undernourished girls/boys are ending up as stunted obese adults, with increased risk of NCDs. Undernourished pregnant women are giving birth to LBW children, who have increased risk of poor growth, cognitive and economic development. The intergenerational malnutrition cycle therefore perpetuates in the region, calling for concerted and coordinated action to address it. The role of proper nutrition prior to and during pregnancy, and for the first 1000 days of the child's life has been established. Effective interventions to achieve this, including breastfeeding, dietary diversification, food fortification and supplementation are known. Coherence of policy, coordination of action and financial commitment for scaling up these interventions are urgently required.

Early Life Nutritional Programming of Health and Disease in the Gambia

Sophie E. Moore

MRC Human Nutrition Research, Cambridge, UK

Exposures during the early life (periconceptual, prenatal and early postnatal) period are increasingly recognised as playing an important role in the aetiology of chronic non-communicable diseases (NCD). The 'Developmental Origins of Health and Disease' (DOHaD) hypothesis asserts that adverse early-life exposures – most notably unbalanced nutrition – leads to an increased risk for a range of NCDs, including coronary heart disease, stroke, hypertension, Type 2 diabetes and osteoporosis, and that disease risk is highest when there is a 'mismatch' between the early- and later-life environments. Thus, the DOHaD hypothesis would predict highest risk in countries where food insecurity creates nutritional vulnerabilities for pregnant women and their offspring, yet with in the same contexts we observe a rapid transition to nutritional adequacy or excess in adulthood.

In this presentation, I will review data from our work conducted in rural Gambia, West Africa. Using demographic data dating back to the 1940s, the follow-up of randomised controlled trials of nutritional supplementation in pregnancy and the 'experiment of nature' that seasonality in this region provides, we have investigated the DOHaD hypothesis in a population with high rates of maternal and infant under-nutrition, a high burden from infectious disease, and an emerging risk of NCDs.

C.4 Early Programming of Taste and Appetite

Friday 14th October, 14.30–16.05

Development of Food Preferences and Appetite in the First Years

Sophie Nicklaus

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Infants are born equipped to ingest nutrients, but have to learn how, what and how much to eat. This must occur early, because the mode of feeding evolves dramatically, from 'tube' feeding in utero to eating table foods with the family. Eating habits established during early years contribute to the development of subsequent eating habits. Therefore, it is fundamental to understand the most important early periods for the development of eating habits and the drivers of this development. Here we will focus on the first three years of postnatal life. Several characteristics of the eating experience contribute to drive infant's eating and to shape preferences and energy intake control: food sensory properties; food energy density, social context of eating. The learning processes involve repeated exposure (including to a variety of flavours), asso-

ciation with post-absorptive consequences (energy density) and with contextual signals (interaction with family members). Beyond the first flavour discoveries during the prenatal and lactation periods (through the infant's exposure to flavours from foods of the mother's diet), the most important phases for learning food preferences and appetite control may be the beginning of complementary feeding. Infants discover the sensory (texture, taste and flavour) and nutritional properties (energy density) of the foods that will ultimately compose their adult diet; parents are still in charge of providing appropriate foods, timing, context for eating. Inter-individual differences in learning, related to temperamental dimensions, to sensitivity to food cues (sensory cues or energy density) are large and also have to be taken into account.

Transmission of Food Aromas into Human Milk – An Active Early Programming Trigger or Rather a Passive, Metabolically-Controlled Phenomenon?

A. Buettner, J. Beauchamp, M. Denzer-Lippmann, F. Kirsch, H. Loos, S. Sandgruber, L. Scheffler, C. Sharapa

University Erlangen-Nuremberg, Erlangen, Germany

Exposure to flavors in early developmental stages is believed to influence food preferences in later stages of human life. Regular consumption of carrot juice during the breastfeeding period, for example, was demonstrated to influence facial expressions in infants whose mothers regularly consumed carrot juice, whereby those infants were found to react more positively to being exposed to carrot flavor themselves. Similarly, changes in consumption patterns and behavioral responses have been observed for garlic consumption, leading to the assumption that aromas are transmitted from foods into human milk and a presumed early conditioning effect. On the other hand, biotransformation processes may lead to substantial modification of ingested food aroma constituents, comprising both degradation and formation of sensorially active substances.

