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Metabolism of bile acids in the post-prandial state

Jarlei Fiamoncini[1], Rui Curi[2] and Hannelore Daniel[3]

[1]INRA, UMR 1019, UNH, CRNH Auvergne, F-63000 Clermont-Ferrand; Clermont Université, Université d'Auvergne, Unité de Nutrition Humaine, BP 10448, F-63000 Clermont-Ferrand, France; [2]University of São Paulo – Institute of Biomedical Sciences, Department of Physiology and Biophysics, São Paulo, Brazil; [3]Technische Universität München, Chair of Nutritional Physiology, Freising, Germany

Correspondence: Jarlei Fiamoncini (jarleifiamoncini@gmail.com)

The modulation of energy expenditure by dietary administration of cholic acid in mice promoted interest in studying bile acid(s) (BA) as adjuvants in the treatment of metabolic diseases such as obesity and diabetes. Bile acids can modulate intermediary metabolism by acting directly on nuclear as well as G-protein-coupled receptors or indirectly through changes in gut microbiota. Despite the potential of BA to affect intermediary metabolism, plasma kinetics and changes in individual BA in blood in the post-prandial state have been neglected for a long time. Minutes after ingestion of a meal (or a glucose challenge), the plasma BA concentration increases as a result of the secretion of bile into the duodenum, followed by intestinal absorption and a systemic circulation spillover. A large inter-individual variability of post-prandial kinetics of plasma BA is documented. Factors such as gender, diet composition, circadian oscillations, and individual capacities for the synthesis and transport of BA play important roles in determining this variability and are discussed in the present short review in light of new findings.

Introduction

Special attention has been given to bile acid(s) (BA) in recent years due to new functions attributed to them [1,2]. These molecules can directly modulate intermediary metabolism by activating different signaling pathways (e.g. the farnesoid X receptor (FXR) or G-protein-coupled receptors such as TGR5) or indirectly by changing the composition of the intestinal microbiota [3,4], which in turn can change gastrointestinal functions, and also via the secretion of gastrointestinal hormones which have systemic effects. A factor that has made BA research more popular is the improvement of their analysis, which was always a critical issue given their low abundance in plasma and the existence of isobaric isomers. A major advancement in the field was attained with the use of tandem mass spectrometry coupled to liquid chromatography for BA identification and quantitation [5–7]. This technique simplifies sample preparation (does not require derivatization as in gas chromatography-mass spectrometry (GC-MS) and allows high-throughput applications. Systematic studies on post-prandial changes of individual BA levels in plasma are scarce and some recent findings on plasma changes were more or less incidental as part of metabolomics discovery approaches. The aim of this review is to provide a brief overview on the metabolism and functions of BA, focusing on the most recent findings regarding their post-prandial kinetics, addressing some problems, and providing some suggestions for future studies.

Bile acid metabolism in a nutshell

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Cholic acid (CA) and chenodeoxycholic acid (CDCA) are the main BA synthesized from cholesterol in the liver and are named primary BA. The last step of BA synthesis in the liver is their conjugation with taurine or glycine at the carboxylic acid side chain, which is catalyzed by bile acid CoA:amino acid *N*-acyltransferase (BAAT) [8]. Glycine is preferably used in humans whereas taurine is mainly used in rodents for conjugation with BA. After their synthesis, BA are stored in the gall

