

Supplementary Materials: Metabolomic Approaches to Explore Chemical Diversity of Human Breast-Milk, Formula Milk and Bovine Milk

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Table S1. The differential metabolites obtained from multivariate and univariate analysis of formula milk relative to human breast milk.

Metabolites	Platform ^a	VIP ^b	FC ^c	<i>p</i> -Value ^d	Class
5-oxoproline	G	1.50	0.06	2.83×10^{-9}	amino acid
alanine	G	1.62	0.06	2.83×10^{-9}	amino acid
phenylalanine	G	1.7	32.91	1.96×10^{-11}	amino acid
glutamine	G	1.56	0.001	8.24×10^{-10}	amino acid
arginine	P	1.23	0.12	1.99×10^{-6}	amino acid
glutamic acid	G	1.61	0.15	3.98×10^{-7}	amino acid
glycine	G	1.32	0.17	2.83×10^{-9}	amino acid
histidine	P	1.18	2.40	1.74×10^{-5}	amino acid
serine	G	1.54	0.03	2.51×10^{-9}	amino acid
tyrosine	P	1.61	0.001	4.78×10^{-8}	amino acid
taurine	N	1.45	1.91	1.04×10^{-4}	amino acid
valine	G	1.55	0.05	2.63×10^{-8}	amino acid
acetylglucosamine	P	1.42	2.15	6.97×10^{-7}	carbohydrate
acetylneuraminic acid	N	1.27	0.44	3.97×10^{-7}	carbohydrate
arabinose	N	1.13	7.43	6.27×10^{-8}	carbohydrate
arabitol	G	1.32	0.35	5.59×10^{-7}	carbohydrate
fructose	G	1.42	30.63	3.20×10^{-5}	carbohydrate
fucose	G	1.21	0.04	9.99×10^{-8}	carbohydrate
glucosamine	N	1.16	1.81	2.82×10^{-9}	carbohydrate
glycerol	G	1.17	0.16	2.94×10^{-8}	carbohydrate
glyceraldehyde	N	1.26	1.51	3.26×10^{-6}	carbohydrate
maltose	G	1.31	5.06	4.42×10^{-7}	carbohydrate
threitol	P	1.29	7.22	9.47×10^{-7}	carbohydrate
xylose	P	1.06	0.16	5.35×10^{-4}	carbohydrate
arachidonic acid	N	1.47	ND ^e	7.70×10^{-16}	fatty acid
capric acid	G	1.51	ND	1.65×10^{-14}	fatty acid
dihydroxypropanoic acid	G	1.01	0.59	2.04×10^{-5}	fatty acid
docosadienoic acid	N	1.62	ND	3.91×10^{-19}	fatty acid
docosahexaenoic acid	P	1.42	ND	1.15×10^{-9}	fatty acid
docosapentaenoic acid	N	1.38	ND	6.59×10^{-11}	fatty acid
eicosadienoic acid	G	1.37	ND	1.07×10^{-10}	fatty acid
eicosapentaenoic acid	N	1.06	ND	5.72×10^{-6}	fatty acid
eicosatrienoic acid	G	1.65	ND	5.02×10^{-21}	fatty acid
eicosenoic acid	G	1.14	ND	7.38×10^{-7}	fatty acid
lauric acid	G	1.53	ND	5.27×10^{-15}	fatty acid
linoleic acid	N	1.50	0.05	6.91×10^{-12}	fatty acid
α -linolenic acid	N	1.50	ND	2.80×10^{-14}	fatty acid
γ -linolenic acid	N	1.47	ND	8.32×10^{-13}	fatty acid
myristic acid	N	1.23	ND	3.68×10^{-8}	fatty acid
myristoleic acid	G	1.18	ND	2.31×10^{-7}	fatty acid
oleic acid	G	1.25	ND	2.57×10^{-8}	fatty acid

Table S1. Cont.

