Integrating metabolic networks and growth biomarkers to unveil potential mechanisms of obesity

Andrea Cremaschi 1,*, Maria De Iorio1,2,3,4, Narasimhan Kothandaraman1, Fabian Yap5, Mya Tway Tint1, Johan Eriksson1,2

1Singapore Institute for Clinical Sciences, A*STAR, Singapore
2Yong Loo Lin School of Medicine, National University of Singapore, Singapore
3Division of Science, Yale-NUS College, Singapore
4Department of Statistical Science, University College London, UK
5Department of Paediatrics, KK Women’s and Children’s Hospital, Singapore
cremaschia@sics.a-star.edu.sg

Abstract

The prevalence of chronic non-communicable diseases such as obesity has noticeably increased in the last decade. The study of these diseases in early life is of paramount importance in determining their course in adult life and in supporting clinical interventions. Recently, attention has been drawn on approaches that study the alteration of metabolic pathways in obese children. In this work, we propose a novel joint modelling approach for the analysis of growth biomarkers and metabolite concentrations, to unveil metabolic pathways related to child obesity. Within a Bayesian framework, we flexibly model the temporal evolution of growth trajectories and metabolic associations through the specification of a joint non-parametric random effect distribution which also allows for clustering of the subjects, thus identifying risk sub-groups. Growth profiles as well as patterns of metabolic associations determine the clustering structure. Inclusion
of risk factors is straightforward through the specification of a regression term. We demonstrate the proposed approach on data from the Growing Up in Singapore Towards healthy Outcomes (GUSTO) cohort study, based in Singapore. Posterior inference is obtained via a tailored MCMC algorithm, accommodating a nonparametric prior with mixed support. Our analysis has identified potential key pathways in obese children that allows for exploration of possible molecular mechanisms associated with child obesity. Dirichlet Process; Gaussian process; Graph-based clustering; Graphical models; Longitudinal data; Metabolomics

1 Introduction

Obesity is a major risk factor for chronic non-communicable diseases such as type-2 diabetes (T2D), metabolic syndrome and cardiovascular diseases. The prevalence of obesity has reached epidemic proportions worldwide and has tripled between 1975 and 2016. In particular in 2016, 39% of adults were overweight and 13% obese [WHO, Accessed: 01-06-2021]. Prevalence of obesity in children has also escalated over the years, increasing from 4% in 1975 to over 18% in 2016 among children and adolescents aged 5-19 years [WHO, Accessed: 01-06-2021]. Overweight or obesity in childhood is critical as it often persists into adulthood due to both physiological and behavioural factors. Indeed, childhood obesity is associated with increased risks of glucose intolerance, hypertension, dyslipidaemia, insulin resistance, and T2D in adulthood [Freemark, 2010]. Therefore, preventing childhood obesity can help disrupt the incidence of metabolic diseases in later life. Several different mechanisms such as insulin resistance, inflammation and metabolic dysregulation mediate the link between obesity and the risk of metabolic diseases. Indeed, there is a complex interplay between genetic determinants, behavioural and environmental factors which contribute to obesity. Yet, relatively little is known regarding its underlying pathophysiology.

In this work, we investigate the complex metabolic pathways in childhood obesity, combing metabolite concentration data (as measured by NMR spectroscopy) with more traditional clinical makers measuring the growth of the children. Metabolites are small molecules that participate in metabolic reactions and are involved in biochemical pathways associated with metabolism in health and disease [Ellul et al., 2019]. As the prevalence of obesity is rapidly increasing in children and adolescents, metabolomics is a powerful
tool to uncover underlying biological mechanisms, to unravel genetic and environmental interactions, to identify therapeutic targets, to facilitate early detection of metabolic diseases and to monitor disease progression. Applying metabolomic techniques in relation to childhood obesity could pave a way in defining biomarkers of future metabolic risk and targets for early detection and intervention. Previous studies in adults have consistently identified metabolic signatures associated with obesity, insulin resistance and T2D. For example, previous research has reported associations between obesity and elevated plasma concentrations of amino acids such as branched-chain amino acids (BCAA, leucine, isoleucine and valine), aromatic amino acids (AA, phenylalanine and tyrosine), gluconeogenesis intermediates and glutamine metabolism which are linked to inflammation of white adipose tissue in obesity [Würtz et al., 2013, Wang et al., 2011, Newgard et al., 2009, Roberts et al., 2020, Takashina et al., 2016, Petrus et al., 2020]. Although the metabolomic literature on adults suffering from obesity, insulin resistance or T2D presents convincing results, metabolic changes related to obesity in younger populations have been poorly identified and findings are often inconsistent and different from those in adult populations [Balikcioglu and Newgard, 2018]. For instance, in contrast to adults, a study on children and adolescents in Germany does not report association between BCAA levels and obesity [Wahl et al., 2012], while concentrations of medium- and long-chain acylcarnitines (C12:1 and C16:1) are reported as higher in obese as compared to normal weight children. This latter association has been replicated in adults. Moreover, further 12 metabolites (glutamine, methionine, proline, nine phospholipids) were found to be significantly altered in obese children. The identified metabolite markers are indicative of oxidative stress and of changes in sphingomyelin metabolism, in β-oxidation, and in pathways associated with energy expenditure. Contrary to adults, Mihalik et al. [2012] show that obese and diabetic children present no evidence of defects in fatty acid or amino acid metabolism as compared to their normal weight peers. In the cohort study Project Viva [Oken et al., 2015], BCAA concentrations have been reported to be higher in obese versus lean children aged 6–10 years [Perng et al., 2014]. Similarly, Butte et al. [2015] report that the concentrations of BCAAs, glutamate, lysine, tyrosine, phenylalanine, and alanine significantly increase in obese children as compared with normal weight children. However, other amino acids such as asparagine, aspartate, glycine, serine, and histidine levels decrease. These results indicate that childhood obesity influences the composition of the serum metabolome,
pointing towards potential biomarkers.

The aim of this work is to identify metabolic signatures of obesity in children with different trajectories of adiposity from 5 to 9 years of age, using data from the Growing Up in Singapore Towards healthy Outcomes (GUSTO) prospective cohort study [Soh et al., 2014]. Metabolome analysis is particularly relevant in Asian populations where the risk of metabolic diseases is higher than in the Western population [Misra and Khurana, 2011] and the GUSTO cohort study provides an optimal platform. GUSTO is a deeply phenotyped prospective cohort involving Singaporean mothers and their children, started in 2009 (pre-natal) by recruiting mothers at the first trimester of pregnancy. A wealth of information is available on both mothers and children. In this work we focus on growth profiles of children and their relationship with metabolic outcomes, as well as more traditional risk factors such as demographics and clinical biomarkers. To this end, we propose a joint model for the growth trajectories and anthropometric measures of children from birth to 9 years of age and a set of metabolites measured at age 8 years in children. The anthropometric indicators are obtained with Quantitative Magnetic Resonance (QMR) techniques [Chen et al., 2018], recording the percentages of fat and lean mass in the children’s body excluding the contribution from the bones, and by height/weight measurements, used to compute the standardised body mass index (Z-BMI). These growth indicators are recorded at different time points in the children’s development: every year from age 5 to 9 for the QMR measures, and at 21 unequally spaced time points for the Z-BMI. These data pose challenges to the statistical analysis given their dimension and missing rates for some of the variables, as not all subjects took part to follow-up visits.