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bladder making up bile in combination with cholesterol, phospholipids, bilirubin, and water [9]. When nutrients reach the small intestine and are sensed by enteroendocrine cells, cholecystokinin (CCK) is released and induces gall bladder contraction resulting in the secretion of bile into the duodenum. Bile promotes the emulsification of dietary lipids for digestion and the absorption of fatty acids, cholesterol, fat-soluble vitamins, and other hydrophobic components of the diet [10]. Up to 95% of secreted BA are finally reabsorbed in the small intestine either via passive diffusion, uniporters, or sodium ion (Na^+)-coupled symporters (SLC10A1), and returned via portal blood to the liver. Despite the very efficient hepatic extraction of BA, a portion is not immediately taken up during the first passage through the liver and thus reaches systemic circulation. This 'escape' from the liver is thus a spillover of BA. After uptake into hepatocytes, BA – not yet conjugated – may be coupled with taurine or glycine. All BA species are then exported via specialized transporters in the canalicular membrane for storage in the gall bladder closing a cycle that can be repeated several times a day, depending on appropriate stimuli. Bile acids that are not absorbed in the small intestine reach the large intestine and are metabolized by the microbiota through de-conjugation and 7 α / β-dehydroxylation before being partly reabsorbed from the colon [11]. Deoxycholic (DCA) and lithocholic (LCA) acids are the most common products of bacterial metabolism of primary BA in humans and are thus named secondary BA.

The unique amphipathic character of BA resides in the distribution of their lipophilic and hydrophilic moieties on opposite sides of the planar structure formed by the steroid nucleus. Normally, two methyl groups are found in the β-face of the molecule and the hydroxy groups are in the α-face (with the exception of ursodeoxycholic acid (UDCA)). Despite being structurally very similar, the main BA found in mammals can be differentiated by the number and position of hydroxy groups (Figure 1). The structural characteristics combined with the glycine or taurine conjugation play a key role in BA polarity [12–14].

Bile acids as signaling molecules

Bile acids have been recognized as signaling molecules with the capacity to affect mammalian energy metabolism. Cholic acid was reported to increase energy expenditure when supplemented in the diet of mice. This was associated with increased gene expression of enzymes involved in fatty acid oxidation, preventing obesity and insulin resistance. Similar effects were elicited by BA in human skeletal myocytes via cyclic adenosine monophosphate (AMP)-mediated activation of deiodinase 2 [15,16] and the effects of active thyroid hormone. These findings promoted research into the use of BA to target obesity and associated diseases.

It has been known since 1999 that BA are ligands of the FXR and can, via this mechanism, alter their own synthesis and transport. However, various other genes are also under FXR control, for example, the short heterodimer partner (SHP) [1,2,17–19]. Activation of SHP and its effect upon HNF-4 or Foxo1 leads to decreased expression of gluconeogenic enzymes, which directly affects glucose homeostasis [3,4,20,21].

TGR5 has been described as a membrane receptor for BA [5–7,22,23] and leads to faster responses than those mediated by FXR. Given the wide expression of TGR5, the effects of BA have been reported in different cell and tissue types such as leukocytes leading to immunomodulatory effects, GLP-1 secretion by the intestine, and deiodinase 2 activation in brown adipose tissue and myocytes [9,22,24].

The potential of BA as modulators of glucose homeostasis was first reported by Garg and Grundy who described glucose-lowering effects induced by cholestyramine therapy [25]. Subsequently, several studies with BA sequestrants reached the same conclusions and differences in circulating BA levels were found between obese and diabetic patients in comparison with lean and healthy individuals [26]. In 2010, two independent studies described the importance of FXR for insulin synthesis and secretion, as well as for the protection of pancreatic islets from lipotoxicity [27,28]. On top of modulating insulin synthesis, FXR also inhibits the expression of different gluconeogenic enzymes – an effect that matches well with their emerging role as post-prandial signaling molecules [29]. BA can thus, via FXR (or other mechanisms), regulate insulin synthesis and secretion and diminish hepatic *de novo* glucose synthesis and output. On the other hand, glucose and insulin have been reported to modulate BA synthesis through an increase in CYP7A1 expression. Li et al. [30] described glucose activating CYP7A1 expression by increasing the acetylation and decreasing methylation of the mouse *cyp7a1* gene promoter.

