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EARNEST

EARly Nutrition programming- long term follow up of Efficacy and Safety Trials and integrated epidemiological, genetic, animal, consumer and economic research

Instrument: Integrated Project

Thematic Priority 5.4.3.1: Food Quality and Safety

Publishable final activity report on Workpackage 3.4:

Integration of molecular and physiological outcomes of perinatal programming

Period covered from 15.04.2005 to 14.10.2010

Start date of project: 15.04.2005

Duration: 5,5 Years

Organisation Name of Lead Contractor for this report: Pr. Michael Symonds and Dr. Sylvain Sebert University of Nottingham (UNOTT)

Introduction

During this research project we focused on establishing the importance of the early nutritional and energetic environment on the long term determination of energy homeostasis. According to the original objective of the workpackage 3.4 our research focussed on establishing early nutritional programming in sheep, an animal model with close similarities with humans with regard to gestational and development outcomes.

Objective of the project

The main objective within the EARNEST integrative project was to demonstrate the importance of the early energetic environment in critical windows of development on the long term susceptibility to metabolic diseases. The importance of a later “obesogenic” environment in potential early programming has been assessed. Three main critical windows have been specifically targeted: **1)** Mid gestation, a period coinciding with early organogenesis, **2)** Late gestation, a stage demonstrating maximal fetal growth and **3)** the lactation/weaning period. We particularly focused during this project at understanding the impact of the early nutritional environment on physiological outcomes of clinical relevance (*e.g.* insulin sensitivity, body composition, appetite) and to integrate such phenotypic metabolic features with relevant alterations upon cellular activity *ex vivo*.

Research rationale and hypotheses

Inadequate early dietary environment during critical stages of development is now recognised as a pivotal risk factor in the long term metabolic health. Numerous life-long epidemiological surveys, corroborated by animal experiments establish that part of the risk of being obese, of contracting cardiovascular diseases and type II diabetes later in life originate from a suboptimal energetic environment during fetal and infant development. Understanding the early mechanisms specifically altered by early nutrition will not only help to impact on metabolic diseases prevalence, it will also contribute to optimising early growth development for healthy growth of the population.

A – Influence of early-to-mid gestational restriction and juvenile obesity in offspring metabolic health

Study objective and design

Early-to-mid gestation is a period critical upon tissues organogenesis and remodelling that may predispose to the development of cardiovascular diseases later in life. However, tissue specific events as well as the interaction within the later development of obesity were unknown and was thus the main objective of our study.

Study design – The design of our study has been published on several occasions (see (1) for details). Briefly Twin-bearing pregnant sheep were either fed a control diet (C) or a nutrient restricted diet (NR; 50 of C) from day 30 to day 80 of gestation (term 145 days). Sheep and lamb were then fed according to requirement during thereafter until weaning (80 days of age). At weaning C and NR offspring were raised in obesogenic environment to favour obesity.

Results

Table 1 Summary of the effect of early-to-mid gestational nutrient restriction and juvenile obesity in the offspring at 1 year of age

	CO	NRO	Ref.
	<i>Birth weight and postnatal growth identical</i>		(2)
	<i>Weight and body composition as adult identical</i>		
Perirenal outcomes			
- Kidneys	Glomerulosclerosis Apoptosis Oxidative stress ER stress	Absence of glomerulosclerosis and exaggerated apoptosis	(3, 4)
- Perirenal adipose tissue	Hypertrophy	Hypertrophy Crown-Like structure ER stress Macrophage infiltration	
Cardiovascular	Hypertension Left ventricle hypertrophy	Hypertension Left ventricle hypertrophy Intra-cardiac lipid infiltration Alteration of FFA trafficking	(5)
Insulin sensitivity	Insulin resistance Hyperinsulinism	Insulin resistance Exaggerated hyperinsulinism	(2)
		Intra hepatic triglycerides	(6)
Appetite control		Hypophagia Leptin resistance Exaggerated FTO expression	(1, 2)

A period of energy deprivation coinciding with early organogenesis in the fetus has multiple consequences. It impacts offspring metabolism in a tissue specific manners. Some of the effects have been found to aggravate the impact of juvenile obesity whereas in certain organ such as the kidney and the hypothalamus, energy restriction may be protective.

Aside from such multi-organ responses we established that common factors could be responsible for such long term outcomes. Nonetheless due to organ specific cell regulation it has organ specific consequences. In fact, the mechanisms that impact oxidative stress and inflammation are consistently reset. In addition cellular factors such as PPAR-gamma and its co-activator (PGC1-Alpha) together with FTO are affected in association with modulation of ectopic lipid infiltration.

Such constant alterations upon cellular oxidative stress and energy metabolism may be of critical importance in the understanding in the early programming of long term metabolic outcomes. It suggests that the early nutritional environment acts to determine cellular energy homeostasis. We therefore hypothesise that the energetic environment during development acts on the long term setting of energy metabolism primarily through changes upon intracellular energy balance. The balance between cellular anabolism and catabolism is respectively regulated by the AMP activate kinase (AMPK) and the mammalian target of rapamycin (mTOR). These two key proteins are indeed able to gauge the energy that is available to a cell and to influence metabolism accordingly (7). Such energy sensing pathways appeared critical in the control of whole body energy homeostasis and can play pivotal functions in the regulation of food intake (7, 8). As a main research objective we further

demonstrated the importance of the early environment in the setting of energy sensing pathways in tissues and organs that controls appetite both centrally and in the periphery. We focused on the importance of such programming effects during the perinatal period. This period is critical in the acquisition of both metabolic function and birth weight and is important in the development of the gastro-intestinal tract. We thus studied the influence of late gestational maternal nutrient restriction and early postnatal growth on the setting of appetite and metabolic health in the offspring exposed to a later obesogenic environment after weaning (See Figure 1 for experimental design).