The main contribution of this work is to provide a joint model for three growth markers and metabolic associations, which allows for data-driven clustering of the children and highlights metabolic pathway involved in child obesity. To this end, in a Bayesian framework, we specify a joint non-parametric random effect distribution on the parameters characterising the longitudinal trajectories of obesity and the graph capturing the association between metabolites. The choice of a nonparametric random effect distribution allows for extra flexibility, heterogeneity in the population as well as data-driven clustering of the subjects.

The paper is structured as follows: Section 2 introduces the proposed approach for the joint modelling of growth trajectories and metabolic associations; Section 3 presents posterior inference results obtained when applying
the proposed methodology to the GUSTO data, highlighting cluster-specific
growth evolution as well as differences in metabolic associations. Section
4 concludes the paper with a discussion. A Supplementary Material file is
available, containing additional Figures, details on the MCMC algorithm and
a comprehensive description of the features of the dataset.

2 Joint modelling of growth trajectories and
metabolites

The main goal of the analysis is to combine information from the longitudinal
responses (i.e. the growth curves) and the metabolic variables (observed
only at one time point) to gain a better understanding of the children’s
development. In particular, we develop a joint model where the longitudinal
outcomes are flexibly modelled via a nonparametric mixture of Gaussian
Processes (GP), while we exploit tools from the Gaussian Graphical Model
(GGM) literature to introduce information from metabolites and their inter-
dependencies. These two components are then linked hierarchically by the
specification of a suitable prior distribution.

Let \( Y = \{ Y_t : t \in \mathbb{R}^+ \} \) be a stochastic process indexed over the positive
real line, in this work representing the time component, and taking values
in \( \mathbb{R} \). Let the realisations of such process be the vectors \( Y_i = (Y_{i1}, \ldots, Y_{in}) \),
observed at times \( t = t_1, \ldots, t_n \) (not necessarily equidistant) for subjects
\( i = 1, \ldots, N \). A possible modelling strategy consists of assuming a Gaus-
sian Process to model each trajectory over time, resulting in a multivariate
Gaussian likelihood distribution for the vectors \( Y_i \). This modelling strat-
egy is flexible and allows for efficient computations, but would not be able
to account effectively for subject heterogeneity, typical of medical studies.
This is evident when observing the empirical distribution of the growth in-
dicators, presenting skewness and heavy tails at the different observed time
points, shown in Figures 7 and 8 in Supplementary Material. A possibility
to overcome these limitations is to model the observations using a mixture
of multivariate Gaussian distributions, adopting a flexible mixing measure
which in turn involves a suitable temporal dependence structure such as the
one offered by the GP. In order to do so, we extend the modelling strategy
introduced in Gelfand et al. [2005] to our context and thus assume the distri-
bution of the vectors \( Y_i \) to be an infinite mixture of multivariate Gaussians
where the mixing measure is given by a Dirichlet Process (DP) prior, centred around a stationary GP. The DP was firstly introduced by Ferguson [1973], and defines a probability measures over the space of probability distributions. A constructive definition of the DP is provided by the stick-breaking representation [Sethuraman, 1994]:

\[ P(\cdot) = \sum_{j=1}^{\infty} w_j \delta_{\psi_j}(\cdot) \]

where \( \delta_x(\cdot) \) is the Dirac’s delta measure taking value 1 at the location \( x \), and 0 otherwise. The infinite sequence of locations \( \{\psi_j\}_{j=1}^{\infty} \) is an i.i.d. sample from a centering measure \( P_0 \), while the infinite sequence of weights \( \{w_j\}_{j=1}^{\infty} \) is constructed in the following way:

\[ w_j = v_j \prod_{i<j} (1 - v_i), \quad j = 2, 3, \ldots, \quad w_1 = v_1 \]

where \( v_1, v_2, \ldots \overset{iid}{\sim} \text{Beta}(1, \alpha) \)

where the mass parameter \( \alpha > 0 \) controls the dispersion of the process around \( P_0 \). An important feature of the DP making it appealing in applications is its almost sure discreteness, implying the possibility of modelling ties in the sample from this distribution, inducing a partition of the indices \( \{1, \ldots, N\} \). When used as a mixing measure, this implies a partition of the subjects sharing the same value of the mixing parameter. As pointed out in Gelfand et al. [2005], this yields a flexible distribution for the vectors \( Y_1, \ldots, Y_N \), which is non-Gaussian and non-stationary, but retains the advantageous mathematical and computational properties of the GP (see Supplementary Section 2 for the details of the algorithm). This, in conjunction with the computational tractability of the DP, allows for efficient posterior inference of the proposed approach. In the application under study, we have \( S = 3 \) distinct processes, namely the percentages of fat and lean mass in the body and the standardised body mass index (Z-BMI) values, and therefore we model the vectors \( Y_i^{(s)} = (Y_{i1}^{(s)}, \ldots, Y_{im_s}^{(s)}) \), for \( s = 1, \ldots, S \) and \( i = 1, \ldots, N \), with a process-dependent covariance kernel in the GP:

\[
K_{s_1s_2}^{t_1t_2} (\sigma^2, \phi^2, \eta^2, \xi_{s_1}, \xi_{s_2}) = \text{Cov} \left( Y_{t_1}^{s_1}, Y_{t_2}^{s_2} \right) = \xi_{s_1} \xi_{s_2} \sigma^2 e^{-\frac{(t_1-t_2)^2}{\phi^2}} + \eta^2 1_{\{s_1=s_2, t_1=t_2\}}
\]
with \( t_i = 1, \ldots, n_{s_1}, t_j = 1, \ldots, n_{s_2}, s_1, s_2 \in \{1, \ldots, S\} \) and \( \eta^2 \) is the nugget parameter, present only on the diagonal elements of the kernel. The covariance kernel presents similar features to the widely-used exponential kernel, accounting for the presence of multiple-processes through the scaling factors \( \xi_s \), for \( s = 1, \ldots, S \). In particular, it includes the positive coefficient \( \sigma^2 \) calibrating the amount of variability in the data, as well as \( \phi^2 \) regulating the impact of the distance between time points on the correlation between observations, and it is stationary since it only depends on time distance \( |t_i - t_j| \).

We will indicate by \( K = \begin{bmatrix} K_{s_1s_2}^{t_i t_j} \end{bmatrix} \) the covariance matrix obtained from this kernel at the observed time points and processes. The choice of random effect distribution will be discussed later.

