Maternal obesity during pregnancy leads to derangements in one-carbon metabolism and the gut microbiota

Rubini, Eleonora; Schenkelaars, Nicole; Rousian, Melek; Sinclair, Kevin D; Wekema, Lieske; Faas, Marijke M; Schoenmakers, Sam; Steegers, Regine

Published in:
American Journal of Obstetrics and Gynecology

DOI:
10.1016/j.ajog.2022.04.013

Publication date:
2022

Maternal obesity during pregnancy leads to derangements in one-carbon metabolism and the gut microbiota: implications for fetal development and offspring wellbeing

Eleonora Rubini, MSc; Nicole Schenkelaars, MD; Melek Rousian, MD, PhD; Kevin D. Sinclair, PhD; Lieske Wekema, MSc; Marijke M. Faas, PhD; Régine P. M. Steegers-Theunissen, MD, PhD; Sam Schoenmakers, MD, PhD

A healthy diet before and during pregnancy is beneficial in acquiring essential B vitamins involved in 1-carbon metabolism, and in maintaining a healthy gut microbiota. Each play important roles in fetal development, immune-system remodeling, and pregnancy-nutrient acquisition. Evidence shows that there is a reciprocal interaction between the one-carbon metabolism and the gut microbiota given that dietary intake of B vitamins has been shown to influence the composition of the gut microbiota, and certain gut bacteria also synthesize B vitamins. This reciprocal interaction contributes to the individual's overall availability of B vitamins and, therefore, should be maintained in a healthy state during pregnancy. There is an emerging consensus that obese pregnant women often have derangements in 1-carbon metabolism and gut dysbiosis owing to high intake of nutritiously poor foods and a chronic systemic inflammatory state. For example, low folate and vitamin B_{12} in obese women coincide with the decreased presence of B vitamin-producing bacteria and increased presence of inflammatory-associated bacteria from approximately mid-pregnancy. These alterations are risk factors for adverse pregnancy outcomes, impaired fetal development, and disruption of fetal growth and microbiota formation, which may lead to potential long-term offspring metabolic and neurologic disorders. Therefore, preconceptional and pregnant obese women may benefit from dietary and lifestyle counseling to improve their dietary nutrient intake, and from monitoring their B vitamin levels and gut microbiome by blood tests and microbiota stool samples. In addition, there is evidence that some probiotic bacteria have folate biosynthetic capacity and could be used to treat gut dysbiosis. Thus, their use as an intervention strategy for obese women holds potential and should be further investigated. Currently, there are many knowledge gaps concerning the relationship between one-carbon metabolism and the gut microbiota, and future research should focus on intervention strategies to counteract B vitamin deficiencies and gut dysbiosis in obese pregnant women, commencing with the use of probiotic and prebiotic supplements.

Key words: folate, microbiota, obesity, offspring, one-carbon metabolism, pregnancy

Introduction

The World Health Organization recommends pregnant women to adhere to a healthy diet, exercise regularly, and take 0.4-mg folic acid supplement per day to ensure a healthy pregnancy and improve pregnancy outcomes.[1] These guidelines are strongly recommended because, during pregnancy, there is a high demand for essential vitamins required to support basic cellular processes involved in developmental programming of fetal and maternal tissues.[2] These include B vitamins involved in one-carbon metabolism (eg, vitamin $B_{9/11}$ [folate] and

From the Department of Obstetrics and Gynaecology, Erasmus University Medical Center, Rotterdam, The Netherlands (Ms Rubini and Drs Schenkelaars, Rousian, Steegers-Theunissen, and Schoenmakers); School of Biosciences, University of Nottingham, Nottingham, United Kingdom (Dr Sinclair); and Division of Medical Biology, Department of Pathology and Medical Biology, University Medical Center Groningen, University of Groningen, Groningen, The Netherlands (Ms Wekema and Dr Faas).

Received Dec. 23, 2021; revised April 4, 2022; accepted April 7, 2022.

The authors report no conflict of interest.

This research was funded by the Department of Obstetrics and Gynaecology of the Erasmus Medical Center (Rotterdam), the European Union's Horizon 2020 research and innovation program under the Marie Skłodowska-Curie grant agreement number 812660 and the ZonMw grant Open Competition 2018 (09120011910046). K.D.S. received funding from the Biotechnology and Biological Sciences Research Council (BB/K017810/1).

Corresponding author: Régine P.M. Steegers-Theunissen, MD, PhD. r.steegers@erasmusmc.nl

• https://doi.org/10.1016/j.ajog.2022.04.013

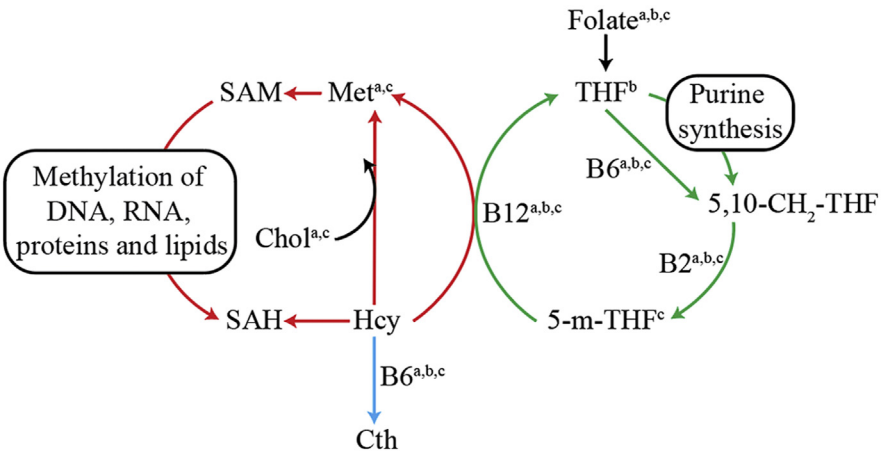
Click Video under article title in Contents at ajog.org

vitamin B_{12} [cobalamin]).[3] Dietary intake of B vitamins is known to influence and maintain a healthy maternal gut microbiota during pregnancy, which plays a major role in nutrient acquisition and absorption,[4] immune remodeling,[5,6] protection against infections,[7] and shaping the fetal immune system and gut microbiota.[8] In addition, certain strains of gut bacteria have been shown to biosynthesize B vitamins,[9,10] which demonstrates that one-carbon metabolism and the gut microbiota are involved in pregnancy health. Derangements in one-carbon metabolism, as a result of vitamin deficiencies, are a risk factor for developmental disorders and adverse pregnancy outcomes, including neural-tube defects (NTDs), miscarriages, preterm birth, and low birthweight.[11–13] Similarly, perturbations of the maternal gut microbiota (termed dysbiosis) are also a risk factor for adverse pregnancy outcomes, such as miscarriage and intrauterine growth restriction, and recent evidence highlights potential long-term effects of maternal dysbiosis on offspring metabolic and neurologic health, resulting in obesity and mental health disorders.[14–16]

The prevalence of obesity (body mass index [BMI] >30 kg/m^2) among pregnant women is increasing worldwide,[17] and obese pregnant women are more likely to adhere to an unhealthy diet, exercise less, and be less compliant with daily folic acid supplementation.[18] Therefore, it is common to find one-carbon metabolism deficiencies and gut dysbiosis in obese pregnant women, which expose the mother and fetus to the aforementioned health risks.[19] However, little is known about the interaction between the gut microbiota and one-carbon metabolism during pregnancy in obese and non-obese women.