Although various studies have demonstrated BA modulate energy metabolism in mice, whether this applies to humans as well is not known. In fact, a study on the association of fasting plasma levels of BA with fasting energy expenditure in humans has failed to establish a connection [31].The effects of BA on energy metabolism are probably more pronounced in the post-prandial state when their plasma levels are increased and insulin secretion and action could be affected by BA.

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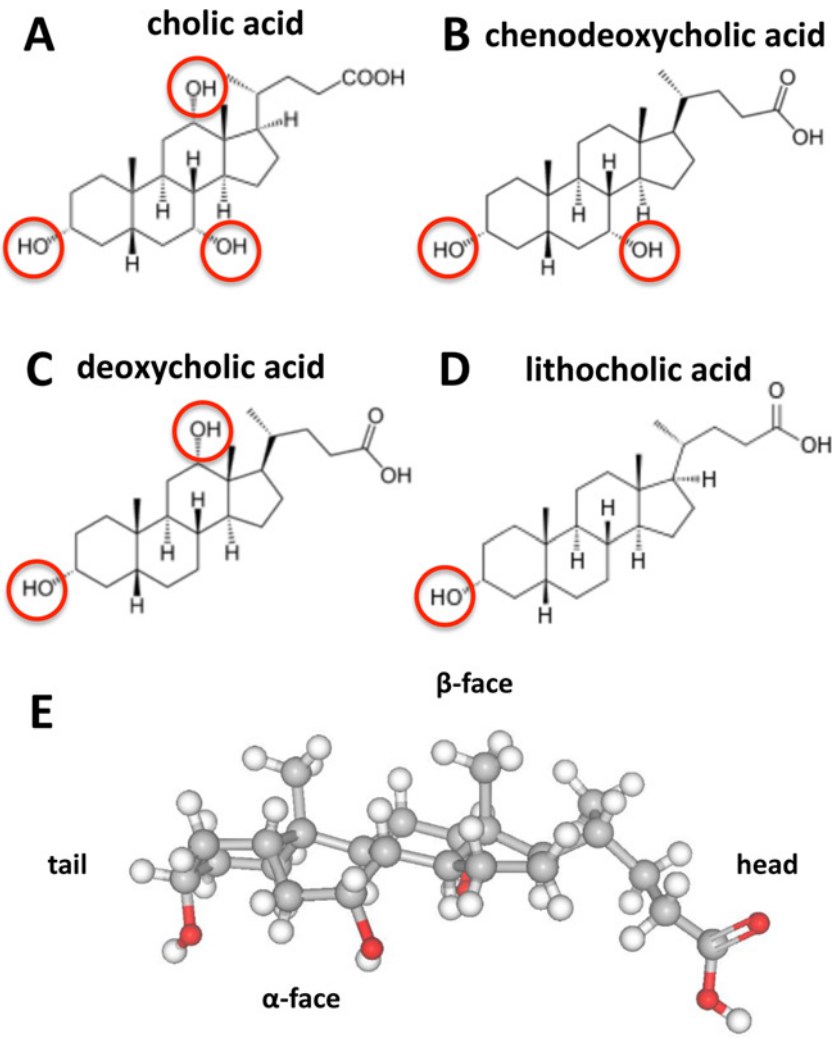

Figure 1. Chemical structure of the most abundant mammalian bile acids in their unconjugated form.

(A–D) Molecular structure of the most abundant bile acids (BA) found in mammals in their unconjugated form. The number and position of hydroxy groups, which are responsible for major physico-chemical differences among the BA species, are highlighted. (**E**) Side view of a three-dimensional (3D) representation of the chemical structure of cholic acid (CA). The distribution of methyl groups in the β-face and hydroxy groups in the α-face of the planar steroid nucleus is responsible for the unique amphipathic character of BA. The information for the chemical structure of CA is available in the PubChem Substance and Compound database through the chemical structure identifier CID: 221493. The CA 3D structure was rendered using Jmol: an open-source Java viewer for chemical structures in 3D. http://www.jmol.org/