The second component of the data is represented by the metabolite concentrations measured at year 8. These type of data are usually modelled via multivariate distributions, most commonly Gaussian. Of particular interest in this analysis is the relationship between the observed metabolites, quantifiable by their correlation structure, with the aim of understanding the dependencies between them and their role in the activation of specific metabolic pathways. We approach this problem by modelling the vectors of metabolites borrowing from the GGMs literature. In this setting, let \( M = (M_1, \ldots, M_{p_M}) \in \mathbb{R}^{p_M} \) be a vector of \( p_M \) metabolites and let \( G = (V, E) \) be a graph defined over the set of nodes \( V = \{1, \ldots, p_M\} \) and with edge set \( E \subset \{(h, k) \in V \times V | h < k\} \) such that if there is a connection between the nodes \( h \) and \( k \), then \((h, k) \in E\). The graph \( G \) can be represented by an adjacency matrix \( A = (A_{hk}) \) such that \( A_{hk} = 1 \) if and only if \((h, k) \in E\). The graph \( G \) is used to represent the correlation structure of the vector of metabolites \( M \), exploiting the property that two elements of the vector are conditionally independent given the rest if and only if the precision matrix is null at the corresponding position [Dempster, 1972]. A zero in the precision matrix corresponds to a zero in the adjacency matrix, which results in the absence of an edge in the graph (and vice-versa, an edge in the graph corresponds to a non-zero element in the precision matrix). The vectors of metabolites are modelled using a multivariate Gaussian distribution with precision matrix \( \Omega_G \), whose prior distribution is defined conditionally to the graph structure \( G \). The standard conditionally conjugate prior distribution is the G-Wishart [Roverato, 2002] with \( \nu \) degrees of freedom, scale matrix \( \Psi \) and graphical encoding \( G \), denoted here as G-Wishart\( (\Omega_G | \nu, \Psi, G) \) and with
p.d.f.:
\[
p(\Omega_G|\nu, \Psi, G) = \frac{1}{I_G(\nu, \Psi)}|\Omega_G|^{(\nu-2)/2} \exp \left\{-\frac{1}{2} \text{tr}(\Psi \Omega_G)\right\} \mathbb{I}(\Omega_G \in \mathbb{P}_G)
\]
where \( \mathbb{P}_G \) indicates the cone of positive-definite square matrices with non-zero entries corresponding to the edges of the graph \( G \), while \( I_G(\nu, \Psi) \) is a normalising constant which is finite if \( \nu > 2 \) and \( \Psi \) is positive-definite [Diaconis and Ylvisaker, 1979]. The prior distribution over the graph structure \( \pi(dG|d) \) is given by the product of Bernoulli priors on each edge with inclusion probability \( d \in (0, 1) \):
\[
\pi(G|d) \propto d^{|E|}(1-d)^{|p_M|/2}^{-|E|}
\]
with \( |E| \) being the number of edges in the graph \( G \) and \( \binom{p_M}{2} \) the total number of possible edges given the dimension \( p_M \).

As mentioned earlier, the aim of this work is to study the relationship between the growth indicators and the metabolite values jointly, still allowing for flexibility. Thus, we specify a joint prior distribution for the hyperparameters of the two sub-models, thus capturing the dependency between the longitudinal and metabolic dimensions. In particular, we specify a joint distribution on the random effect vector \( \theta \) in the longitudinal part of the model, on the precision matrix \( \Omega_G \) and embedded graph \( G \). The random effect vector \( \theta \) of length \( p_Y = \sum_{s=1}^{S} n_s \) is obtained by concatenating the random intercept vectors relative to each longitudinal process, s.t. \( \theta = (\theta^{(1)}, \ldots, \theta^{(S)}) \). Let \( \psi_i = (\theta_i, \Omega_{G_i}, G_i) \) for \( i = 1, \ldots, N \) be the array of subject-specific parameters of interest. We specify a the DP prior on the arrays \( \psi_1, \ldots, \psi_N \). We
obtain the following model:

\[ Y_i^{(s)} | \theta_i^{(s)}, \beta_{s}, X_i^Y, \tau_s^2 \sim \text{N}_{n_s}(Y_i^{(s)} | \theta_i^{(s)} + \beta_{s} X_i^Y, \text{diag}(1/\tau_s^2)) \]

\[ \beta^Y = [\beta_Y^1, \ldots, \beta_Y^{S}] \sim \text{MN}_{p_Y \times q_Y}(\beta^Y | 0, \mathbb{I}_{p_Y}, \mathbb{I}_{q_Y}) \]

\[ \tau_s^2 \sim \text{inv-gamma}(\tau_s^2 | 3, 2) \]

\[ M_i | \beta^M, X_i^M, \Omega_G \sim \text{N}_{p_M}(M_i | \beta^M X_i^M, \Omega_{G_i}) \]

\[ \beta^M \sim \text{MN}_{p_M \times q_M}(\beta^M | 0, \mathbb{I}_{p_M}, \mathbb{I}_{q_M}) \]

\[ \Omega_{G_i} | \nu, \Psi, G_i \sim \text{G-Wishart}(\Omega_G | \nu, \Psi, G_i) \]

where \( N_{p}(Y | \mu, \Omega) \) is the \( p \)-dimensional Gaussian distribution for the vector \( Y \) with mean \( \mu \) and precision matrix \( \Omega \). where the DP prior is imposed on the \( p_Y \)-dimensional concatenated random effects \( \theta_1, \ldots, \theta_N \) in order to introduce dependencies within and between processes via the definition of the GP kernel in (1), on the graph and on the precision matrix. Finally, a conjugate Gaussian prior distribution is imposed on the mean vector of the GP in the centring measure \( P_0 \). We specify a joint Matrix-Normal prior distribution for the column-wise concatenated matrix of coefficients \( \beta^Y \in \mathbb{R}^{p_Y \times q_Y} \), where \( p_Y = \sum_{s=1}^{S} n_s \) and \( q_Y \) is the number of covariates used. The prior distribution for the matrix of coefficients \( \beta^Y \) has zero mean matrix and identity covariance matrices. Notice that this setting implies a different regression coefficient being estimated at each observed time point for each covariate included in the model, allowing to capture changes in the temporal effect of risk factors. We write \( \text{gamma}(x | a, b) \) and \( \text{inv-gamma}(x | a, b) \) to denote the Gamma and the inverse-Gamma distribution for \( x > 0 \) with means \( a/b \) and \( b/(a - 1) \), respectively. We allow for covariate effects on the metabolite concentrations via the terms \( \beta^M X_i^M \), for \( i = 1, \ldots, N \) and use a prior specification for the matrix of coefficients analogous to the one for \( \beta^Y \).
The main contribution of the proposed approach is the ability to cluster individuals based on their growth profiles and metabolic associations through joint modelling of longitudinal and multivariate markers. Modelling of multiple graphs has been proposed before in the Bayesian framework [e.g. Peterson et al., 2015, Tan et al., 2017, Shaddox et al., 2020] with groups specified a priori, but not in the context of graph-based (unsupervised) clustering, which is achieved by our modelling strategy. To the best our knowledge, this approach has not been proposed in the statistical literature before. This modelling choice implies that the base measure $P_0$ of the DP is a mixed measure, due to the presence of the graph structure in the sample from the DP. In particular, $P_0$ is defined on the product space $\mathbb{R}^{pY} \times \mathbb{P}_G \times \mathcal{G}_{PM}$, where $\mathcal{G}_{PM}$ represents the space of all possible graphs of dimension $p_M$. In general, when using the DP, the base measure $P_0$ is chosen to be non-atomic, allowing for the computation of the predictive distributions. However, examples of applications requiring a mixed base measure in the specification of the DP are found in the literature [among others, see MacLehose et al., 2007, Dunson et al., 2008, Guindani et al., 2009]. As reported in existing work [see, for instance, Kim et al., 2009, Canale et al., 2017], the mixed measure setting is not problematic in the case of the DP, since the predictive distributions remain unchanged, and thus posterior inference via a Pólya urn algorithm can still be achieved, and we exploit this result. One of the main features of this approach is that the unique values associated with the clusters are not necessarily different, due to the mixed nature of the centering measure $P_0$. For instance, in our setting, ties between the graph structures associated with the clusters can be observed (but not among the random effects or precision matrices).