Taking these issues into account, this article aims to: (1) summarize the evidence of interactions between obesity, one-carbon metabolism, and the gut microbiota; (2) provide evidence that gut dysbiosis mediates derangements in one-carbon metabolism in obese individuals; and (3) report the risks for offspring health in obese pregnancies. Understanding interactions between the gut microbiota and one-carbon metabolism in obese and non-obese women will favor more targeted interventional approaches to improve treatment during pregnancy, prevent adverse pregnancy outcomes, and improve offspring health.

FIGURE
One-carbon metabolism and the origin of its substrates and cofactors



One-carbon metabolism is composed of folate (*green arrows*) and methionine (*red arrows*) cycles and transsulfuration pathway (*blue arrow*). Metabolites within the pathway can originate from the diet (a), from gut microbiota (b), and/or from synthetic supplementation (c). Substrates: Bet, Chol, Cth, DHF, Hcy, 5,10-CH_2-THF, 5-m-THF, Met, THF, SAM, and SAH. Cofactors: vitamins B2, B6, and B12. Image adapted from a previous publication from the authors.[34]

Bet, betaine; *Chol*, choline; *Cth*, cystathione; *DHF*, dihydrofolate; *Hcy*, homocysteine; *5,10-CH$_2$-THF*, 5,10-methylenetetrahydrofolate; *5-m-THF*, 5-methyltetrahydrofolate; *Met*, methionine; *THF*, tetrahydrofolate; *SAM*, S-adenosylmethionine; *SAH*, S-adenosylhomocysteine.

Rubini. Maternal obesity during pregnancy leads to derangements in one-carbon metabolism and the gut microbiota. Am J Obstet Gynecol 2022.

One-Carbon Metabolism

One-carbon metabolism is composed of the interlinked folate and methionine cycles and the transsulfuration pathway, which are central to cellular function, providing substrates for DNA synthesis and epigenetic regulation (Figure). Folate from dietary, synthetic, or bacterial sources enters the folate cycle once reduced to its biologically active form, tetrahydrofolate (THF). THF is irreversibly reduced to 5-methyl-THF via B_6 and/or B_2 vitamin cofactors, providing purines for DNA repair. 5-methyl-THF can only enter the methionine cycle by donating a one-carbon unit for the remethylation of homocysteine to methionine via vitamin B_{12}. In the methionine cycle, methionine is converted to S-Adenosyl methionine, which serves as a universal methyl donor for biosynthetic and epigenetic processes. Lastly, homocysteine is remethylated to methionine if there is demand for tissue methyl donors, or enters the transsulfuration pathway where it is irreversibly transformed to cystathionine. Elevated homocysteine levels occur when there is insufficient folate or vitamin B_{12} to allow the conversion of homocysteine to methionine; therefore, homocysteine is perceived as a sensitive marker for derangements in one-carbon metabolism.[20–23]

B vitamins that drive the folate and methionine cycles are essential vitamins that cannot be produced by any cell of the human body. Consequently, sources of B vitamins arise from dietary components such as green vegetables, red meat, dairy, or synthetic supplementation or from the local gut microbes such as *Bacteroidetes*, *Proteobacteria*, and *Fusobacteria* (Table).

One-carbon metabolism during the periconceptional period and pregnancy

One-carbon metabolism plays a key role during the periconceptional period and

TABLE
Summary of most common bacterial phyla of the human gut microbiota and their role in health and disease

Phylum	Genera	One-carbon metabolites biosynthesis	Biological activity	Function	Health-related associations	Reference
Bacteroidetes[a]	Bacteroides Prevotella	Vitamin B_2, B_6 and B_{12} Folate	Butyrate production (fermentation) Bile acid metabolism Transformation of toxic compounds Degradation of carbohydrates	Protection against inflammation Nourishment of intestinal barrier	↓ in obesity ↓ with age ↑ in IBS	Magnúsdóttir et al,[81] 2015; Engevik et al,[57] 2019; Stojanov et al,[100] 2020; Thomas et al,[107] 2011; Ottman et al,[87] 2012; Vaiserman et al,[108] 2020; Santacruz et al,[109] 2010
Firmicutes[a]	Lactobacillus Bacillus Clostridium Enterococcus Ruminococcus	Vitamin B_2[b] Folate[c]	Butyrate production (fermentation)	Extraction of energy from food	↑ in obesity ↑ with age ↑ with weight gain	Magne et al,[69] 2020; Magnúsdóttir et al,[81] 2015; Ottman et al,[58] 2012; Vaiserman et al,[108] 2020
Actinobacteria	Bifidobacterium	Folate	Acetate production (fermentation)	Stimulation of the immune system Resistance to colonization by pathogens Probiotic properties in the colon	Beneficial for treating gastrointestinal diseases	Strozzi and Mogna,[82] 2008; Picard et al,[110] 2005
Proteobacteria	Escherichia coli	Vitamin B_2 Folate	Aerobic activity Minor anaerobic fermentation	Protection against inflammation and pathogens	↑ in intestinal diseases	Magnúsdóttir et al,[81] 2015; Rizzatti et al,[111] 2017; Shin et al,[112] 2015; Christofi et al,[113] 2019
Fusobacteria	Fusobacterium	Vitamin B_2 and B_{12} Folate	Anaerobic activity	Protection against pathogens	↑ in colorectal cancer	Magnúsdóttir et al,[81] 2015; Zhou et al,[114] 2018; Keyy et al,[115] 2018; Wu et al,[116] 2019
Verrucomicrobia	Akkermansia	N/A	Anaerobic activity	Mucus degradation	↑ in long-term fasting ↑ in malnutrition	Belzer et al,[117] 2012

DHF, dihydrofolic acid; *IBS*, irritable bowel syndrome; *N/A*, not applicable; *THF*, tetrahydrofolic acid.