Chemo-molecular studies on this topic are scarce, thus our goal is to focus on characterizing the underlying physiological and chemical processes involved in the potential aroma transmission processes in human milk. Sensorially-relevant aroma substances from different nutritional interventions, together with their potential metabolites, are monitored in milk of human subjects, as well as in their urine and exhaled breath. Biotransformation and transmission, as well as elimination processes, are thereby monitored under real-life conditions. A specific focus is made on every-day dietary constituents that are of relevance for breastfeeding mothers.

Early Exposure in Promoting and Programming Healthy Eating

Marion M. Hetherington

School of Psychology, University of Leeds, Leeds, UK

Flavour exposure begins *in utero* and after birth babies experience a variety of flavours through breastmilk. Experience of flavour continues during complementary feeding which is a sensitive period for developing preferences for food flavours which can be enhanced by repeated exposure. In a series of small scale studies, we have explored the ways in which mothers provide their solid foods to their infants, specifically vegetables and we have tested systematically a step by step approach to adding novel vegetable flavours to breast or formula milk to encourage liking and intake, as well as exposure to novel vegetables during the pre-school period. Across all studies conducted to date through both the Ha-bEat and VIVA projects, a consistent finding emerges that age predicts intake of familiar and novel vegetables, that intake increases by the 5th exposure and that individual differences contributing to food avoidance/approach are significantly associated with willingness to consume vegetables. Older children are more reluctant to try new vegetables and they express greater levels of neophobia than young children. Overall, food preference and acceptance develop early in life, it is enhanced by repeated and varied exposure specifically to novel vegetables forming the foundation of healthy eating.

A.5 Nutritional Epigenetics of Obesity

Friday 14th October, 16.30–18.05

Does Epigenetic Variation Impact Type 2 Diabetes?

Charlotte Ling

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It is well established that combinations of genetic and environmental factors affect the susceptibility for metabolic disease e.g. type 2 diabetes. While ageing, obesity and physical inactivity represent non-genetic risk factors for metabolic disease, genome-wide association studies have identified common genetic variants associated with disease. However, epigenetic modifications such as DNA methylation and histone modifications may also promote metabolic disease. Indeed, studies from our group demonstrate that epigenetic variation is involved in the pathogenesis of type 2 diabetes and altered metabolism in humans. We have identified altered DNA methylation patterns in pancreatic islets, the liver, skeletal muscle and adipose tissue from patients from type 2 diabetes compared with non-diabetic controls. We have also shown that environmental factors, including exercise and diet, affect the DNA methylation pattern in human skeletal muscle and adipose tissue. Moreover, we recently demonstrated that polymorphisms associated with type 2 diabetes directly modify the epigenetic pat-

tern in human tissues. Finally, our data also support that an impaired intrauterine environment affect the epigenome in humans, which may contribute to metabolic diseases. Together, our studies support that epigenetic variation contributes to metabolic disease and type 2 diabetes.

Development, Epigenetics and Later Obesity

Keith Godfrey

Professor of Epidemiology & Human Development, Director, Centre for the Developmental Origins of Health and Disease, and Director, NIHR Southampton Biomedical Research Centre in Nutrition, Southampton, UK

Within the Southampton Women's Survey, we have shown greater adiposity in the offspring in association with higher maternal adiposity, poor quality maternal diets in pregnancy, low maternal vitamin D status, excess gestational weight gain, and short duration of breastfeeding. Studies in animals, and initial data in humans, suggests the environment during early life induces altered phenotypes in ways which are influenced or mediated by epigenetic mechanisms. Most is known about DNA methylation changes, and our studies have identified and replicated associations between a number of perinatal methylation marks and the child's later adiposity. Integrating the genotype with epigenetic marks holds the promise of better understanding the biology that underlies the complex interactions of inherited and environmental components that define the developmental origins of common non-communicable diseases. Neonatal methylomes contain molecular memory of the individual in utero experience but are also partly a consequence of DNA sequence polymorphisms that result in methylation quantitative trait loci (methQTLs) and, potentially, the interaction between fixed genetic variation and environmental influences. Among 237 neonates from the Singapore GUSTO cohort we found 1423 punctuate regions of the methylome that were highly variable across individuals, termed variably methylated regions; around 25% of these regions were best explained by MethQTLs, with the remainder best explained by the interaction of genotype with different in utero environments, including maternal smoking, maternal depression, maternal BMI, infant birth weight, gestational age, and birth order. Future insights will depend on considering both fixed genetic variation and environmental factors in interpreting epigenetic variation.