Plasma kinetics of bile acids in the post-prandial state

The peripheral venous blood concentration of BA measured in the post-prandial state represents, in essence, the hepatic spillover. Hepatic BA extraction is very efficient and even though there is a linear correlation between the concentration of BA in systemic circulation and their concentration in portal blood, the absolute values are quite different [10,32,33]. Hepatic uptake of BA from portal blood is influenced by their physico-chemical properties and lipophilicity is negatively correlated with the V_{max} for uptake. As a consequence, CA is transported more efficiently than CDCA and taurine-conjugated BA are more efficiently taken up than unconjugated substrates [11,34]. A fact that is frequently overlooked is the more lipophilic BA – DCA and CDCA – can be transported together with the lipid

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fraction of the diet via the lymph [15,16,35] to enter the circulation. Even though the amount of BA transported via this route seems small, it may affect BA-dependent metabolic regulation; particularly when considering the effects of BA on inflammation (via TGR5 and FXR) and modulation of immune cell function, which was shown to be affected by a simple meal [36–38]. This constitutes a still poorly explored triangle involving inflammation, digestion, and energy expenditure.

The appearance of BA in the circulation in the first hour after a meal is the result of bile secretion into the small intestine and their fast absorption. Considering the average oro-cecal transit time, it is unlikely that BA secreted following food ingestion reach the large intestine in the first hour following a meal and therefore bacterial activity does not have a marked acute effect on post-prandial plasma BA concentrations [39,40]. This implies that the increase in primary and conjugated BA in circulation in response to a meal represents primarily the bile composition in BA [7,14,41].

Comparisons of studies on post-prandial BA secretion can only be performed if the different experimental designs are carefully considered. Secretion and plasma kinetics of BA vary largely with the composition of the meal and the period of the day. It is also influenced by gender and displays large inter-individual variations. On top of that, the slow clearance of BA from circulation after the post-prandial peak hampers interpretation of the results. In fact, experimental protocols are usually designed to assess the metabolism of markers with faster kinetics, such as glucose.

Plasma kinetics of bile acids during an oral glucose tolerance test

Shaham and co-workers described a 2-fold increase in glycochenodeoxycholic acid (GCDCA) and a 3-fold increase in taurochenodeoxycholic acid (TCDCA) in plasma that lasted for two hours after an oral glucose tolerance test (OGTT) with 75 g of glucose in healthy male subjects (aged 18–30 years) [42]. Maximal concentration of plasma BA was reached after 30 minutes. The rapid rise – as fast as glucose – can be taken as an indicator of absorption from the upper small intestine that precedes absorption in the ileum, which is usually considered as the major site for intestinal BA uptake. BA are thus amongst the metabolites that change most in plasma during an OGTT. In fact, Zhao et al. reported plasma levels of glycodeoxycholic acid (GDCA), GCDCA, and glycocholic acid (GCA) to increase by 4.5- and 6-fold in Caucasian subjects (48 ± 2.5 yrs). These authors also described the biphasic appearance of BA in plasma with one peak at 30 minutes and a second peak after 120 minutes [43]. Angelin et al. [32] in a pioneering study (performed in 1982) measured BA in portal and periphery blood sampled during a mixed-meal challenge. They described the biphasic appearance of BA in the portal blood of most patients that was not always observed in peripheral blood [32]. Matysik et al. obtained similar results but reported a 1.85-fold decrease in the concentration of non-conjugated BA during the course of the OGTT, performed in healthy men and women aged 18 to 49 years [44]. The authors attributed the latter observation to activation of FXR, leading to repression of CYP7A1 – the rate-limiting enzyme of BA synthesis [1]. Following the decrease in non-conjugated BA levels in serum during the OGTT, a decrease in the concentration of 7α-hydroxy-4-cholesten-3-one was reported, which was assumed to reflect 7α-hydroxylation mediated by Cyp7a1 [45]. The increase in plasma BA levels following glucose ingestion is a striking finding per se since these acids are not needed for glucose absorption. However, it was shown in 1985 that glucose alone causes CCK secretion and, in turn, promotes gall bladder contraction [10].