It is common (especially in medical/epidemiological research) that the set of metabolites $M$ is used as predictor, often in a regression setting. From this perspective, model (3) shows similarities with existing Bayesian non-parametric literature on product partition models with covariates [PPMx, Müller et al., 2011]. In particular, it can be shown that the marginal distribution of the random partition induced by the DP measure can be factorised in terms including the covariates (i.e., the metabolites) within each cluster. Let $\rho_N = \{C_1, \ldots, C_{K_N}\}$ be the partition of the indices $\{1, \ldots, N\}$ induced by the sample $(\psi_1, \ldots, \psi_N)$, where we indicate by $C_j$ the $j$-th cluster of size $n_j = |C_j|$, for $j = 1, \ldots, K_N$. Considering the metabolites observations $M$ as covariates, and following the PPMx approach, we have that the prior for
the partition $\rho_N$ is:

$$p(\rho_N \mid M) = V(N, K_N, \alpha) \prod_{j=1}^{K_N} C(C_j) \mathcal{H}(M_j^*)$$  \hspace{1cm} (4)$$

where $C(C_j)$ is the **cohesion** of the $j$-th cluster $C_j$, $\mathcal{H}(M_j^*)$ is the **similarity** of the metabolites for the subjects belonging to cluster $j$, denoted as $M_j^* := \{M_i : i \in C_j\}$, for $j = 1, \ldots, K_N$, and $V(N, K_N, \alpha)$ is a constant derived from the marginal distribution of the partition $\rho_N$. Information about the partition is included via the cohesion function $C$, while the contribution of the covariates to the clustering structure is expressed via the similarity function $\mathcal{H}$, facilitating the clustering of subjects with similar covariates. Under our modelling assumptions it can be shown that (4) is given by:

$$p(\rho_N \mid M) \propto p(\rho_N) p(M \mid \rho_N) =$$

$$V(N, K_N, \alpha) \prod_{j=1}^{K_N} \Gamma(n_j) \int \left( \prod_{i \in C_j} p(M_i \mid \Omega G_j, G_j) \right) P_0(d\Omega G_j, dG_j)$$ \hspace{1cm} (5)

The expression of $V(N, K_N, \alpha)$ can be derived from the eppf of the DP with general base measure, following Argiento et al. [2019], Pitman [2006]. The integral can be simplified further by integrating out the precision matrix $\Omega G_j$, which is a-priori distributed according to a $G$--Wishart($\nu, \Psi, G$), yielding to the ratio of normalising constants in (2):

$$p(\rho_N \mid M) \propto p(\rho_N) p(M \mid \rho_N) =$$

$$\alpha^{K_N} \frac{\Gamma(\alpha)}{\Gamma(\alpha + N)} \prod_{j=1}^{K_N} \Gamma(n_j) \sum_{G \in \mathcal{G}_{p_M}} \frac{I_G(\nu + n_j, \Psi^{(j)})}{I_G(\nu, \Psi)} \pi(G)$$ \hspace{1cm} (6)

where $\Psi^{(j)} = \Psi + \sum_{i \in C_j} M_i M_i^\top$, and where the part relative to the precision matrix has been integrated out, leaving only the sum over all possible graphs of size $p_M$. The latter sum is finite and theoretically could be computed for each cluster. In principle, this would allow us to devise an algorithm similar to the one proposed in the original PPMx models. In practice, due to its computational burden, such approach is unfeasible for graphs of even moderate sizes. As such, we opt for a conditional algorithm (see Supplementary Section 2), which does not marginalise over the random measure. The algorithm is based on Metropolis-within-Gibbs sampling, with adaptive steps...
for those parameters which are non-conjugate in the proposed model. The update of the DP parameters follows a Pólya Urn scheme for non-conjugate models [Neal, 2000, Favaro and Teh, 2013], adjusted for the presence of the non-conjugate graphical structure. The updates of the graph and precision matrix within each cluster are tackled using the Birth-and-Death algorithm of Mohammadi and Wit [2015].

3 Posterior inference

In this Section we present the application of the proposed modelling strategy to the data from the GUSTO cohort. The longitudinal data are composed of the fat and lean percentages measured at years 5 to 9 of the children, together with the Z-BMI values from birth to year 9 at non-equally spaced time points, such that $n_1 = n_2 = 5$ and $n_3 = 21$. The size of the concatenated vectors of growth indicators is $p_Y = 31$. The list of $p_M = 35$ metabolites measured at year 8 is reported in Supplementary Table 1. We apply a logit transformation to the fat and lean percentages, in order to map the observations to the real line, while the Z-BMI is standardised. Many of the metabolites present different ranges of values, skewness, and non-normality. In order to correct for these features, we apply a Box-Cox transformation to each metabolite individually [Sakia, 1992], and then center and re-scale the observations such that their component-wise empirical means and variances are equal to 0 and 1, respectively.

Additionally to longitudinal and metabolic information, demographic variables and other clinical characteristics, such as ethnicity, pre-pregnancy maternal BMI, findings from oral glucose tolerance test (OGTT) and gender of the child, are available. The full list of covariates is reported in Supplementary Table 2. The same set of covariates is used in both regression components of the model, therefore $X^Y = X^M$ and $q_Y = q_M = 14$. The covariates present a relatively low percentage of missing values, as reported in the last column of Supplementary Table 2, which are imputed using the R package mice. After imputation, the continuous covariates are centred and scaled to have mean zero and variance one. We do not include any intercept term in the regression components.

We set the hyperparameters for $\tau_s^2$, $\xi_s$ for $s = 1, \ldots, S$ and $\sigma^2$, $\phi^2$ and $\eta^2$ so that their prior means and variances are both equal to 1; we follow Jones et al. [2005] and set $d = 2/(p_M - 1) \approx 0.06$ inducing sparsity in the
graph structure; applying Eq. (14) and (15) in Jara et al. [2007], we fix the mass parameter of the DP to $\alpha = 0.18$, yielding $\mathbb{E}(K_N) = 1.31$ and $\text{Var}(K_N) = 1.13$; the hyperparameters of the centering measure $P_0$ are set as $\mu_\theta = 0$, $\nu = p_M + 2 = 37$ and $\Psi = 10\|p_M\|$. To fit the model, the algorithm is run for 50000 iteration after an initial short burn-in period of 100 iterations used to initialise the adaptive components of the algorithm. Then, after a longer burn-in of 40000 iterations, a posterior chain of 5000 iterations is saved by thinning the remaining 10000 every 2.