Epigenome-Wide DNA-Methylation and Body Composition at Age 5.5 Years in the European Childhood Obesity Project (CHOP)-Study

Peter Rzehak¹, Marcela Covic¹, Richard Saffery², Eva Reischl³, Simone Wahl³, Veit Grote¹, Martina Weber¹, Annick Xhonneux⁴, Jean-Paul Langhendries⁴, Natalia Ferre⁵, Ricardo Closa-Monasterolo⁵, Joaquin Escribano⁵, Elvira Verducci⁶, Enrica Riva⁶, Piotr Socha⁷, Dariusz Gruszfeld⁷, Berthold Koletzko¹ for the European Childhood Obesity Trial Study group

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Background: Epigenetic plasticity is considered to play an important role for developmental programming towards childhood and adult obesity. Epigenome-wide methylation signatures in relation to BMI and less frequently to body composition have been studied in adults but rarely in children.

Methods: In this epigenome-wide-association-study (EWAS) in 374 children from four European countries participating in the European Childhood Obesity Project, body size (BMI, WHO-standardized BMI) and body composition measures (fat mass (kg), fat free mass (kg), fat mass index (kg/m²) and fat free mass index (kg/m²)) were regressed on 431313 CpG methylation values determined in blood cells using the Illumina HumanMethylation 450 BeadChip array. EWAS analyses were adjusted for study characteristics (sex, age at blood draw, study centre, parental education) biological (white blood cell types) and technical effects (control probe derived principal components).

Results: BMI, fat mass, fat free mass and height related indices were associated with >200 CpGs in >32 genes. Consistent associations with at least four body size and composition measures were found for CpGs in 5 genes, *KLHL6*, *ZDHHC17*, *FARP1*, *SNED1/IRE-BP1* and *CILP2* and in three measures for 27 further genes.

Conclusion: This EWAS provides novel evidence linking DNA methylation at *KLHL6*, *ZDHHC17/HIP14*, *SNED1/IRE-BP1*, *CILP2*, and *FARP1* genes with increased body size and body composition measures in preschool children. Replication in further populations is required to corroborate these findings. Assessment of epigenetic stability of these associations over the life course and investigation into metabolic function and gene expression seems worthwhile.

Funding: Commission of the European Communities, the 7th Framework Programme, contract FP7-289346-EARLY NUTRITION and the European Research Council Advanced Grant ERC-2012-AdG – no.322605 META-GROWTH.

B.5 Catch-Up Growth

Friday 14th October, 16.30–18.05

Proven Benefits of Nutritional Modifications of Patterns of Growth

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Growth patterns have been characterized with the use of different models, often depending on the purpose of their application. Certain growth patterns in early childhood have been associated with adverse or beneficial health outcomes in later life. Particularly rapid weight gain in infancy has been associated with increased risk of subsequent obesity. Among many determinants of early growth, feeding practices have been considered to be of major importance. This is a systematic summary of current evidence on the influence of selected nutritional interventions or exposures (such as protein intake in infancy and breastfeeding) on growth patterns, described with the use of different growth parameters. At least 2 electronic databases and other sources of data were searched in order to identify all relevant studies. An attempt to translate particular effects of nutritional modifications on growth into selected long-term health outcomes (such as overweight and obesity) has been made.