Plasma kinetics of bile acids in response to fat-containing meals

During an oral lipid tolerance test (OLTT) in healthy women and men aged 18–54 years, Schmid et al. reported a 3-fold increase in primary and secondary BA, a 5-fold increase in taurine-conjugated BA, and a 4.3-fold increase in glycine-conjugated BA levels in plasma [46]. These authors also described decreased levels of the unconjugated BA CDCA and LCA but no changes were observed in CA, DCA, UDCA, and hyodeoxycholic acid (HDCA) levels. It is worth mentioning that in that study, serum was sampled every second hour for six hours and the test drink contained exclusively lipids (84 g) with only trace amounts of carbohydrates. Haeusler et al. reported a 3-fold increase in total BA following a meal containing 75 g of glucose, 40 g of Parmesan cheese, and 50 g of egg (16% protein, 56% carbohydrates, and 28% fat) [47]. In contrast with findings by Matysik et al. (2011) with an OGTT, Haeusler et al. described increased

concentrations of 7α-hydroxy-4-cholesten-3-one during the mixed-meal test. Interestingly, in that study, the maximal plasma BA concentration was achieved only 2 hours after the meal and sampling was interrupted after 3 hours, when BA were still at maximal plasma concentrations [47,44]. That study enrolled subjects of both genders and reported that despite obese subjects tend to have higher circulating BA concentrations, the post-prandial increase in plasma BA levels was blunted in comparison with non-obese subjects. Ewang-Emukowhate et al. studied the post-prandial response of lean male subjects (aged 21 years) to a diet containing 46.6 g of carbohydrates, 28.5 g of fat, and 10.4 g of proteins. Glycine-conjugated BA concentrations were increased 6-fold in the circulation, peaking after 90 minutes, whereas the plasma concentration of unconjugated BA did not have any alteration in response to the test meal [48]. Sonne et al. investigated post-prandial responses to an OGTT and to liquid meals with 2.5, 10, and 40 g of lipids in healthy and diabetic men and women aged 42–71 years. These authors reported that plasma concentrations of BA and gall bladder emptying are closely associated with the fat content in the meals and there was no difference between diabetic patients and healthy controls [49,50]. The peak levels of BA in plasma were observed between the first and second hour, and glycine- and taurine-conjugated BA dominated the response, whereas the concentration of unconjugated BA showed no comparable changes. Despite no differences in the gall bladder emptying response, diabetic patients displayed higher concentrations of BA than healthy controls [49].

In summary, mixed meals (fat-containing) generally induce a higher secretion of BA than an OGTT. Despite the well-known relationship between CCK secretion and gall bladder emptying with the fat content of the meal, we still do not know if other factors are also involved [10,50]. The increase in circulating BA levels is not only more pronounced after the mixed meals but also remains increased for longer time periods in comparison with the OGTT. There is some disagreement on post-prandial kinetics of unconjugated BA. Some authors described no alterations, whereas others reported decreasing concentrations. There is, however, no doubt that plasma levels of primary BA, particularly glycine- and taurine-conjugated species, increase following ingestion of a meal or glucose. Information gathered from recent studies addressing blood post-prandial kinetics of BA is summarized in Table 1.