### 3.1 Posterior Inference on Clustering Allocation

An advantage of the proposed approach is the ability of provide posterior inference on the clustering of the subjects, therefore allowing for the identification of groups of children characterised by specific growth trajectories and metabolic associations. As posterior estimate of the random partition $\rho_N$, we report the clustering configuration minimising the Binder’s loss function [Binder, 1978], which corresponds the expected loss derived from the two possible misclassification errors, occurring when a pair of subjects is erroneously clustered together or separately. The use of Binder’s criterion yields a partition composed of three clusters of sizes 124, 71 and 32, respectively. We refer to it as the Binder partition, and label the clusters by their decreasing size. The number of clusters identified by Binder’s method coincides with the posterior mode of the random variable counting the number of clusters, reported in Figure 1 of Supplementary Material, together with the posterior co-clustering probability for each pair of subjects. The results show little uncertainty in the distribution of the number of clusters and cluster assignment. In order to visualize the data and their estimated partition, we display the mean of the longitudinal growth data within each cluster in Figure 1. As it is evident from the top panel of the Figure, the longitudinal growth patterns display an intuitive clustering structure, separating children with low fat and high lean percentages from those with high fat and low lean percentages. The biggest cluster is composed of children with moderate values of fat and lean percentages. The Z-BMI curves follow a similar pattern, but only after 15 months of age.

As exploratory analysis, we also look at the empirical mean of the metabolite concentrations within each cluster, shown in Figure 2. The three clusters exhibit different mean patterns, highlighting differences in the three groups also on a metabolic level. It is evident that the mean level of almost all
metabolites in Cluster 1 is around zero, while in the smaller clusters the distributions are centred away from zero and often in opposite directions between them, supporting the hypothesis that they capture different metabolic mechanisms.

Summarizing the posterior distribution of the latent variables $\psi_1, \ldots, \psi_N$ is not trivial, due to the existence of label-switching problems arising when working in a Bayesian nonparametric setting. Therefore, in order to understand the results of the clustering analysis, we run an additional MCMC chain, with the same number of iterations, after fixing the random partition $\rho_N$ to be equal to the Binder partition. By doing so, we are able to provide the posterior distribution of the values of $\psi^*$ within each of the three clusters. We show in Figure 3 the posterior estimates of the graph structures within each of the three clusters (the corresponding estimates of the precision matrices are shown in Supplementary Figure 2). The estimates are the median graphs, obtained selecting the edges whose posterior inclusion probability is greater than 0.5 [Barbieri and Berger, 2004]. As it can be observed, the number of estimated connections in the graphs is highest in Cluster 1, as well as the intensity of the entries of the corresponding precision matrix (see Supplementary Figure 2). In all clusters, we can identify a group of metabolites linked together, corresponding to fatty acids, phosphoglycerides, apolipoproteins and cholesterol (see Figure 3 and Supplementary Figure 2, top left corners), while associations between smaller groups of metabolites involving some amino acids and ketone bodies present different patterns in the three clusters. We provide a discussion and suggest a biological interpretation of such differences in Section 3.3.

### 3.2 Posterior Inference on Regression Coefficients

We now discuss posterior inference on the regression coefficients for the three responses in (3). As described in Section 2, we estimate the effect of the covariates on each growth process at different time points. We report in Supplementary Figures 3, 4 and 5 the posterior means and 95% credible intervals (CI) for the entries of the matrix $\beta^Y$. We consider as relevant those predictors whose 95% CI does not contain the value zero, highlighted in red in the Figures. Interestingly, the covariates which influence the fat and lean percentages at most of the five time points include gestational age, gender of the children, maternal pre-pregnancy BMI and ethnicity, confirming existing results obtained from the same cohort [Aris et al., 2017, Ong et al., 2021].
Figure 1: Posterior mean trajectories within each cluster identified by the Binder’s partition. Each row represent a growth indicator (Fat/Lean/Z-BMI), while each column refers to a cluster.

Moreover, some covariates have different effects across time, such gender and highest education degree of the mother. Similar results on the effect of these covariates on the evolution of the Z-BMI trajectories are also reported in Supplementary Figure 5. Results on the effects of covariates on metabolites levels are discussed in Supplementary Material Section 1.2.

### 3.3 Differential network analysis

To quantify the differences between the cluster-specific networks presented in Figure 3, we estimate a differential network [Valcárcel et al., 2011, Tan et al., 2017], providing an approach based on the joint posterior distribution of the graph structures within each cluster to establish whether the differences
among the cluster-specific are statistically relevant. We perform three pairwise comparisons of the networks characterising the three clusters estimated by minimising the Binder’s loss function. A differential network only shows those connections for which the absolute difference between the posterior edge inclusion probabilities of two graphs is greater than 0.9. Specifically, for two clusters $k_1$ and $k_2$ we require that $|\hat{\pi}^{k_1}_{ij} - \hat{\pi}^{k_2}_{ij}| > 0.9$, where $\hat{\pi}^k_{ij}$ is the posterior inclusion probability of the edge between nodes $i$ and $j$ in cluster $k$, estimated using the MCMC output obtained after fixing the random partition $\rho_N$ to the Binder partition, as previously done in the context of cluster-specific network estimation (see Figure 3). The resulting differential networks are shown in Figures 4, 5 and 6 (left panels). The differential networks are characterised by a different number of edges, with the one between Cluster 1 and 2 showing more differential connections (see Figures 4(a), 5(a) and 6(a)).
However, there are key metabolites common to all three differential networks: (i) some amino acids such as glycine, leucine and tyrosine; (ii) glycoprotein acetyllys; (iii) docosahexaenoic acid (DHA, an omega 3 fatty acid); (iv) acetate. Lipids (HDL and triglycerides) are involved only in the first two differential networks.

The roles of the metabolites involved in the differential networks can be further explored by performing an Ingenuity Pathway Analysis [IPA, Krämer et al., 2014]. IPA generates network maps between molecules (see Figures 4(b), 5(b) and 6(b)), and compares them with multiple metabolic pathways (i.e. the linked series of chemical reactions occurring within a cell) available in the literature, assigning a score to each comparison. The scores are generated based on the negative logarithm of the significance level obtained by performing Fisher’s exact hypergeometric test when comparing the estimated differential network and the known pathways in the IPA library. For canonical pathway analysis, values of $-\log(p\text{-value}) > 2$ are used to detect significant activation. Figure 7 shows a summary of the pathways identified as significant via the IPA methodology. We identify a total of 18 pathways through IPA that are common between the three differential networks, denoted by 1-2, 2-3, and 1-3 in Figure 7. Of the 18 pathways, key ones are tRNA charging and biosynthetic pathways for amino acid biosynthesis. The
pathways associated with biosynthesis include acidic amino acid glutamine, aliphatic simple amino acids alanine, glycine, and aromatic amino acid tyrosine and its precursor phenylalanine undergoing degradation. The commonality among the three groups indicates an active amino acid biosynthesis machinery with the initiation from tRNA charging which is a requisite for translation and transcription of protein biosynthesis through the binding of amino acids. It has been previously shown that altered tRNA aminoacylation, modification and fragmentation are associated with β-cell failure, obesity and insulin resistance [Arroyo et al., 2021]. All the amino acid pathways common to the three differential networks are associated with obesity as well as metabolic syndrome. In Figure 7, comparisons between differential networks 1-3 (green) and 2-3 (yellow) shows clear patterns of insulin metabolism as well as activation of signalling pathways related to dysglycemia. The pattern could provide evidence of early events leading to insulin resistance as well as transition to a hyperglycemic state and onset of obesity as evident from IL-12 [Interleukin 12, Nam et al., 2013], apelin (a peptide) [Dray et al., 2008] and growth hormone (GH) signalling [Høgild et al., 2019]. Studies show IL-12 family cytokines as prospective regulators that could cause insulin resistance due to obesity in tissues and plasma [Nam et al., 2013]. The comparison between Clusters 1 and 2 (blue) does not yield any unique pathway via the IPA. This is important since it refers to the comparison between normal and low Z-BMI trajectories (see Figure 1). Comparing common pathways between the differential networks 1-2 and 2-3 shows only one common pathway, i.e. phenylalanine degradation I (Aerobic), indicating biosynthesis of tyrosine which is a feeder molecule for acetoacetate involved in the synthesis of acetyl-CoA (acetyl coenzyme A), which is important for dietary intake and energy balance. Comparison between Clusters 2 and 3 shows primarily differences in amino acid biosynthesis and degradation along with insulin receptor signalling, while 1-3 had only one unique pathway, i.e. T2D mellitus signalling. The intersection between the set of pathways highlighted by comparisons 1-2 and 1-3 contains lipid signalling pathways, ceramide signalling, sphingosine-1-phosphate signalling. The lipid signalling is characteristic of a transition from normal BMI to an obese phenotype. This might indicate a transition from increase in amino acid biosynthesis/degradation and following appearance of lipid biosynthesis. The sphingomyelin class of lipids such as ceramides is associated with differential networks 1-2 and 1-3. Sphingolipids plays an important role in obesity and correlated co-morbidities [Torretta et al., 2019]. A list of the metabolites identified as commonly dif-
ferentially expressed between the three Clusters and associated pathways of activation is reported in Table 1.