Long-Term Adverse Effects of Catch-Up Growth

Atul Singhal

The Childhood Nutrition Research Centre, Institute of Child Health, University College London, London, UK

The impact of catch-up growth (the higher than expected rate of growth seen following recovery from illness or starvation) or growth acceleration (upward centile crossing for weight or length) on later health has generated considerable interest. The fact that 'catch-up growth' occurs in animal species as diverse as mammals, birds, and fish, as well as humans, suggests that this pattern of growth must have a survival advantage. However, the observation that animals or humans do not usually grow as fast as they are capable of (e.g. as seen during catch-up growth) suggests that faster growth must also have a biological cost. There is, therefore, a trade-off in order to optimise growth trajectories between short-term gains and long-term costs.

In infants born prematurely, faster post-natal growth improves long-term cognitive function but is associated with later risk factors for cardiovascular disease. So, on balance, current policy is to promote faster growth by increasing nutrient intake. Similarly, in low birth weight infants from low income countries, a global problem affecting 20 million births per year, the short-term benefits of faster post-natal growth may outweigh long-term disadvantages. However, whether similar considerations apply to infants from countries in transition is uncertain. For term infants from devel-

oped countries, promoting catch-up growth by nutritional supplementation has few advantages for short- or long-term health, but may increase long-term risk of obesity and metabolic disease. The present review considers the long-term effects of faster post-natal growth focusing on the biology and clinical impact.

C.5 Translational Application of Programming Evidence. What Is Needed?

Friday 14th October, 16.30–18.05

Why Early Prevention of Childhood Obesity Is More Than a Medical Concern – A Health Economic Approach

Diana Sonntag

Mannheim Institut for Public Health (MIPH), Medical Faculty of Heidelberg University, Mannheim/Heidelberg, Germany

Childhood obesity is more than a medical concern; it is receiving an increasing economic attention. This is mainly due to substantial cost burden, particularly later in life. Childhood obesity results in direct and indirect costs; the direct costs are predominantly healthcare costs due to change in the risk of developing obesity-related comorbidities and responding to treatment, while the indirect costs are related to sick leave, reduced productivity and premature death. Indeed international cost-of-illness studies provide robust evidence that societies will face substantial additional lifetime costs related to childhood obesity. In Germany, for example, individuals who were overweight or obese during childhood cause 3–5 times higher lifetime costs than individuals who were normal weight as child.

Given high excess lifetime costs of childhood overweight and obesity, an obvious policy question is: how would changing the current childhood obesity trend result in cost savings and how to reallocate cost savings to implement new pediatric preventive programs? Recent systematic literature reviews confirm that there is an urgent need to conduct health economic evaluations assessing whether a new pediatric preventive programs offers good value for money. However, health economic evaluations had often not being planned from the beginning of an intervention and making it thus, difficult for intervention developers, health care specialists and economists to coordinate their efforts in an effective fashion. It is therefore advisable to involve health economists during the design phase of an intervention. Equally necessary is the development of a toolbox for efficient data acquisition.

Science, Policy and Consumers, Understanding Infant Feeding Communication Practices

Monique M. Raats

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Decisions about whether to breastfeed infants, and when to introduce complementary foods, are important health decisions that are in part influenced by the information environments in which parents and other caregivers find themselves. The emphasis in written advisory materials appears to relate to short term health effects of infant feeding decisions (e.g. infections or food allergy) rather than longer term consequences (the development of chronic conditions such as obesity, diabetes, cardiovascular disease and cancer later in life). This is in line with protection motivation theory that purports that more immediate threats are more likely to be more effective at generating health protecting behaviours than threats that are remote. The lack of emphasis on the long-term health effects of diet, and absence of any mention of programming theories in information materials, may reflect the uncertainties in the underlying science, and lack of consensus about mechanisms and impact. The prevalence of health-related claims and symbols on foods intended for babies and infants (e.g., milk formulas and follow-on foods) will also be discussed. Where the scientific evidence is scarce or inconclusive, policies and associated information for consumers may be cautionary. There may be scope to improve infant feeding practices by increasing the quantity and specificity of messages about health effects, including the implications of nutrition programming. Further research is required to explore how people process, interpret and respond to alternative means of providing information on infant feeding practices and related policies.

How Do We Get Parents Involved?