[a] Bacteroidetes and Firmicutes constitute 90% of the gut bacteria population. The term folate comprises the active forms of folate DHF and THF; [b] Only 50% of Firmicutes have vitamin B_2 biosynthetic capacity; [c] Only *Lactobacillus* has sufficient evidence for folate production

Rubini. Maternal obesity during pregnancy leads to derangements in one-carbon metabolism and the gut microbiota. Am J Obstet Gynecol 2022.

pregnancy by providing substrates for the biosynthesis of DNA, RNA, proteins, and lipids, which are necessary for developmental processes such as cell replication and differentiation.[24–26] Adequate periconceptional folate and vitamin B_{12} levels increase the probability of fertilization success,[27] improve preimplantation embryo quality,[28] increase implantation rates, reduce the incidence of NTDs, and improve live birth rates.[29] In addition, folate requirements are higher in pregnancy because of uterine- and placental-cell proliferation, fetal-tissue differentiation and growth, and increased erythrocyte production.[30] Folate and/or vitamin B_{12} deficiencies can lead to adverse pregnancy outcomes. It is well established that folate deficiency is a severe risk factor for fetal NTDs and other congenital anomalies,[30–32] whereas vitamin B_{12} deficiency during early pregnancy is a risk factor for preterm birth and adverse offspring neurodevelopment.[11,12] Moreover, elevated homocysteine is associated with miscarriages, gestational hypertension, and preterm birth.[13] Therefore, pregnant women are strongly recommended to consume micronutrient-rich foods and use folic acid supplementation from as early as 5 to 6 months before conception up to 12 weeks of gestation, preferably continuing until the end of pregnancy, to enhance fertility, prevent pregnancy complications, and reduce the risk of impairments to fetal development.[24,33–35]

Obesity and one-carbon metabolism during pregnancy

Obesity is characterized by high adiposity, associated with a wide range of health risks including insulin resistance, cardiovascular diseases, and metabolic and endocrine imbalances, and contributes to a chronic inflammatory physiological state.[36,37] One of the etiologic factors that induces obesity is

excessive caloric input owing to consumption of an unbalanced diet predominantly composed of nutritionally poor and high-caloric food, which often leads to micronutrient deficiencies.[38] Consequently, derangements in one-carbon metabolism are more common in obese individuals.[39] In particular, low levels of vitamin B_{12} and folate are associated with high BMI,[40−42] and it is known that, despite regular folic acid supplement use, obese women retain low fasting serum folate levels and have a lower folate absorption phase than normal-weight women.[19,43] This is also observed in early, mid, and late pregnancy, when obese women tend to have lower serum vitamin B_{12} and folate concentrations, and higher odds of vitamin B_{12} and folate deficiency than non-obese pregnant women.[40,44−52] In fact, offspring of obese women have a 2-fold higher risk of being born with NTDs despite maternal folic acid supplement use.[53] With regard to homocysteine, increased levels during mid-pregnancy are observed with increasing prepregnancy BMI.[46,48] The emerging consensus is that the requirements for one-carbon metabolites during all stages of pregnancy are not met in obese women[19] despite micronutrient supplementation.[40] Therefore, levels of one-carbon metabolites should be monitored carefully and, if necessary, be improved by intervention strategies that involve lifestyle counseling with a focus on diet. For the time being, although the evidence is disputable, obese women are recommended to take higher doses of folic acid during pregnancy than non-obese women.[19]

The Gut Microbiota

The human gut microbiota consists of a complex and dynamic mixture of microorganisms, including bacteria, viruses, and fungi, which coexist in the gastrointestinal tract.[54] The gut microbiota is shaped in early life and more definitively established in adulthood, depending on age, ethnicity, antibiotics, medication, and, importantly, diet.[55] For the purpose of the current review, only bacterial microbiota will be addressed because of the availability of literature and our current understanding of its relationship to maternal one-carbon metabolism.

Bacterial microbiota perform essential activities such as food digestion, release of short-chain fatty acids for energy and metabolic processes, immunomodulation, and antimicrobial protection, and they are important suppliers of essential vitamins including intermediates of one-carbon metabolism.[56,57] An overview of the most important bacterial phyla and genera in the human gut and their primary roles in health and disease is provided in the Table. In general, healthy gut microbiota has a stable and balanced composition of *Firmicutes* and *Bacteroidetes* and, to lesser extent, *Actinobacteria, Proteobacteria,* and *Verrucomicrobia*.[56] *Firmicutes* are mostly responsible for carbohydrate metabolism and energy extraction, whereas *Bacteroidetes* have more mixed functions including degradation of polysaccharides, activation of the immune system, regulation of pathogenic gut bacteria, and transformation of toxic compounds.[58,59] *Actinobacteria*, *Proteobacteria*, and *Verrucomicrobia* are involved in decomposition of organic compounds, supporting the immune system, and mucus degradation.[59] Dysbiosis occurs when there is a disruption in the composition of the residential bacterial community, either in number or in phyla or genera predominance, resulting in loss of gut homeostasis and bacterial function, which is associated with inflammatory bowel diseases and metabolic disorders such as obesity and diabetes mellitus type 2.[60]

The maternal gut microbiota during pregnancy

A healthy maternal gut microbiota is essential during pregnancy because it contributes to obstetrical outcomes and long-term health sequelae for the mother and child. Various studies have shown that the gut microbiota remodels and fluctuates during pregnancy depending on gestational age. In particular, a substantial shift in bacteria phylogenies occurs from the second to third trimester, characterized by a reduction in bacterial richness (alpha diversity), with predominance of *Proteobacteria* and *Actinobacteria,* contributing to the inflammatory response in pregnancy, and a decrease in butyrate-producing bacteria, which have antiinflammatory effects (Table).[61−64] Rodent studies report the same phylogenetic patterns but a dominance of *Proteobacteria* in early pregnancy.[6,63,65] Other experimental studies have shown that transplantation of third-trimester microbiota into germ-free mice increases fat deposition, inflammation, and insulin sensitivity compared with transplantation of first-trimester microbiota into germ-free mice.[61,66] This suggests that the gut microbiota may adapt to sustain the superior metabolic demands of pregnancy and to support the immune system.[5,7] Recently, it has been shown that changes in the gut microbiota correlate to changes in the maternal immune response in pregnant mice, suggesting a role for the maternal microbiota in adapting the maternal immune response to pregnancy to protect the semiallogeneic fetus.[6] In contrast to studies showing changes in the maternal microbiota, there are also studies that report a relatively stable microbiota throughout gestation given that pregnant and nonpregnant women have a similar core microbiota of *Firmicutes* and *Bacteroidetes*, which are responsible for the provision of nutrition and vitamins to the host and other gut microbiota (Table).[67,68] Therefore, further research of the impact of pregnancy on maternal microbiota during pregnancy is necessary.