| Clin. LDL Chol | HDL Chol | Triglycerides | Phosphoglycerides | Cholines | Sphingomyelins | APO B | APO A1 | Omega 3 | Omega 6 | PUFA | MUFA | SFA | Linoleic acid | DHA | Alanine | Glutamine | Glycine | Histidine | Isoleucine | Leucine | Phenylalanine | Tyrosine | Glucose | Lactate | Pyruvate | Citrate | b-OHbutyrate | Acetate | Acetoacetate | Acetone | Creatinine | Albumin | Glycoprotein acetyls |
|----------------|----------|--------------|-------------------|----------|----------------|--------|--------|---------|---------|-------|-------|-----|----------------|-----|---------|----------|---------|----------|-----------|--------|-------------|----------|---------|--------|----------|--------|---------------|--------|----------------|---------|---------------|--------|------------------|

Figure 4: Posterior differential network and result of IPA between Clusters 1 and 2. Threshold for edge inclusion set to 0.9.

4 Conclusions

We propose a Bayesian semiparametric model which allows clustering of subjects based on both longitudinal trajectories and patterns of metabolic association. The work is motivated by a study in early mechanisms of obesity, but it is applicable to several scenarios presenting data with similar features. Excess bodyweight is one of the leading risk factors contributing to the overall burden of disease worldwide [McMillen et al., 2009]. Childhood obesity is one of the major health problems in western countries and it is increasingly affecting Asian countries. The excessive accumulation of adipose tissue causes inflammation, oxidative stress, apoptosis, and mitochondrial dysfunctions, leading to the development of severe co-morbidities including type-2 diabetes mellitus, liver steatosis, cardiovascular, and neurodegenerative diseases which can develop early in life [Faienza et al., 2019].

Our analysis has identified potential key pathways in obese children in
order to explore possible molecular mechanisms associated with child obesity (see Table 1). We identified 18 metabolic pathways common to the three differential networks, the majority of which involves amino acids. An analysis of these associations reveals multiple biochemical pathways such as aromatic amino acid metabolism, branched-chain amino acid metabolism, glutathione metabolism, gluconeogenesis, tricarboxylic acid cycle, anti-inflammatory pathways and lipid metabolism. The analysis shows comprehensive initiation of amino biosynthesis as well as precursor molecule degradation, NAD biosynthesis, TCA cycle responsible for providing feeder molecules to sustain the flux required for fat metabolism through synthesis and degradation of aromatic amino acids as well as precursors for acetyl-CoA. The pathways that are unique to each set are able to filter out lipid pathways responsible for BMI/obesity and dyslipidemia, as well as onset of diabetes.

Finally, our results suggest that alterations in amino acid metabolism may play an important role in adiposity and dyslipidemia in children which may be relevant to the susceptibility of metabolic diseases later in life. Our findings are consistent with recent findings which investigate the relationship between obesity in children and pathways (and their combinations) related with amino acid, lipid and glucose metabolism [see, for example, Matsumoto
Figure 6: Posterior differential network and result of IPA between Clusters 2 and 3. Threshold for edge inclusion set to 0.9.

et al., 2021].
Figure 7: Venn diagram of the results of the IPA analysis performed on each differential network resulting from the pair-wise comparisons between the graphical structures estimated within each cluster. Each circle in the diagram refers to one comparison (e.g., 1-2 refers to the comparison between Cluster 1 and Cluster 2). The numbers in the diagram indicate the number of unique pathways found to statistically significant via IPA.
Table 1: Metabolites identified as commonly differentially expressed between the three clusters and associated pathways of activation derived from existing literature.

| Metabolite              | Primary Pathway                                      |
|-------------------------|------------------------------------------------------|
| Tyr                     | Aromatic amino acid metabolism                       |
| Leu                     | BCAA metabolism [Roberts et al., 2020]               |
| Ala                     | Gluconeogenesis [Roberts et al., 2020]              |
| Cystine                 | Glutathione metabolism [Roberts et al., 2020]       |
| Gly                     | Glutathione metabolism [Roberts et al., 2020]       |
| Glycoprotein acetyl     | Chronic inflammation [Ritchie et al., 2015]         |
| DHA                     | Energy expenditure                                   |
|                         | Lipid catabolism [Kuda, 2017]                        |
|                         | Anti-inflammatory pathways [Poudyal et al., 2011]    |
| Acetate                 | Ketogenesis, TCA cycle [Fletcher et al., 2019]       |
| HDL, Triglycerides      | Energy expenditure fat utilization [Canfora and Blaak, 2017] |
|                         | Fatty acid metabolism                                |
Acknowledgements

This research is supported by the Singapore National Research Foundation under the Translational and Clinical Research (TCR) Flagship, and Open Fund Large Collaborative Grant (OFLCG) Programmes and administered by the Singapore Ministry of Health’s National Medical Research Council (NMRC), Singapore - NMRC/TCR/004-NUS/2008; NMRC/TCR/012-NUHS/2014; OFLCG/MOH-000504. Additional funding is provided by the Singapore Institute for Clinical Sciences, Agency for Science Technology and Research (A*STAR), Singapore.
References

R. Argiento, A. Cremaschi, and M. Vannucci. Hierarchical normalized completely random measures to cluster grouped data. *Journal of the American Statistical Association*, 2019.

Izzuddin M Aris, Ling-Wei Chen, Mya Thway Tint, Wei Wei Pang, Shu E Soh, Seang-Mei Saw, Lynette Pei-Chi Shek, Kok-Hian Tan, Peter D Gluckman, Yap-Seng Chong, et al. Body mass index trajectories in the first two years and subsequent childhood cardio-metabolic outcomes: a prospective multi-ethnic asian cohort study. *Scientific reports*, 7(1):1–10, 2017.