Inter-individual variability of bile acid kinetics in the post-prandial state

Bile acids are among the endogenous metabolites with the highest inter-individual variability. Even in the fasting state, circulating BA concentrations can vary by more than 10-fold between individuals. Their concentrations in plasma originates from diverse processes that can largely vary between individuals [51–54]. Food, drugs, and diurnal variation [51,53,55] are the major extrinsic factors that affect plasma BA composition. The individual capacities for BA synthesis, gall bladder function, intestinal motility, enterocyte uptake, and the transport of BA in the circulation, as well as hepatocyte uptake and microbiota biotransformation, are important intrinsic factors behind the large inter-individual variability of BA kinetics in the post-prandial state. Variations in genes involved in BA synthesis and turnover explain part of the variability observed between individuals. Genetic variations in transporters involved in BA uptake from the intestine (SLC10A2, SLC10A1, and SLCO1A2) as well as for uptake into hepatocytes and canalicular secretion such as (ABCA3, ABCG5, and ABCC2) have been reported and may cause the observed differences in BA handling between individuals. Analysis of plasma levels should thus use multivariate statistical analysis (e.g. cluster analysis) allowing individual groupings based on their similarities rather than using the mean levels within a cohort.

Despite the very large inter-individual variations in BA metabolism, post-prandial responses are very steady across time in the same individual – low intra-individual variation [47]. Bathena et al. reported high inter-individual but low intra-individual variability in four consecutive samplings [52]. A list of the different factors that contribute to the high inter-individual variation in BA metabolism and plasma levels is given in Figure 2.

Gender-related differences in bile acid metabolism

Controversial findings have been reported in BA metabolism between men and women. Gälman et al. reported 30% higher BA synthesis rates in men compared with women and other authors found the BA pool to be significantly smaller in women than in men [54,56,57]. Bathena et al. investigated a larger population and did not observe

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Table 1 Summary of kinetic parameters of circulating BA in the post-prandial state

Authors	Reference	Test	Δ BA	Δ G-BA	Δ T-BA	Δ U-BA	Time of BA peak (min)	Subjects	Duration of test (h)
Shaham et al.	[28]	OGTT	NA	≈2.5	≈3	NA	30	healthy, 18–30 y.o.	2
Zhao et al.	[29]	OGTT	NA	4.5 to 6	NA	NA	30	healthy, ≈ 48 y.o.	2
Matysik et al.	[30]	OGTT	1.22	1.63	1.33	-1.85	60 (t=30 NA)	healthy, mixed, 18–49 y.o.	2
Schmid et al.	[32]	OLTT	3	4.25	4.98	-2	120	healthy, mixed, 18–55 y.o.	6
Haeusler et al.	[33]	MMTT	4.31	7.80	5.88	1.72	120	non-obese, mixed, ≈ 39 y.o.	3
			2.86	4.08	4.33	1.16	120	obese , mixed, ≈ 37 y.o.	3
Ewang-Emukowhate et al.	[34]	MMTT	3.8	6	NA	1	90	healthy, men, ≈ 21 y.o.	2
Sonne et al.	[35]	OGTT	3**	3^	3^	1^	30	T2D, ≈ 59 y.o.	4
		MMTT *	5**	6^	6^	2^	60	T2D, ≈ 59 y.o.	4
Sonne et al.	[35]	OGTT	2**	2^	3^	1^	30	healthy, ≈ 59 y.o.	4
		MMTT *	5**	5^	10^	2^	90	healthy, ≈ 59 y.o.	4

Δ refers to the maximal change in concentration of the specified BA (expressed as fold change), at any time during the tests.
BA = bile acids.
G-BA = glycine-conjugated bile acids.
T-BA = taurine-conjugated bile acids.
U-BA = unconjugated bile acids.
NA = data not available.
OGTT = oral glucose tolerance test.
OLTT = oral lipid tolerance test.
MMTT = mixed-meal tolerance test.
y.o. = years old.
* in this table only data from the high-fat diet is reported.
** based on visual inspection of the graphic representation of post-prandial changes in total BA.
^ based on visual inspection of the graphic representation of the most abundant BA in each class (GCDCA, TCDCA, and DCA). Care should be taken as in this table we report changes from T=0 until the maximal value and not area under the curve as Sonne and co-workers presented the data in the original manuscript.

differences in fasting plasma concentrations of BA between men and women [52,54], although it has been known since the early 1980s that sex hormones affect BA metabolism.