Silke Mader

Chairwoman of the Executive Board, EFCNI, Munich, Germany

For parents of a preterm infant everything turns upside-down from one day to another with challenging and unexpected situations. Silke Mader, Co-founder and chairwoman of the European Foundation for the Care of Newborn Infants (EFCNI), knows from her own experiences that a family needs much more than medical care and support – parents also need to be empowered and involved in their parental role from the beginning.

Since a few years EU research grants demand patient involvement, but this is for both, scientists and patients a new development. The challenge for a respective collaboration and partnership in research projects is to accept and to understand both opinions. It is a challenge, but also a chance for getting the best possible care, treatment and research for preterm and newborn infants. To face these challenges and to involve parents, research with children and parents needs more time and special communication trainings for developing a valuable and equal partnership. It is important to be transparent with all information, potential risk factors or side effects to get the acceptance from the families. Therefore, understand-

able patient and parent information has to be provided and the voices of parents and children have to be accepted and respected.

EFCNI collaborates with scientific and health professional societies as well as parent organisations to improve the situation for preterm and newborn health in Europe. Silke Mader and team members of EFCNI are involved in many research programmes and studies, having functions in the boards of different societies as patient representatives.

A.6 The Future of Early Life Research

Friday 14th October, 10.10–11.45

Finding Molecular Trackers of Early Life Environment in High-Dimensional Data

Joanna D. Holbrook

Singapore Institute for Clinical Sciences, Singapore

If the seeds of disease are sown early in our lives, it must be possible to detect changes to our biology, at the molecular level right at the beginning of disease etiology. Increasingly we are finding molecular correlates with early life environments, some of which can predict later life disease phenotypes. One particularly productive source of molecular biomarkers are DNA methylation marks, which putatively combine the influences of genotype and early life environment. We have turned to methylome wide association studies (methWAS) to search for these epigenetic trackers of early life environment and disease trajectory. MethWAS has inherent challenges in terms of statistical power and confounding but has uncovered some important general truths as well as robust examples of molecular trackers. Here I discuss our experiences of methWAS in the GUSTO birth cohort and how combining with data from other cohorts at different stages of the lifecourse has yielded hypotheses about cause and effect of DNA methylation biomarkers, early environment and disease.

Genetics and Epigenetics in DOHaD – Future Perspectives

Richard Saffery

Cancer and Disease Epigenetics, Murdoch Childrens Research Institute and University of Melbourne, Melbourne, Australia

There is now compelling evidence for epigenetic variation as a mediator of developmental programming effects in humans. Specific exposures can induce predictable variation in the epigenome prior to birth, much of which is stable postnatally. It is also clear that genetic variation plays a key role in shaping the epigenome and modulating the effects of environmental exposures. For non-communicable diseases (NCD), the emerging picture is one of underlying genetic risk (shaped over generations) that in-

teracts with the cumulative modern environment to determine risk. The breadth of inter-relationships is complex and variable, dependent on dose, duration and timing of exposure and underlying genetic profile. Thus, understanding NCD aetiology is far more complex than simply mapping genetic variation, requiring detailed measurement of exposures and phenotype with regular biological sampling of different tissues. There have been recent notable successes, largely driven by four key factors:

1. Availability of longitudinal pregnancy cohorts established decades ago.
2. Advances in 'omics technologies in reliability, sensitivity and affordability.
3. A wider acceptance of the DOHaD field and fetal programming.
4. An acknowledgement that the impact of data from team studies is greater than the sum of their individual outputs.

The field is still relatively young, but the way forward is clear. Modern longitudinal pregnancy cohorts are essential, preferably utilising standardised exposure and phenotype measures with extensive and harmonised biospecimen collection. Multidimensional 'omics data remains prohibitively expensive and analytical approaches suboptimal, but this is likely to change in the near future. It is likely that our understanding of DOHaD causal pathways will mature rapidly in parallel to the many international pregnancy cohorts now underway.