Obesity and the maternal gut microbiota during pregnancy

The composition of gut microbiota in obese pregnant women differs from that of normal-weight pregnant women. In obese women, dysbiosis is represented by an increase in *Firmicutes*, resulting in a high gut *Firmicutes*-to-*Bacteroidetes* ratio.[69] Many genera of the *Firmicutes* have enzymes that metabolize carbohydrates to extract energy from food, thus a high prevalence of *Firmicutes* may increase calorie absorption, predispose to weight gain, and is additionally a

symptom of gut inflammation and intestinal permeability.[69–72] Gomez et al[73] (2016) and Zacarias et al[74] (2018) report that obese pregnant women have a high *Firmicutes*-to-*Bacteroidetes* ratio, an increase in *Actinobacteria* in mid and late pregnancy, and a decrease in bacterial diversity in the third trimester of pregnancy compared with non-obese women.[73,74] According to this evidence, obesity during pregnancy can lead to a change in bacterial phyla that is not observed in non-obese pregnancies. Such microbial shifts are also observed in animal models including rodents.[65,74–77] Various studies have shown a causal role for the gut microbiota in obese individuals regarding weight gain and metabolic disturbances. Turnbaugh et al[78] (2006) demonstrated that germ-free mice inoculated with gut microbiota of obese mice, consisting of a high *Firmicutes*-to-*Bacteroidetes* ratio, gain more body fat and acquire metabolic disturbances. In addition, Ridaura et al[79] (2013) observed that transplanting human fecal microbiota from obese individuals to germ-free mice on a low-fat diet caused mice to develop increased fat mass and obesity-associated metabolic phenotypes. This was confirmed when microbiota from lean donors infused to recipients with metabolic syndrome (BMI >30 kg/m^2 and fasting plasma glucose >5.6 mmol/L) restored insulin sensitivity and increased butyrate-producing intestinal bacteria, exerting immunomodulatory and antiinflammatory properties.[80] More research is required to determine if this shift in bacterial populations is a consequence or cause of maternal obesity, and to determine its possible positive or negative impact on fetal growth and development.

Linking One-Carbon Metabolism to The Microbiota of Obese Pregnant Women

One-carbon metabolism and the gut microbiota are strongly connected. The reciprocal interaction between B vitamins of one-carbon metabolism and gut bacteria contributes to the overall availability of B vitamins and gut homeostasis. Despite diet being the best-known source of essential vitamins, several bacterial phyla produce B vitamins in quantities that match dietary intake.[81,82] One of the most prevalent phyla in the gut, *Bacteroidetes,* is the predominant producer of vitamin B_2, B_6, and B_{12} (Table). Furthermore, most B vitamins required for one-carbon metabolism play a role in supporting bacterial survival and fitness in the gut.[83] Animal and human studies report that vitamin B_6, folate, and B_{12} deficiencies contribute to disturbances of bacterial gut homeostasis.[10,84,85]

Gut dysbiosis and derangements in one-carbon metabolites occur more often in obese than non-obese pregnant women; however, it is not known if this is the cause or consequence of maternal obesity. Often, consumption of diets poor in micronutrients and rich in energy-dense food groups is associated with lower blood concentrations of folate and vitamin B_{12}.[86] This may be exacerbated by the changes in patterns of bacterial populations associated with such dietary regimes.[10,87] Low serum folate and vitamin B_{12} in pregnant obese women coincide with the decreased presence of B vitamin-producing *Bacteroidetes* and an increased prevalence of *Firmicutes* from mid-pregnancy onward.[73] Consequently, folate and vitamin B_{12} may be synthesized to a lesser extent because of the loss of bacterial richness and diversity. Despite this reasoning, given that there are many bacterial strains that synthesize one-carbon metabolic intermediates, it is hard to define the exact roles of each bacteria genus in pregnancy.

Offspring Health

Derangements of maternal one-carbon metabolism and maternal gut dysbiosis, present in obese pregnancies, represent a risk for fetal and offspring health. Low serum folate concentrations in obese women during late pregnancy are associated with an increased prevalence of fetal macrosomia, preterm birth, increased placental weight, and increased offspring BMI.[88,89] In addition, gut dysbiosis in obese women may be strongly associated with offspring predisposition to metabolic and neurodevelopmental abnormalities, and debate continues on whether or not this is mediated by the offspring microbiota.[90,91] Current literature seems to support that the fetal microbiota is colonized by the maternal bacteria via transplacental transport.[92] These observations are supported by rodent studies, which reveal that high-fat diets lead to gut dysbiosis and offspring with altered birthweight, fat mass, and percentage of body fat.[6,90,93,94] In conclusion, although not much is known about the effects of derangements in one-carbon metabolism and maternal gut dysbiosis on offspring health in obese pregnancies, these factors seem to play an important role in offspring weight and body fat content, and should therefore be rigorously monitored as early in pregnancy as possible to avoid dysbiosis and micronutrient deficiencies known to occur from mid-pregnancy.

Clinical Implications

It is clinically relevant to know that B vitamin availability depends on multiple factors, such as diet, metabolism and gut microbiota, which opens the window for multiple treatment strategies. In this case, if vitamin supplementation is insufficient to restore B vitamin levels in obese women, microbiota homeostasis can be considered as a second target for treating the deficiency. Currently, there is no effective intervention strategy for (obese) pregnant women with derangements in one-carbon metabolism and/or gut dysbiosis. Lifestyle care, including a combination of dietary advice and folic acid/multivitamin supplement use, is currently the most used treatment approach.[95,96] However, its effectiveness is either variable or unknown. Alongside lifestyle care tailored to obese pregnant (or preconceptional) women, we would advise to integrate microbiota analyses of stool samples and blood tests for one-carbon–related metabolites/cofactors throughout pregnancy, particularly homocysteine plasma levels given that this is a sensitive marker of derangements in one-carbon metabolism. Currently, several stool sampling methods are used to analyze the gut microbial population, and they could be

integrated in clinical practice during pregnancy visits.[97] The 2 must be independently but simultaneously monitored because a healthy microbiota does not exclude derangements in one-carbon metabolism and vice versa.