Metabolites	Platform ^a	VIP ^b	FC ^c	p-Value ^d	Class
palmitic acid	G	1.16	0.02	2.47×10^{-5}	fatty acid
palmitoleic acid	P	1.59	ND	1.04×10^{-17}	fatty acid
pentadecanoic acid	N	1.53	ND	2.83×10^{-16}	fatty acid
tridecanoic acid	G	1.15	ND	4.30×10^{-7}	fatty acid
2-ketoglutaramic acid	G	1.28	26.24	3.03×10^{-5}	TCA cycle
2-ketoglutaric acid	G	1.21	4.23	9.17×10^{-6}	TCA cycle
citric acid	G	1.43	11.56	1.70×10^{-10}	TCA cycle
fumaric acid	G	1.66	28.54	1.05×10^{-8}	TCA cycle
adenine	P	1.13	21.15	1.54×10^{-9}	vitamin
carnitine	G	1.39	4.19	2.61×10^{-8}	vitamin
lipoic acid	P	1.47	0.10	4.90×10^{-8}	vitamin
niacinamide	P	1.45	32.38	4.59×10^{-4}	vitamin
orotic acid	G	1.28	12.58	7.09×10^{-8}	vitamin
pantothenic acid	N	1.15	1.81	1.73×10^{-6}	vitamin
glycocholic acid	P	1.41	0.11	2.29×10^{-7}	bile acid
hydroxycholic acid	P	1.57	0.10	3.65×10^{-8}	bile acid
4-methoxycinnamic acid	P	1.36	4.37	4.05×10^{-9}	other
acetylcystathionine	P	1.49	0.09	1.12×10^{-7}	other
aldosterone	P	1.35	0.16	2.28×10^{-6}	other
urea	G	1.84	22.18	2.79×10^{-9}	other

^a G: data from GC-TOFMS; N: data from UPLC-QTOFMS in negative mode; P: data from UPLC-QTOFMS in positive mode; ^b VIP (variable importance in projection) score from PLS-DA model; ^c FC (fold change) of different metabolites in formula milk compared to human breast milk; ^d p-value from the Mann–Whitney U test; ^e below the limit of detection.

Table S2. The differential metabolites obtained from multivariate and univariate analyses of bovine milk relative to human breast milk.

Metabolites	Platform ^a	VIP ^b	FC ^c	p-Value ^d	Class
5-oxoproline	G	1.39	0.32	3.19×10^{-9}	amino acid
alanine	G	1.37	0.36	7.18×10^{-8}	amino acid
arginine	P	1.24	0.04	6.92×10^{-8}	amino acid
glutamic acid	G	1.52	0.35	2.83×10^{-9}	amino acid
glutamine	G	1.29	0.18	1.90×10^{-7}	amino acid
phenylalanine	G	1.42	25.79	1.82×10^{-11}	amino acid
proline	G	1.22	2.3	7.39×10^{-7}	amino acid
serine	G	1.55	0.13	2.82×10^{-9}	amino acid
taurine	N	1.33	0.10	2.82×10^{-9}	amino acid
tyrosine	P	1.36	0.001	4.78×10^{-8}	amino acid
valine	G	1.31	0.34	4.81×10^{-8}	amino acid
3'-ketolactose	N	1.26	3.30	6.68×10^{-7}	carbohydrate
4-ketoglucose	G	1.35	0.18	9.52×10^{-8}	carbohydrate
acetylglucosamine	P	1.74	12.64	4.92×10^{-9}	carbohydrate
allofuranose	G	1.35	17.07	2.49×10^{-9}	carbohydrate
arabinose	N	1.13	3.36	3.30×10^{-6}	carbohydrate
fucose	G	1.41	0.11	5.77×10^{-6}	carbohydrate
glucosamine	N	1.22	1.56	4.95×10^{-9}	carbohydrate
glyceraldehyde	N	1.04	1.97	4.97×10^{-9}	carbohydrate
maltose	G	1.13	0.20	2.04×10^{-4}	carbohydrate

Table S2. Cont.