Hormonal variations due to pregnancy or during the menstrual cycle have been linked to alterations in BA metabolism in women and gallstone formation is clearly more frequent in women than men [58,59]. Female mice also have a larger BA pool than male mice similar to differences in the expression of BA synthesis enzymes [60]. Moreover, old female mice present higher levels of circulating conjugated BA than male animals and have increased expression of the rate-limiting enzyme Cyp7a1 [61].

Following an OGTT, women have a more pronounced increase in taurine- and glycine-conjugated BA in plasma than men [62]. Schmid et al. found that after the sixth hour in an OLTT, circulating concentrations of taurine-conjugated BA were higher in women than men, despite no differences in fasting state [46]. They also reported an effect of hormonal contraception on plasma levels of taurine-conjugated BA. Despite the studies mentioned above, the mechanisms causing the gender-related differences in BA metabolism remain to be established.

Conclusions and perspectives

Considering the magnitude and duration of the post-prandial BA increase in circulation and our dietary habits with various meals and snacks during the day, the study of BA metabolism and their metabolic effects merit special attention. Plasma concentrations of BA following food intake are several-fold higher than in the fasting state and therefore their effects on the regulation of metabolism are likely to be more pronounced during the post-prandial period. Assessment of post-prandial BA metabolism should take into consideration the rather long periods in which BA

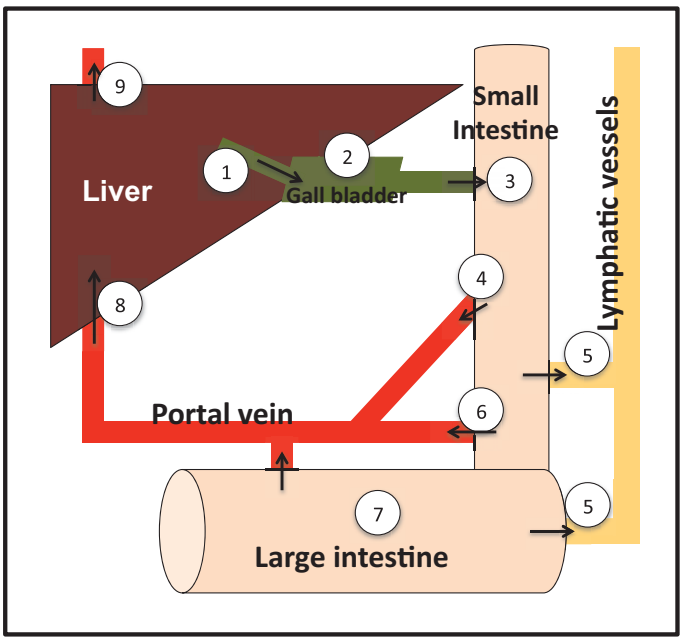

Figure 2. Schematic representation of the inter-organ metabolism of bile acids and factors that promote inter-individual variability of post-prandial BA kinetic metabolites.

1 Individual capacities for BA synthesis from cholesterol as well as their conjugation with taurine or glycine determine their production rate.

2 Bile acids are transported to the gall bladder, where they are concentrated and stored. Different transporters contribute to the transport of BA from the apical side of hepatocytes, via the biliary duct to the gall bladder.

3 Upon response to post-prandial stimuli (CCK, for example), the gall bladder contracts and secretes bile into the duodenum. The volume of the gall bladder, the responsiveness to CCK, and conditions like cholestasis and the presence of gallstones affect individual capacities to secret bile and BA into the intestine.

4 and 6 Bile acids are taken up from the intestinal lumen by passive diffusion in the upper parts of the small intestine. In the distal part (ileum), active transport efficiently takes up the remaining BA and transports them into the portal vein. Different genes are involved, such as apical and basolateral membrane transporters as well as BA binding proteins. They contribute to the inter-individual variation observed in post-prandial BA kinetics. Intestinal motility (which varies from person to person) and the composition of the diet determine the period of time needed for BA reach the ileum.