Furthermore, the field is now focusing on the use of probiotics and/or prebiotics as an intervention strategy in the treatment of gut dysbiosis. Probiotic bacteria (most common are *Lactobacillus* and *Bifidobacterium* species) confer long-lasting health benefits when administered in adequate quantities, such as when treating chronic bowl diseases and infections.[98,99] *Lactobacillus* and *Bifidobacterium* species have folate biosynthetic capacity and are used to restore the *Firmicutes*-to-*Bacteroidetes* ratio in gut dysbiosis.[100] In addition, adding probiotics to the diet can favor an increase in bacterial diversity, which counteracts the loss of diversity seen in gut microbiomes of obese patients. Conversely, prebiotics are substrates (mostly nondigestible carbohydrates) that are selectively utilized by gut microorganisms to nourish probiotic bacteria and other gut bacteria.[101] Indeed, the consumption of prebiotic-rich foods has been shown to support probiotic bacteria activity, particularly by increasing gut microbiota concentrations of *Lactobacillus* and *Bifidobacterium* species.[102] Currently, there is contradictory evidence on the efficacy of probiotic and prebiotic interventions during pregnancy, particularly because the doses have to be tailored to the composition of the host microbiota, which is individual-specific.[103] However, both have been demonstrated to be safe for use during pregnancy.[103−105] The use of probiotics and prebiotics during pregnancy in obese women holds potential and should be investigated because there is currently no evidence that they confer substantial benefits for obese pregnant women.

Future Studies

From the foregoing discussion, we can conclude that there are numerous knowledge gaps concerning the relationship between one-carbon metabolism and the gut microbiota in nonobese and obese pregnancies, although it is known that both play a major role in pregnancy health and fetal development, and the subsequent long-term health of offspring. It would be important to know which specific bacteria are associated with low serum folate and vitamin B_{12} during pregnancy. Future research should focus on intervention strategies, such as blended periconception lifestyle care approaches,[106] to detect and counteract B vitamin deficiencies and gut dysbiosis in obese pregnant women, starting with the use of probiotic supplements. Lastly, given that current knowledge is based on maternal obesity only, further studies need to address other potential mechanisms and the relationship between one-carbon metabolism and gut bacteria in other circumstances prone to vitamin deficiencies, such as in patients with metabolic disorders, those who have a family history of pregnancy complications, or those who harbor genetic polymorphisms in any one of a number of one-carbon metabolism genes.

Conclusion

In this review, we propose that the association between one-carbon metabolism and the gut microbiota contributes to the overall availability of B vitamins and to gut homeostasis of the individual, which subsequently determines maternal and fetal health during pregnancy. Despite limited evidence, it seems that dietary patterns in obese pregnancies are associated with derangements in one-carbon metabolism, aggravated by changes in patterns of bacterial populations associated with poor diets. These are risk factors for adverse pregnancy outcomes, impaired fetal development, and disruption of fetal growth and microbiota formation, leading to potential long-term offspring metabolic and neurologic disorders. Further studies are required to find effective intervention strategies for derangements in one-carbon metabolism and gut dysbiosis tailored to obese pregnant women. Disturbances in metabolism that are detected during early pregnancy or even during the preconceptional period create a window of opportunity for clinicians to provide lifestyle care and to prevent adverse pregnancy outcomes. ■

ACKNOWLEDGMENTS

We offer our special thanks to Wichor Bramer, PhD, for his assistance with the literature search.

REFERENCES

1. Tunçalp Ö, Pena-Rosas JP, Lawrie T, et al. WHO recommendations on antenatal care for a positive pregnancy experience-going beyond survival. BJOG 2017;124:860–2.
2. Cetin I, Bühling K, Demir C, et al. Impact of micronutrient status during pregnancy on early nutrition programming. Ann Nutr Metab 2019;74:269–78.
3. Clare CE, Brassington AH, Kwong WY, Sinclair KD. One-carbon metabolism: linking nutritional biochemistry to epigenetic programming of long-term development. Annu Rev Anim Biosci 2019;7:263–87.
4. Glenwright AJ, Pothula KR, Bhamidimarri SP, et al. Structural basis for nutrient acquisition by dominant members of the human gut microbiota. Nature 2017;541:407–11.
5. Kamada N, Seo S, Chen GY, Núñez G. Role of the gut microbiota in immunity and inflammatory disease. Nat Rev Immunol 2013;13:321–35.
6. Faas MM, Liu Y, Borghuis T, van Loo-Bouwman CA, Harmsen H, de Vos P. Microbiota induced changes in the immune response in pregnant mice. Front Immunol 2019;10:2976.
7. Khosravi A, Yáñez A, Price JG, et al. Gut microbiota promote hematopoiesis to control bacterial infection. Cell Host Microbe 2014;15:374–81.
8. Nyangahu DD, Lennard KS, Brown BP, et al. Disruption of maternal gut microbiota during gestation alters offspringmicrobiota and immunity. Microbiome 2018;6:124.
9. Gurwara S, Ajami NJ, Jang A, et al. Dietary nutrients involved in one-carbon metabolism and colonic mucosa-associated gut microbiome in individuals with an endoscopically normal colon. Nutrients 2019;11:613.
10. Lurz E, Horne RG, Määttänen P, et al. Vitamin B12 deficiency alters the gut microbiota in a murine model of colitis. Front Nutr 2020;7:83.
11. Rogne T, Tielemans MJ, Chong MF, et al. Associations of maternal vitamin B12 concentration in pregnancy with the risks of preterm birth and low birth weight: a systematic review and meta-analysis of individual participant data. Am J Epidemiol 2017;185:212–23.
12. Molloy AM, Kirke PN, Brody LC, Scott JM, Mills JL. Effects of folate and vitamin B12 deficiencies during pregnancy on fetal, infant, and child development. Food Nutr Bull 2008;29:S101–11.
13. Mascarenhas M, Habeebullah S, Sridhar MG. Revisiting the role of first trimester homocysteine as an index of maternal and fetal outcome. J Pregnancy 2014;2014:123024.