Maximising the Impact of DOHaD Research

Mark Hanson

University of Southampton, London, UK

The concepts which DOHaD research has established can have substantial impact in several areas. First, including them in educational curricula will help to engage young people in devising and evaluating interventions for primary prevention of NCD risk early in the lifecourse, to improve their future health and that of the next generation. The wider public is also ready to engage with such concepts, as the perception that NCD risk is not explicable largely in terms of genetics, and that adult lifestyle interventions are relatively ineffective, is spreading. Moreover, DOHaD research in developmental epigenetics is finally showing the irrelevance of the nature/nurture dichotomy, and receiving much media coverage. Secondly, DOHaD concepts are vital to the training of a wide range of healthcare professionals in order to meet 21st Century health challenges. Whatever their specialty, these professionals need to ensure that young people are aware of the need to adopt a healthy lifestyle, especially those of inaccessible population groups.

Next, there are substantial implications for public health policy. The scale of the DOHaD contribution to e.g. later NCD risk may not be known, but the interventions needed in the preconception, pregnancy and postnatal periods are not rocket science. Arguably, they should not be deferred until RCTs have been performed or the obligation on states to institute them be dodged by transferring responsibility to individuals/parents.

Lastly, the SDGs open new avenues for international cooperation and a mandate for translating DOHaD research into action.

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B.6 Obesity Prevention and Intervention in Infants and Young Children

Friday 14th October, 10.10–11.45

Early Obesity Prevention – The Role of Life Style. Experience from Toybox and Other Intervention Trials

Piotr Socha

The Children's Memorial Health Institute, Warsaw, Poland

Obesity risk seems to be strictly related to nutrition and dietary habits in early life. Early interventions can decrease obesity risk in children and their eating habits. Early education of parents was tested in a randomized controlled trial in 667 mothers, who started education already in pregnancy. Results were analyzed at the age of 2 y – BMI was lower in the intervention group (16.53 vs 16.82) and there was positive effect on vegetable consumption and TV viewing in those children (Wen LM, BMJ 2012). A systematic review of randomised controlled trials showed that interventions specifically designed to build maternal self-efficacy around infant feeding had impact on weight outcomes (Redsell SA, Maternal Child Nutr 2016).

We performed the EU project called ToyBox which was a multifactorial evidence based approach using behavioural models in understanding and promoting fun, healthy food, play and policy for the prevention of obesity in early childhood. The results of the intervention are being evaluated, still interesting finding concerning behaviours of preschoolers were also analyzed and published. Behaviours differ among countries and for example the specific problem for Poland and Belgium is high soft drink consumption. Children in both groups improved their eating habits but the effect as more significant in the intervention group for prepacked fruit juice consumption (AS Pinket 2016).

Acknowledgement: Toybox group led by Y Manios.

Food Choices, Lifestyles and the Prevention of Overweight and Obesity in Children: Evidence from the IDEFICS Cohort and The I.Family Study

Wolfgang Ahrens

University of Bremen, Bremen Institute for Prevention Research and Social Medicine, Bremen, Germany

As extension of the IDEFICS cohort the I.Family study investigated the impact of dietary, behavioural and socio-economic factors on non-communicable chronic diseases in European children.

In 2007/2008 the baseline examination (T0) of 16,228 children aged 2 to 9.9 years took place in Belgium, Cyprus, Estonia, Germany, Hungary, Italy, Spain and Sweden. 11,041 of these boys and girls participated in the first follow-up examination (T1) in 2009/2010 and 7,105 in the second (T3) in 2013/2014. Half of them

participated in a community-oriented primary prevention programme on obesity. Parents reported lifestyle data for their small children and families while self-reports were collected from adolescents. All children underwent detailed physical examinations.

We observed unfavourable health outcomes due to the lack of sleep and physical activity, excessive exposure to TV, unhealthy diet and rapid weight gain in pre-school age. An unfavourable built environment was associated with reduced physical activity levels. Exposure to TV advertisements was associated with unhealthy food choices. From T0 to T1 the prevalence of overweight/obesity increased to a similar degree in the control (18.0% to 22.9%) and the intervention group (19.0% to 23.6%). However, the intervention prevented unfavourable changes in sedentary time and light physical activity in those receiving a medium-high intervention dose. Also, intervention children being overweight/obese at baseline had a significantly greater probability of normalised weight status after 2 years. Although the intervention did not reduce the incidence of overweight/obesity its results may guide future interventions. This cohort enables us to identify early life factors affecting later health outcomes.