5 Usually overlooked, a portion of BA is transported into the lymphatic vessels together with the lipid component of the diet. Individualities in lipoprotein metabolism can also account for variability at this step.

7 The small portion of BA that reaches the large intestine is subjected to microbiota-driven metabolism, being de-conjugated and converted into secondary BA. Most of the BA are transported back to the portal vein. This is probably one of the hottest points for the individual variability in BA plasma composition and metabolism. Intestinal microbiota can be greatly influenced by environmental and genetic factors and directly affects BA plasma composition. In turn, BA modulate the microbiological community of the large intestine.

8 When entering the liver, portal blood delivers BA coming from the intestine that are efficiently taken up by hepatocytes, closing the BA entero-hepatic circulation. In this step, the individual capacities of hepatic basolateral transporters determine the individual variability of BA kinetics.

9 Despite the very efficient hepatic uptake of BA, some are not immediately taken up in the first passage through the liver, reaching systemic circulation. This ‘escape’ from the liver is called the spillover of BA.

concentrations remain increased and blood sampling should be performed over longer periods of time and not be limited to the usual 2 or 4 hours often used in dietary challenges. Perhaps the most difficult aspect to explain is the large inter-individual variability and what it means for the responses when BA serve as ligands for FXR or TGR5. Bile acids are important signaling molecules closely associated with diet, microbiota, and intermediary metabolism.

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The combination of metabolite profiling with the assessment of genetic variation and gut microbiota composition analysis is thus required to better define the role of BA in the control of intermediary metabolism and inter-individual variability.

Summary

- Plasma concentrations of bile acids (BA) increase rapidly after a meal. The magnitude and time scale of this increase is proportional to the fat content of the diet and despite high inter-individual variability, in average maximal values are observed after 1 hour.
- The entero-hepatic circulation of BA requires the activity of different transporters in different cell types. The diversity of transporters involved in BA secretion and uptake give margin to large interindividual variability regarding the appearance of BA in systemic circulation in the post-prandial state.
- Inter-individual variability of BA kinetics in the post-prandial state is one of the major limitations to figure out the systemic effects of BA. Studies of the effects of BA on metabolism should consider the inter-individual variability of their plasma kinetic following a meal.

Abbreviations

ABCA3, ATP binding cassette Subfamily A member 3; ABCC2, ATP binding cassette Subfamily C member 2; ABCG5, ATP binding cassette Subfamily G member 5; AMP, adenosine monophosphate; BA, bile acids; BAAT, bile acid; CoA:amino acid N-acyltransferase; CA, cholic acid; CCK, cholecystokinin; CDCA, chenodeoxycholic acid; CYP7A1, cytochrome P450 Family 7 Subfamily A Member 1; DCA, deoxycholic acid; FXR, farnesoid X receptor; GCA, glycocholic acid; GC-MS, gas chromatography coupled to mass spectrometry; GCDCA, glycochenodeoxycholic acid; GDCA, glycodeoxycholic acid; GLP-1, glucagon like peptide 1; HDCA, hyodeoxycholic acid; HNF-4, hepatic nuclear factor 4; LCA, lithocholic acid; MMTT, mixed meal tolerance test. OGTT, oral glucose tolerance test; OLTT, oral lipid tolerance test; SLC10A1, solute carrier Family 10 member 1; SLC10A2, solute carrier Family 10 member 2; SLCO1A2, solute carrier organic anion transporter family member 1A2; SHP, short heterodimer partner; TCDCA, taurochenodeoxycholic acid; TGR5, G Protein-coupled bile acid receptor 1; UDCA, ursodeoxycholic acid.

Author Contribution

All authors contributed equally to the preparation of this article.

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Competing Interests

The authors have no competing interests in relation to this manuscript.

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