14. Zhang D, Huang Y, Ye D. Intestinal dysbiosis: an emerging cause of pregnancy complications? Med Hypotheses 2015;84:223–6.
15. Zhang Z, Xue C, Ju M, et al. Maternal gut dysbiosis alters offspring microbiota and social interactions. Microorganisms 2021;9:1742.
16. Di Gesù CM, Matz LM, Buffington SA. Diet-induced dysbiosis of the maternal gut microbiome in early life programming of neurodevelopmental disorders. Neurosci Res 2021;168:3–19.
17. Poston L, Caleyachetty R, Cnattingius S, et al. Preconceptional and maternal obesity: epidemiology and health consequences. Lancet Diabetes Endocrinol 2016;4:1025–36.
18. Masho SW, Bassyouni A, Cha S. Pre-pregnancy obesity and non-adherence to multivitamin use: findings from the National Pregnancy Risk Assessment Monitoring System (2009-2011). BMC Pregnancy Childbirth 2016;16:210.
19. van der Windt M, Schoenmakers S, van Rijn B, Galjaard S, Steegers-Theunissen R, van Rossem L. Epidemiology and (patho)physiology of folic acid supplement use in obese women before and during pregnancy. Nutrients 2021;13:331.
20. McMullin MF, Young PB, Bailie KE, Savage GA, Lappin TR, White R. Homocysteine and methylmalonic acid as indicators of folate and vitamin B12 deficiency in pregnancy. Clin Lab Haematol 2001;23:161–5.
21. Vashi P, Edwin P, Popiel B, Lammersfeld C, Gupta D. Methylmalonic acid and homocysteine as indicators of vitamin B-12 Deficiencyin cancer. PLoS One 2016;11:e0147843.
22. Green R. Indicators for assessing folate and vitamin B-12 status and for monitoring the efficacyof intervention strategies. Am J Clin Nutr 2011;94:666S–72S.
23. Blom HJ, Smulders Y. Overview of homocysteine and folate metabolism. With special references to cardiovascular disease and neural tube defects. J Inherit Metab Dis 2011;34:75–81.
24. Steegers-Theunissen RP, Twigt J, Pestinger V, Sinclair KD. The periconceptional period, reproduction and long-term health of offspring: the importance of one-carbon metabolism. Hum Reprod Update 2013;19:640–55.
25. Cai S, Quan S, Yang G, et al. One carbon metabolism and mammalian pregnancy outcomes. Mol Nutr Food Res 2021;65:e2000734.
26. Kalhan SC. One carbon metabolism in pregnancy: impact on maternal, fetal and neonatal health. Mol Cell Endocrinol 2016;435:48–60.
27. Gaskins AJ, Afeiche MC, Wright DL, et al. Dietary folate and reproductive success among women undergoing assistedreproduction. Obstet Gynecol 2014;124:801–9.
28. Boxmeer JC, Macklon NS, Lindemans J, et al. IVF outcomes are associated with biomarkers of the homocysteine pathwayin monofollicular fluid. Hum Reprod 2009;24:1059–66.
29. Gaskins AJ, Chiu YH, Williams PL, et al. Association between serum folate and vitamin B-12 and outcomes of assistedreproductive technologies. Am J Clin Nutr 2015;102:943–50.
30. Greenberg JA, Bell SJ, Guan Y, Yu YH. Folic acid supplementation and pregnancy: more than just neural tube defect prevention. Rev Obstet Gynecol 2011;4:52–9.
31. Zhao W, Mosley BS, Cleves MA, Melnyk S, James SJ, Hobbs CA. Neural tube defects and maternal biomarkers of folate, homocysteine, and glutathione metabolism. Birth Defects Res A Clin Mol Teratol 2006;76:230–6.
32. Wilson RD; GENETICS COMMITTEE; MOTHERISK. Pre-conceptional vitamin/folic acid supplementation 2007: the use of folic acid in combination with a multivitamin supplement for the prevention of neural tube defects and other congenital anomalies. J Obstet Gynaecol Can 2007;29:1003–13.
33. Korsmo HW, Jiang X. One carbon metabolism and early development: a diet-dependentdestiny. Trends Endocrinol Metab 2021;32:579–93.
34. Rubini E, Baijens IMM, Horánszky A, et al. Maternal one-carbon metabolism during the periconceptional period and human foetal brain growth: a systematic review. Genes (Basel) 2021;12:1634.
35. Li B, Zhang X, Peng X, Zhang S, Wang X, Zhu C. Folic acid and risk of preterm birth: a meta-analysis. Front Neurosci 2019;13:1284.
36. Ellulu MS, Patimah I, Khaza'ai H, Rahmat A, Abed Y. Obesity and inflammation: the linking mechanism and the complications. Arch Med Sci 2017;13:851–63.
37. Sikaris KA. The clinical biochemistry of obesity-more than skin deep. Heart Lung Circ 2007;16(Suppl3):S45–50.
38. Damms-Machado A, Weser G, Bischoff SC. Micronutrient deficiency in obese subjectsundergoing low calorie diet. Nutr J 2012;11:34.
39. Kaidar-Person O, Person B, Szomstein S, Rosenthal RJ. Nutritional deficiencies in morbidly obese patients: a new form of malnutrition? Part A: vitamins. Obes Surg 2008;18:870–6.
40. Knight BA, Shields BM, Brook A, et al. Lower circulating B12 is associated with higher obesity and insulin resistance during pregnancy in a non-diabetic white British population. PLoS One 2015;10:e0135268.
41. Kimmons JE, Blanck HM, Tohill BC, Zhang J, Khan LK. Associations between body mass index and the prevalence of low micronutrient levels among US adults. MedGenMed 2006;8:59.
42. Mojtabai R. Body mass index and serum folate in childbearing age women. Eur J Epidemiol 2004;19:1029–36.
43. Da Silva VR, Hausman DB, Kauwell GP, et al. Obesity affects short-term folate pharmacokinetics in women of childbearing age. Int J Obes (Lond) 2013;37:1608–10.
44. O'Malley EG, Reynolds CME, Cawley S, Woodside JV, Molloy AM, Turner MJ. Folate and vitamin B12 levels in early pregnancy and maternal obesity. Eur J Obstet Gynecol Reprod Biol 2018;231:80–4.
45. Scholing JM, Olthof MR, Jonker FA, Vrijkotte TG. Association between pre-pregnancy weight status and maternal micronutrient status in early pregnancy. Public Health Nutr 2018;21:2046–55.
46. Bjørke-Monsen AL, Ulvik A, Nilsen R, et al. Impact of pre-pregnancy BMI on B vitamin and inflammatory status in early pregnancy: an observational cohort study. Nutrients 2016;8:776.
47. Shen M, Chaudhry SH, MacFarlane AJ, et al. Serum and red-blood-cell folate demonstrate differential associations with BMI in pregnant women. Public Health Nutr 2016;19:2572–9.
48. Adaikalakoteswari A, Wood C, Mina TH, et al. Vitamin B12 deficiency and altered one-carbon metabolites in early pregnancy is associated with maternal obesity and dyslipidaemia. Sci Rep 2020;10:11066.
49. Kim H, Hwang JY, Kim KN, et al. Relationship between body-mass index and serum folate concentrations in pregnant women. Eur J Clin Nutr 2012;66:136–8.
50. Sukumar N, Venkataraman H, Wilson S, et al. Vitamin B12 status among pregnant women in the UK and its association with obesity and gestational diabetes. Nutrients 2016;8:768.
51. Martino J, Segura MT, García-Valdés L, et al. The impact of maternal pre-pregnancy body weight and gestational diabetes on markers of folate metabolism in the placenta. Nutrients 2018;10:1750.
52. Zhou YB, Si KY, Li HT, et al. Trends and influencing factors of plasma folate levels in Chinese women at mid-pregnancy, late pregnancy and lactation periods. Br J Nutr 2021;126:885–91.
53. Rasmussen SA, Chu SY, Kim SY, Schmid CH, Lau J. Maternal obesity and risk of neural tube defects: a metaanalysis. Am Jobstet Gynecol 2008;198:611–9.
54. Valdes AM, Walter J, Segal E, Spector TD. Role of the gut microbiota in nutrition and health. BMJ 2018;361:k2179.
55. Thursby E, Juge N. Introduction to the human gut microbiota. Biochem J 2017;474:1823–36.
56. Jandhyala SM, Talukdar R, Subramanyam C, Vuyyuru H, Sasikala M, Nageshwar Reddy D. Role of the normal gut microbiota. World J Gastroenterol 2015;21:8787–803.
57. Hill MJ. Intestinal flora and endogenous vitamin synthesis. Eur J Cancer Prev 1997;6:S43–5.
58. Ottman N, Smidt H, de Vos WM, Belzer C. The function of our microbiota: who is out there and what do they do? Front Cell Infect Microbiol 2012;2:104.
59. Rinninella E, Raoul P, Cintoni M, et al. What is the healthy gut microbiota composition? A