M. N. Arroyo, J. A. Green, M. Cnop, and M. Igoillo-Esteve. tRNA biology in the pathogenesis of diabetes: Role of genetic and environmental factors. *International Journal of Molecular Sciences*, 22(2):496, 2021.

P. G. Balikcioglu and C. B. Newgard. Metabolomic signatures and metabolic complications in childhood obesity. In *Pediatric Obesity*, pages 343–361. Springer, 2018.

M. M. Barbieri and J. O. Berger. Optimal predictive model selection. *The annals of statistics*, 32(3):870–897, 2004.

J. A. Bell, C. J Bull, M. J. Gunter, D. Carslake, A. Mahajan, G. D. Smith, N. J. Timpson, and E. E. Vincent. Early metabolic features of genetic liability to type 2 diabetes: cohort study with repeated metabolomics across early life. *Diabetes Care*, 2020.

D. A. Binder. Bayesian cluster analysis. *Biometrika*, 65(1):31–38, 1978.

N. F. Butte, Y. Liu, I. F. Zakeri, R. P. Mohney, N. Mehta, V. S. Voruganti, H. Göring, S. A. Cole, and A. G. Comuzzie. Global metabolomic profiling targeting childhood obesity in the hispanic population. *The American journal of clinical nutrition*, 102(2):256–267, 2015.

A. Canale, A. Lijoi, B. Nipoti, and I. Prünster. On the pitman–yor process with spike and slab base measure. *Biometrika*, 104(3):681–697, 2017.

E. E. Canfora and E. E. Blaak. Acetate: a diet-derived key metabolite in energy metabolism: good or bad in context of obesity and glucose homeostasis? *Current opinion in clinical nutrition and metabolic care*, 20(6):477–483, 2017.
L-W Chen, M-T Tint, Marielle V Fortier, Izzuddin M Aris, LP-C Shek, Kok Hian Tan, Victor Samuel Rajadurai, Peter D Gluckman, Y-S Chong, Keith M Godfrey, et al. Body composition measurement in young children using quantitative magnetic resonance: a comparison with air displacement plethysmography. *Pediatric obesity*, 13(6):365–373, 2018.

Arthur P Dempster. Covariance selection. *Biometrics*, pages 157–175, 1972.

P. Diaconis and D. Ylvisaker. Conjugate priors for exponential families. *The Annals of statistics*, pages 269–281, 1979.

C. Dray, C. Knauf, D. Daviaud, A. Waget, J. Boucher, M. Buléon, P. D. Cani, C. Attané, C. Guigné, and C. Carpéné. Apelin stimulates glucose utilization in normal and obese insulin-resistant mice. *Cell metabolism*, 8(5):437–445, 2008.

D. B. Dunson, A. H. Herring, and S. M. Engel. Bayesian selection and clustering of polymorphisms in functionally related genes. *Journal of the American Statistical Association*, 103(482):534–546, 2008.

S. Ellul, M. Wake, S. A. Clifford, K. Lange, P. Würtz, M. Juonala, T. Dwyer, J. B. Carlin, D. P. Burgner, and R. Saffery. Metabolomics: population epidemiology and concordance in australian children aged 11–12 years and their parents. *BMJ open*, 9(Suppl 3), 2019.

Maria Felicia Faienza, Gabriele D’Amato, Mariangela Chiarito, Graziana Colaianni, Silvia Colucci, Maria Grano, Filomena Corbo, and Giacomina Brunetti. Mechanisms involved in childhood obesity-related bone fragility. *Frontiers in endocrinology*, 10:269, 2019.

S. Favaro and Y. W. Teh. Mcmc for normalized random measure mixture models. *Statistical Science*, 28(3):335–359, 2013.

T. S. Ferguson. A bayesian analysis of some nonparametric problems. *The annals of statistics*, pages 209–230, 1973.

J. A. Fletcher, S. Deja, S. Satapati, X. Fu, S. C. Burgess, and J. D. Browning. Impaired ketogenesis and increased acetyl-coa oxidation promote hyperglycemia in human fatty liver. *JCI insight*, 4(11), 2019.
M. Freemark. *Pediatric obesity: Etiology, pathogenesis, and treatment.* Springer, 2010.

A. E. Gelfand, A. Kottas, and S. N. MacEachern. Bayesian nonparametric spatial modeling with dirichlet process mixing. *Journal of the American Statistical Association*, 100(471):1021–1035, 2005.

M. Guindani, P. Müller, and S. Zhang. A bayesian discovery procedure. *Journal of the Royal Statistical Society: Series B (Statistical Methodology)*, 71(5):905–925, 2009.

M. L. Høgild, A. M. Bak, S. B. Pedersen, J. Runghy, J. Frystyk, N. Møller, N. Jessen, and J. O. L. Jørgensen. Growth hormone signaling and action in obese versus lean human subjects. *American Journal of Physiology-Endocrinology and Metabolism*, 316(2):E333–E344, 2019.

A. Jara, M. J. Garcia-Zattera, and E. Lesaffre. A dirichlet process mixture model for the analysis of correlated binary responses. *Computational Statistics & Data Analysis*, 51(11):5402–5415, 2007.

B. Jones, C. Carvalho, A. Dobra, C. Hans, C. Carter, and M. West. Experiments in stochastic computation for high-dimensional graphical models. *Statistical Science*, pages 388–400, 2005.

S. Kim, D. B. Dahl, and M. Vannucci. Spiked dirichlet process prior for bayesian multiple hypothesis testing in random effects models. *Bayesian analysis (Online)*, 4(4):707, 2009.

A. Krämer, J. Green, J. Pollard Jr, and S. Tugendreich. Causal analysis approaches in ingenuity pathway analysis. *Bioinformatics*, 30(4):523–530, 2014.

O. Kuda. Bioactive metabolites of docosahexaenoic acid. *Biochimie*, 136:12–20, 2017.

R. F MacLehose, D. B. Dunson, A. H. Herring, and J. A. Hoppin. Bayesian methods for highly correlated exposure data. *Epidemiology*, pages 199–207, 2007.

Shirou Matsumoto, Tomomi Nakamura, Fusa Nagamatsu, Jun Kido, Rieko Sakamoto, and Kimotoshi Nakamura. Metabolic and biological changes in
children with obesity and diabetes. *World Journal of Meta-Analysis*, 9(2): 153–163, 2021.

I Caroline McMillen, Leewen Rattanatray, Jaime A Duffield, Janna L Morrison, Severence M MacLaughlin, Sheridan Gentili, and Beverley S Muhlhausler. The early origins of later obesity: pathways and mechanisms. *Early nutrition programming and health outcomes in later life*, pages 71–81, 2009.

S. J. Mihalik, S. F. Michaliszyn, J. De Las Heras, F. Bacha, S. Lee, D. H. Chace, V. R. DeJesus, J. Vockley, and S. A. Arslanian. Metabolomic profiling of fatty acid and amino acid metabolism in youth with obesity and type 2 diabetes: evidence for enhanced mitochondrial oxidation. *Diabetes care*, 35(3):605–611, 2012.

A Misra and L Khurana. Obesity-related non-communicable diseases: South asians vs white caucasians. *International journal of obesity*, 35(2):167–187, 2011.