Metabolites	Platform ^a	VIP ^b	FC ^c	<i>p</i> -Value ^d	Class
mannose	G	1.35	16.38	4.97×10^{-9}	carbohydrate
ribose	G	1.34	1.44	2.60×10^{-7}	carbohydrate
arachidonic acid	N	1.58	ND ^e	8.70×10^{-19}	fatty acid
capric acid	G	1.49	0.04	5.51×10^{-15}	fatty acid
docosadienoic acid	N	1.62	ND	3.91×10^{-19}	fatty acid
docosahexaenoic acid	P	1.24	ND	1.01×10^{-8}	fatty acid
docosapentaenoic acid	N	1.35	ND	7.68×10^{-11}	fatty acid
eicosadienoic acid	G	1.35	ND	8.23×10^{-9}	fatty acid
eicosapentaenoic acid	N	1.04	ND	5.72×10^{-6}	fatty acid
eicosatrienoic acid	G	1.61	ND	8.93×10^{-21}	fatty acid
eicosenoic acid	G	1.09	0.04	1.47×10^{-6}	fatty acid
lauric acid	G	1.32	0.04	4.21×10^{-9}	fatty acid
linoleic acid	N	1.72	ND	7.12×10^{-15}	fatty acid
α -linolenic acid	N	1.47	ND	3.18×10^{-14}	fatty acid
γ -linolenic acid	N	1.69	ND	8.32×10^{-13}	fatty acid
myristic acid	N	1.44	0.17	2.83×10^{-13}	fatty acid
myristoleic acid	G	1.10	0.07	1.21×10^{-6}	fatty acid
oleic acid	G	1.27	ND	1.89×10^{-8}	fatty acid
palmitic acid	G	1.33	31.87	3.41×10^{-15}	fatty acid
palmitoleic acid	P	1.55	ND	2.54×10^{-17}	fatty acid
pentadecanoic acid	N	1.23	0.44	1.47×10^{-8}	fatty acid
stearic acid	G	1.18	3.15	4.77×10^{-6}	fatty acid
tridecanoic acid	G	1.13	ND	4.30×10^{-7}	fatty acid
2-ketoglutaramic acid	G	1.44	28.24	5.21×10^{-9}	TCA cycle
cis-aconitic acid	G	1.62	4.08	4.97×10^{-9}	TCA cycle
citric acid	N	1.65	15.07	4.96×10^{-9}	TCA cycle
fumaric acid	G	1.62	34.53	2.29×10^{-10}	TCA cycle
malic acid	G	1.70	13.05	4.97×10^{-9}	TCA cycle
biotin	P	1.49	2.79	4.97×10^{-9}	vitamin
lipoic acid	P	1.40	0.09	4.95×10^{-9}	vitamin
orotic acid	G	1.26	10.78	1.14×10^{-2}	vitamin
tocopherol	N	1.23	0.01	3.53×10^{-7}	vitamin
glycocholic acid	P	1.44	0.03	1.32×10^{-8}	bile acid
hydroxycholic acid	P	1.47	0.03	2.33×10^{-8}	bile acid
2-ethoxy-acetic acid	N	1.52	0.005	1.98×10^{-9}	other
2-indolecarboxylic acid	N	1.64	0.78	4.97×10^{-9}	other
3-aminopropionaldehyde	N	1.45	1.67	7.15×10^{-9}	other
3-hydroxybutanoic acid	G	1.58	8.71	3.39×10^{-8}	other
4-methoxycinnamic acid	N	1.74	16.96	4.96×10^{-9}	other
cholesterol	G	1.40	2.36	8.53×10^{-8}	other
creatinine	P	1.57	2.23	4.97×10^{-9}	other
indole-5,6-quinone	N	1.42	0.57	7.19×10^{-9}	other
methylenebutanedioic acid	G	1.10	1.44	1.03×10^{-5}	other
pipecolic acid	G	1.53	3.32	8.05×10^{-9}	other
uracil	G	1.52	2.20	4.97×10^{-9}	other
uridine	G	1.23	2.61	9.81×10^{-7}	other

^a G: data from GC-TOFMS; N: data from UPLC-QTOFMS in negative mode; P: data from UPLC-QTOFMS in positive mode; ^b VIP (variable importance in projection) score from PLS-DA model; ^c FC (fold change) of different metabolites in bovine milk compared to human breast milk; ^d *p*-value from the Mann–Whitney U test; ^e below the limit of detection.