Determining Factors and Critical Periods in the Formation of Eating Habits: Results from the HabEat Project

Sylvie Issanchou

French National Institute for Agricultural Research, Paris, France

Eating habits form early during childhood and are likely to track until the beginning of adulthood. Thus, understanding the formation of eating habits is important.

In the HabEat* project we focused on the development of preferences for vegetables since they are the less liked foods for children. Based on the analyses of data from different European cohorts, HabEat found that breast milk may facilitate the consumption of vegetables in later childhood. HabEat found some evidence that introducing a variety of different vegetables in the complementary feeding period increases later acceptance of novel foods. HabEat also found that repeated exposure is as much or more efficient than flavour-flavour learning to increase vegetable intake even for 2- to 6-year-old children who are more likely to be neophobic.

Food intake adjustment in young children (aged 3 to 6 years) was also studied. HabEat found that when children ate a preload of energy-dense food less than one hour before a meal, they ate less during the meal but adjusted their food intake only partially for the energy ingested from the preload. HabEat found that when palatable foods were available freely after a meal, most children ate in the absence of hunger and consumed extra energy, and the extra-consumption was higher for children whose parents who used 'Food as a reward' than for children whose parents did not use this practice.

* The research leading to these results has received funding from the European Community's Seventh Framework Programme (FP7/ 2007–2013) under the grant agreement n.FP7-245012-HabEat.

C.6 Personalised Nutrition and Counseling

Friday 14th October, 10.10–11.45

Personalised Nutrition – What Does It All Mean?

Rosalie Grivell

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Background: There is much known about the importance of diet and physical activity during pregnancy. In both normal and overweight/obese women, diet and physical activity may mediate many health outcomes for women and their infants, as does weight gain during pregnancy. Related to this, gestational weight gain that is excessive in one pregnancy may further influence health for the woman later in life. Significant efforts are being made to develop and assess various interventions in pregnancy that could potentially improve diet, physical activity and related health. Many aspects may be considered when developing and implementing interventions in this field. Drawing on our research group's experience in designing and running large scale clinical trials in pregnancy, this presentation will attempt to provide an overview of personalised counselling and nutrition as it relates to improving outcomes for women and their infants.

Framing Personalized Nutrition and the Food4me Experience

Hannelore Daniel

Technische Universität München, Munich, Germany

Like in many other sectors, diversification and individualization is also driving the food markets with products and services. This is currently mainly based on food/taste preferences and enjoyment in product lines such as coffee, chocolate or beverages. The highest level of personalization is achieved by offers to compose your 'own' food item such as a breakfast cereal or chocolate.

HEALTH is considered as a key market driver. When taken into the food and nutrition sector, the key question is, how health-promotion can be achieved at the level of the individual and the foods consumed. What can be predicted is that a wide range of web-based health services will become available and those will also employ numerous electronic devices that allow assessment of food intake and measurements of a variety of lifestyle parameters (exercise, sleep, leisure time) and health indicators (blood pressure and glucose, metabolite profiles etc). Whether genetics should/will be included is to be seen, but is likely. Based on these parameters individualized dietary recommendations but also menu plans can be generated and those customized menus may be preordered in a restaurant or for home-delivery. Electronic devices will also be available as shopping guides. It is predicted that the entire supply of foods becomes personalized as more and more consumers outsource this to a food or menu service provider

(e-commerce). Health insurances may be part of such services and the measures of compliance may be used in adjusting individual health insurance plans. Such a system of course challenges some fundamental principles of liberal societies and it remains to be seen how societies scope with this. Personalization can not only be the highest level of possible services but will clearly also be the highest level of personal responsibility. The EU-project *Food4me*

was the largest project ever for testing the concept of personalized nutrition in the field. With 1.600 participants from seven European countries different levels of personalisation in dietary advice systems were defined and tested for compliance and their efficacy for changing dietary intake and life style parameters. I shall be presenting the Food4me framework, its main tools and the key findings.