A. Mohammadi and E. C. Wit. Bayesian structure learning in sparse gaussian graphical models. *Bayesian Analysis*, 10(1):109–138, 2015.

P. M"uller, F. Quintana, and G. L. Rosner. A product partition model with regression on covariates. *Journal of Computational and Graphical Statistics*, 20(1):260–278, 2011.

H. Nam, B. S. Ferguson, J. M. Stephens, and R. F. Morrison. Impact of obesity on il-12 family gene expression in insulin responsive tissues. *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease*, 1832(1):11–19, 2013.

R. M. Neal. Markov chain sampling methods for dirichlet process mixture models. *Journal of computational and graphical statistics*, 9(2):249–265, 2000.

C. B. Newgard, J. An, J. R. Bain, M. J. Muehlbauer, R. D. Stevens, L. F. Lien, A. M. Haqq, S. H. Shah, M. Arlotto, C. A. Slentz, and J. Rochon. A branched-chain amino acid-related metabolic signature that differentiates obese and lean humans and contributes to insulin resistance. *Cell metabolism*, 9(4):311–326, 2009.
E. Oken, A. A. Baccarelli, D. R. Gold, K. P. Kleinman, A. A. Litonjua, D. De Meo, J. W. Rich-Edwards, S. L. Rifas-Shiman, S. Sagiv, E. M. Taveras, and S. T. Weiss. Cohort profile: project viva. *International journal of epidemiology*, 44(1):37–48, 2015.

Yi Ying Ong, Jonathan Y Huang, Navin Michael, Suresh Anand Sadanathan, Wen Lun Yuan, Ling-Wei Chen, Neerja Karnani, S Sendhil Velan, Marielle V Fortier, Kok Hian Tan, et al. Cardiometabolic profile of different body composition phenotypes in children. *The Journal of Clinical Endocrinology & Metabolism*, 106(5):e2015–e2024, 2021.

W. Perng, M. W. Gillman, A. F. Fleisch, R. D. Michalek, S. M. Watkins, E. Isganaitis, M. Patti, and E. Oken. Metabolomic profiles and childhood obesity. *Obesity*, 22(12):2570–2578, 2014.

C. Peterson, F. C. Stingo, and M. Vannucci. Bayesian inference of multiple gaussian graphical models. *Journal of the American Statistical Association*, 110(509):159–174, 2015.

P. Petrus, S. Lecoutre, L. Dollet, C. Wiel, A. Sulen, H. Gao, B. Tavira, J. Laurencikiene, O. Rooyackers, A. Checa, and I. Douagi. Glutamine links obesity to inflammation in human white adipose tissue. *Cell metabolism*, 31(2):375–390, 2020.

J. Pitman. *Combinatorial stochastic processes: Ecole d’été de probabilités de saint-flour xxxii-2002*. Springer, 2006.

H. Poudyal, S. K. Panchal, V. Diwan, and L. Brown. Omega-3 fatty acids and metabolic syndrome: effects and emerging mechanisms of action. *Progress in lipid research*, 50(4):372–387, 2011.

S. C. Ritchie, P. Würtz, A. P. Nath, G. Abraham, A. S. Havulinna, L. G. Fearnley, A. Sarin, A. J. Kangas, P. Soininen, K. Aalto, and I. Seppälä. The biomarker glyca is associated with chronic inflammation and predicts long-term risk of severe infection. *Cell systems*, 1(4):293–301, 2015.

J. A. Roberts, V. R. Varma, C. Huang, Y. An, A. Oommen, T. Tanaka, L. Ferrucci, P. Elango, T. Takebayashi, S. Harada, and M. Iida. Blood metabolite signature of metabolic syndrome implicates alterations in amino acid metabolism: findings from the baltimore longitudinal study of aging
A. Roverato. Hyper inverse Wishart distribution for non-decomposable graphs and its application to Bayesian inference for Gaussian graphical models. *Scandinavian Journal of Statistics*, 29(3):391–411, 2002.

R. M. Sakia. The box-cox transformation technique: a review. *Journal of the Royal Statistical Society: Series D (The Statistician)*, 41(2):169–178, 1992.

J. Sethuraman. A constructive definition of dirichlet priors. *Statistica sinica*, pages 639–650, 1994.

Elin Shaddox, Christine B Peterson, Francesco C Stingo, Nicola A Hana-nia, Charmion Cruickshank-Quinn, Katerina Kechris, Russell Bowler, and Marina Vannucci. Bayesian inference of networks across multiple sample groups and data types. *Biostatistics*, 21(3):561–576, 2020.

S. Soh, M. T. Tint, P. D. Gluckman, K. M. Godfrey, A. Rifkin-Graboi, Y. H. Chan, W. Stünkel, J. D. Holbrook, K. Kwek, Y. S. Chong, and S. M. Saw. Cohort profile: Growing up in singapore towards healthy outcomes (gusto) birth cohort study. *International journal of epidemiology*, 43(5):1401–1409, 2014.

C. Takashina, I. Tsujino, T. Watanabe, S. Sakaue, D. Ikeda, A. Yamada, T. Sato, H. Ohira, Y. Otsuka, N. Oyama-Manabe, and Y. M. Ito. Associations among the plasma amino acid profile, obesity, and glucose metabolism in japanese adults with normal glucose tolerance. *Nutrition & metabolism*, 13(1):1–10, 2016.

L. S. L. Tan, A. Jasra, M. De Iorio, and T. M. D. Ebbels. Bayesian inference for multiple gaussian graphical models with application to metabolic association networks. *The Annals of Applied Statistics*, pages 2222–2251, 2017.

Enrica Torretta, Pietro Barbacini, Nasser M Al-Daghri, and Cecilia Gelpi. Sphingolipids in obesity and correlated co-morbidities: the contribution of gender, age and environment. *International journal of molecular sciences*, 20(23):5901, 2019.
B. Valcárcel, P. Würtz, N. K. S. al Basatena, T. Tukiainen, A. J. Kangas, P. Soininen, M. R. Järvelin, M. Ala-Korpela, T. M. Ebbels, and M. De Iorio. A differential network approach to exploring differences between biological states: an application to prediabetes. *PLoS One*, 6(9):e24702, 2011.

S. Wahl, Z. Yu, M. Kleber, P. Singmann, C. Holzapfel, Y. He, K. Mittelstrass, A. Polonikov, C. Prehn, W. Römisch-Margl, and J. Adamski. Childhood obesity is associated with changes in the serum metabolite profile. *Obesity facts*, 5(5):660–670, 2012.

T. J. Wang, M. G. Larson, R. S. Vasan, S. Cheng, E. P. Rhee, E. McCabe, G. D. Lewis, C. S. Fox, P. F. Jacques, C. Fernandez, and C. J. O’donnell. Metabolite profiles and the risk of developing diabetes. *Nature medicine*, 17(4):448–453, 2011.

WHO. World health organisation - *Obesity and overweight*, Accessed: 01-06-2021. https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight.

P. Würtz, P. Soininen, A. J. Kangas, T. Rännemaa, T. Lehtimäki, M. Kähönen, J. S. Viikari, O. T. Raitakari, and M. Ala-Korpela. Branched-chain and aromatic amino acids are predictors of insulin resistance in young adults. *Diabetes care*, 36(3):648–655, 2013.