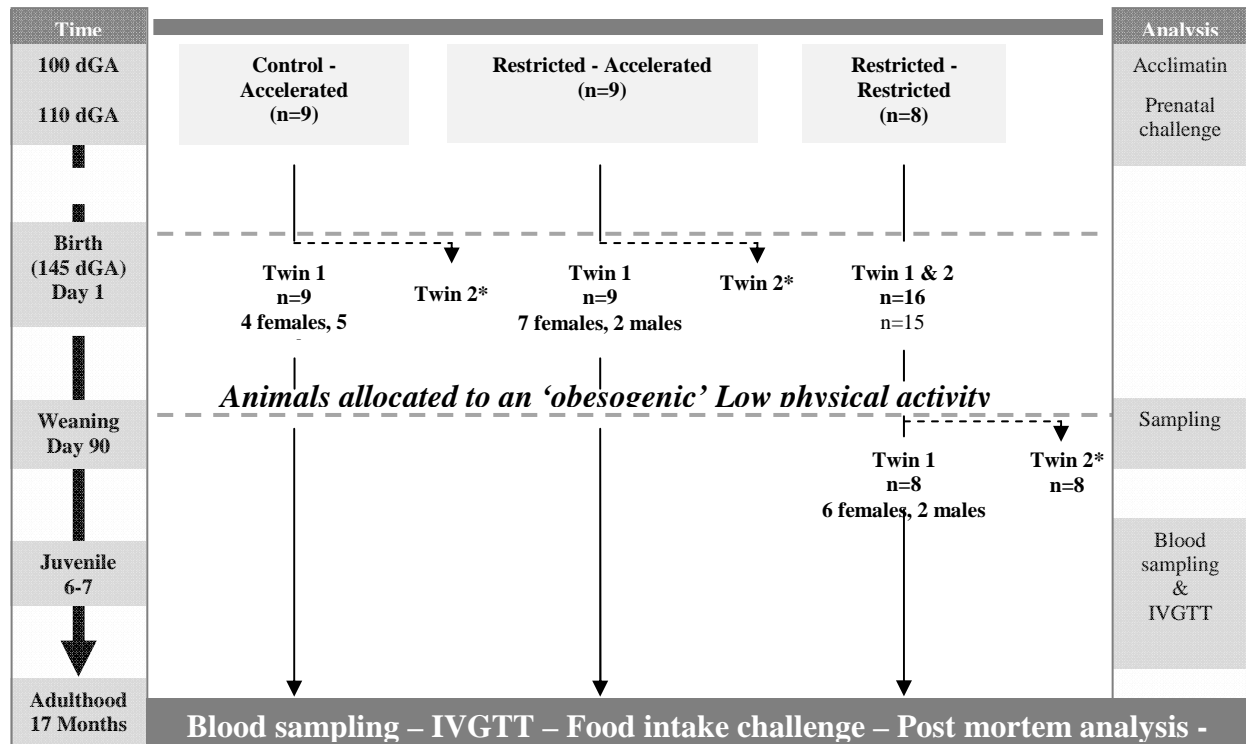


Figure 1: Summary of experimental design.

Results

Table 2: Impact of maternal nutrient restriction in late gestation and of postnatal growth velocity in the 17 months old offspring (effect compared to control animals)

	RA	RR
	<i>Low birth weight (IUGR)</i>	
Postnatal growth	Accelerated	Standard
Postweaning growth	Accelerated	Accelerated
Insulin sensitivity	Insulin resistance Hyperinsulinism Glucose intolerance	Insulin resistance Hyperinsulinism Glucose intolerance
Appetite control	Hypophagia hyperleptinemia Up regulation of central energy sensing Down regulation of gastric mTOR, SIRT1, UCP2 and CPT1	Normal appetite Normal leptin Up regulation of central energy sensing Down regulation of gastric mTOR, SIRT1, UCP2 and CPT1 Upregulation of AMPk dependent pathway and mitochondrial biogenesis
	Altered cellular proliferation in the gastric mucosa	Normal cell proliferation of the gastric mucosa

Clearly we succeeded in inducing IUGR associated with early postnatal growth acceleration. We found that such a change in growth during the perinatal period had a long term impact on the control of whole body energy metabolism. Although standardisation of early postnatal growth had no effects on long term insulin sensitivity and glucose tolerance, it protected the RR offspring against hyperleptinemia. One of the main results was that beside a similar effect on the programming upon the central control of appetite in the hypothalamus, modest early growth in IUGR offspring may help prevent long term adverse outcomes. Such adaptations may have been driven by the impact of early milk intake in gut development in which AMPK-dependant energy sensing pathway was up regulated. In addition we found a long term impact on cellular proliferation in the adults suggesting programming of oxidative stress.

Conclusion of 5.5 years of research:

Critical pioneering findings have been established with marked progresses in the understanding of the impact of early nutrition have been made possible and we further demonstrate the importance of studying such essential questions in large animal models that allow more direct translation into human practices. The take home message of this project could be that “the early energetic environment primarily acts through the setting of cellular energy homeostasis via the determination of energy sensing pathways”. This can be of pivotal importance for influencing the long term feature of inadequate early growth associated with IUGR, premature birth and maternal obesity. It is in fact known that specific nutritional compounds such as LC PUFA, resveratrol and other antioxidant may act directly to modify energy sensing pathways suggesting potential therapeutic interventions that can now be tested.

Key References

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