changing ecosystem across age, environment, diet, and diseases. Microorganisms 2019;7:14.
60. Petersen C, Round JL. Defining dysbiosis and its influence on host immunity and disease. Cell Microbiol 2014;16:1024–33.
61. Koren O, Goodrich JK, Cullender TC, et al. Host remodeling of the gut microbiome and metabolic changes during pregnancy. Cell 2012;150:470–80.
62. Ferrocino I, et al. Changes in the gut microbiota composition during pregnancy in patients withgestational diabetes mellitus (GDM). Sci Rep 2018;8:12216.
63. Smid MC, Ricks NM, Panzer A, et al. Maternal gut microbiome biodiversity in pregnancy. Am J Perinatol 2018;35:24–30.
64. Mor G, Cardenos I, Abrahams V, et al. Inflammation and pregnancy: the role of the immune system at the implantation site. Ann N Y Acad Sci 2011;1221:80–7.
65. Gohir W, Whelan FJ, Surette MG, Moore C, Schertzer JD, Sloboda DM. Pregnancy-related changes in the maternal gut microbiota are dependent uponthe mother's periconceptional diet. Gut Microbes 2015;6:310–20.
66. Mor G, Aldo P, Alvero AB. The unique immunological and microbial aspects of pregnancy. Nat Rev Immunol 2017;17:469–82.
67. DiGiulio DB, Callahan BJ, McMurdie PJ, et al. Temporal and spatial variation of the human microbiota during pregnancy. Proc Natl Acad Sci U S A 2015;112:11060–5.
68. Yang H, Guo R, Li S, et al. Systematic analysis of gut microbiota in pregnant women and its correlations with individual heterogeneity. NPJ Biofilms Microbiomes 2020;6:32.
69. Magne F, Gotteland M, Gauthier L, et al. The *Firmicutes/Bacteroidetes* ratio: a relevant marker of gut dysbiosis in obese patients? Nutrients 2020;12:1474.
70. Le Chatelier E, Nielsen T, Qin J, et al. Richness of human gut microbiome correlates with metabolic markers. Nature 2013;500:541–6.
71. Ley RE, Bäckhed F, Turnbaugh P, Lozupone CA, Knight RD, Gordon JI. Obesity alters gut microbial ecology. Proc Natl Acad Sci U S A 2005;102:11070–5.
72. de La Serre CB, Ellis CL, Lee J, Hartman AL, Rutledge JC, Raybould HE. Propensity to high-fat diet-induced obesity in rats is associated with changes in the gut microbiota and gut inflammation. Am J Physiol Gastrointest Liver Physiol 2010;299:G440–8.
73. Gomez-Arango LF, Barrett HL, McIntyre HD, et al. Connections between the gut microbiome and metabolic hormones inearly pregnancy in overweight and obese women. Diabetes 2016;65:2214–23.
74. Zacarías MF, Collado MC, Gómez-Gallego C, et al. Pregestational overweight and obesity are associated with differences in gut microbiota composition and systemic inflammation in the third trimester. PLoS One 2018;13:e0200305.
75. Connor KL, Chehoud C, Altrichter A, Chan L, DeSantis TZ, Lye SJ. Maternal metabolic, immune, and microbial systems in late pregnancy vary with malnutrition in mice. Biol Reprod 2018;98:579–92.
76. Mann PE, Huynh K, Widmer G. Maternal high fat diet and its consequence on the gut microbiome: a rat model. Gut Microbes 2018;9:143–54.
77. Li X, Rensing C, Taylor WL, et al. Papio spp. Colon microbiome and its link to obesity in pregnancy. J Med Primatol 2018;47:393–401.
78. Turnbaugh PJ, Ley RE, Mahowald MA, Magrini V, Mardis ER, Gordon JI. An obesity-associated gut microbiome with increased capacity for energyharvest. Nature 2006;444:1027–31.
79. Ridaura VK, Faith JJ, Rey FE, et al. Gut microbiota from twins discordant for obesity modulate metabolism in mice. Science 2013;341:1241214.
80. Vrieze A, Van Nood E, Holleman F, et al. Transfer of intestinal microbiota from lean donors increases insulin sensitivity in individuals with metabolic syndrome. Gastroenterology 2012;143:913–6.e7.
81. Magnúsdóttir S, Ravcheev D, de Crécy-Lagard V, Thiele I. Systematic genome assessment of B-vitamin biosynthesis suggests co-operation among gut microbes. Front Genet 2015;6:148.
82. Strozzi GP, Mogna L. Quantification of folic acid in human feces after administration of Bifido bacterium probiotic strains. J Clin Gastroenterol 2008;42:S179–84.
83. Uebanso T, Shimohata T, Mawatari K, Takahashi A. Functional roles of B-vitamins in the gut and gut microbiome. Mol Nutr Food Res 2020;64:e2000426.
84. Mayengbam S, Chleilat F, Reimer RA. Dietary vitamin B6 deficiency impairs gut microbiota and host and microbial metabolites in rats. Biomedicines 2020;8:469.
85. Kok DE, Steegenga WT, Smid EJ, Zoetendal EG, Ulrich CM, Kampman E. Bacterial folate biosynthesis and colorectal cancer risk: more than just a gutfeeling. Crit Rev Food Sci Nutr 2020;60:244–56.
86. Allen LH. Causes of vitamin B12 and folate deficiency. Food Nutr Bull 2008;29:S20–34.
87. Engevik MA, Morra CN, Röth D, et al. Microbial metabolic capacity for intestinal folate production and modulation of host folate receptors. Front Microbiol 2019;10:2305.
88. Berglund SK, García-Valdés L, Torres-Espinola FJ, et al. Maternal, fetal and perinatal alterations associated with obesity, overweight and gestational diabetes: an observational cohort study (PREOBE). BMC Public Health 2016;16:207.
89. Wang S, Ge X, Zhu B, et al. Maternal continuing folic acid supplementation after the first trimester of pregnancy increased the risk of large-for-gestational-age birth: a population-based birth cohort study. Nutrients 2016;8:493.
90. Bruce-Keller AJ, Fernandez-Kim SO, Townsend RL, et al. Maternal obese-Type gut microbiota differentially impact cognition,anxiety and compulsive behavior in male and female offspring in mice. PLoS One 2017;12:e0175577.
91. Bhagavata Srinivasan SP, Raipuria M, Bahari H, Kaakoush NO, Morris MJ. Impacts of diet and exercise on maternal gut microbiota are transferred to offspring. Front Endocrinol (Lausanne) 2018;9:716.
92. Miko E, Csaszar A, Bodis J, et al. The maternal-fetal gut microbiota axis: physiological changes, dietary influence, and modulation possibilities. Life (Basel) 2022;12:424.
93. Paul HA, Bomhof MR, Vogel HJ, Reimer RA. Diet-induced changes in maternal gut microbiota and metabolomic profiles influence programming of offspring obesity risk in rats. Sci Rep 2016;6:20683.
94. Paul HA, Collins KH, Bomhof MR, Vogel HJ, Reimer RA. Potential impact of metabolic and gut microbial response to pregnancy and lactation in lean and diet-induced obese rats on offspring obesity risk. Mol Nutr Food Res 2018;62.
95. Oostingh EC, Koster MPH, van Dijk MR, et al. First effective mHealth nutrition and lifestyle coaching program for subfertile couples undergoing in vitro fertilization treatment: a single-blinded multicenter randomized controlled trial. Fertil Steril 2020;114:945–54.
96. van Dijk MR, Koster MPH, Oostingh EC, Willemsen SP, Steegers EAP, Steegers-Theunissen RPM. A mobile app lifestyle intervention to improve healthy nutrition in women before and during early pregnancy: single-center randomized controlled trial. J Med Internet Res 2020;22:e15773.
97. Short MI, Hudson R, Besasie BD, et al. Comparison of rectal swab, glove tip, and participant-collected stool techniques for gut microbiome sampling. BMC Microbiol 2021;21:26.
98. Fijan S. Microorganisms with claimed probiotic properties: an overview of recent literature. Int J Environ Res Public Health 2014;11:4745–67.
99. Palumbo VD, Romeo M, Marino Gammazza A, et al. The long-term effects of probiotics in the therapy of ulcerative colitis: a clinical study. Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub 2016;160:372–7.
100. Stojanov S, Berlec A, Štrukelj B. The influence of probiotics on the Firmicutes/Bacteroidetes ratio in the treatment of obesity and inflammatory bowel disease. Microorganisms 2020;8:1715.
101. Olvera-Rosales LB, Cruz-Guerrero AE, Ramírez-Moreno E, et al. Impact of the gut microbiota balance on the health-disease relationship: the importance of consuming probiotics and prebiotics. Foods 2021;10:1261.
102. Gibson GR, Probert HM, Loo JV, Rastall RA, Roberfroid MB. Dietary modulation of the human colonic microbiota: updating the concept of prebiotics. Nutr Res Rev 2004;17:259–75.

103. Baldassarre ME, Palladino V, Amoruso A, et al. Rationale of probiotic supplementation during pregnancy and neonatal period. Nutrients 2018;10:1693.
104. Sheyholislami H, Connor KL. Are probiotics and prebiotics safe for use during pregnancy and lactation? A systematic review and meta-analysis. Nutrients 2021;13:2382.
105. Jarde A, Lewis-Mikhael AM, Moayyedi P, et al. Pregnancy outcomes in women taking probiotics or prebiotics: a systematic review and meta-analysis. BMC Pregnancy Childbirth 2018;18:14.
106. van der Windt M, van der Kleij RM, Snoek KM, et al. Impact of a blended periconception lifestyle care approach on lifestyle behaviors: before-and-after study. J Med Internet Res 2020;22:e19378.
107. Thomas F, Hehemann JH, Rebuffet E, Czjzek M, Michel G. Environmental and gut *Bacteroidetes*: the food connection. Front Microbiol 2011;2:93.
108. Vaiserman A, Romanenko M, Piven L, et al. Differences in the gut Firmicutes to Bacteroidetes ratio across age groups in healthy Ukrainian population. BMC Microbiol 2020;20:221.
109. Santacruz A, Collado MC, García-Valdés L, et al. Gut microbiota composition is associated with body weight, weight gain and biochemical parameters in pregnant women. Br J Nutr 2010;104:83–92.
110. Picard C, Fioramonti J, Francois A, Robinson T, Neant F, Matuchansky C. Review article: Bifido bacteria as probiotic agents - physiological effects and clinical benefits. Aliment Pharmacol Ther 2005;22:495–512.
111. Rizzatti G, Lopetuso LR, Gibiino G, Binda C, Gasbarrini A. Proteobacteria: a Common Factor in Human Diseases. BioMed Res Int 2017;2017:9351507.
112. Shin NR, Whon TW, Bae JW. Proteobacteria: microbial signature of dysbiosis in gut microbiota. Trends Biotechnol 2015;33:496–503.
113. Christofi T, Panayidou S, Dieronitou I, Michael C, Apidianakis Y. Metabolic output defines Escherichia coli as a health-promoting microbe against intestinal Pseudomonas aeruginosa. Sci Rep 2019;9:14463.
114. Zhou Z, Chen J, Yao H, Hu H. Fusobacterium and colorectal cancer. Front Oncol 2018;8:371.
115. Kelly D, Yang L, Pei Z. Gut microbiota, Fusobacteria, and colorectal cancer. Diseases 2018;6:109.
116. Wu J, Li Q, Fu X. Fusobacterium nucleatum contributes to the carcinogenesis of colorectal cancer by inducing inflammation and suppressing host immunity. Transl Oncol 2019;12:846–51.
117. Belzer C, de Vos WM. Microbes inside–from diversity to function: the case of Akkermansia. ISME J 2012;6:1449–58.