Gut Microbiota and Infant Colic: is there a Place for Prebiotics, Probiotics, and Postbiotics?

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ABSTRACT

The consumption of probiotics, prebiotics, and postbiotics, or a combination of them, can contribute to maintaining a healthy intestinal microbiota as it allows the regulation of its dysbiosis in the case of some diseases or disorders, mainly in functional gastrointestinal disorders (FGIDs). The gut microbiome is an essential player in the pathophysiology of FGIDs through its metabolic and nutritional functions, the maintenance of intestinal mucosal integrity, and the regulation of the immune response. Research results thus far indicate that probiotics, prebiotics, and postbiotics may have direct and clinically relevant immunomodulatory effects. There is evidence regarding the prescription of this family of biotics in healthy individuals to improve overall health and alleviate symptoms in many conditions like infantile colic.

The colonization and microbiota establishment begins at birth; the first 2-3 years of life are critical for developing an abundant and diverse microbial community. Several scientific studies performed by traditional culture-dependent techniques and more recently by molecular techniques have observed differences in the bacterial populations of healthy infants and those suffering from FGIDs, the latter characterized by an increase in pathogenic species and a lower population of bifidobacteria and lactobacilli, compared to the former. In this context, the intestinal microbiota plays a leading role in the onset of these disorders, including infantile colic, through its metabolic and nutritional functions, maintenance of the integrity of the intestinal mucosa, and regulation of the immune response. That has opened the door to the study of prebiotics, probiotics, and postbiotics usage in the treatment and or prevention of infantile FGIDs. Vaginal and term delivery and breastfeeding are fundamental in the constitution of a healthy microbiota. As supportive tools, there are efficacy studies that support the administration of this family of biotics, mainly in cases where lactation is not possible or is limited.

Key words: infant colic, intestinal microbiota, probiotics, prebiotics, postbiotics.

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INTRODUCTION

The intestine: the anatomical site with the most abundance and diversity of microorganisms and immunological cells

The first 1000 days of life is a pediatric concept that implies the period considered between fertilization and the baby's first two years, a fundamental stage in which a window of opportunity opens for proper gestation, birth, colonization of the child's body, training of the immune system, development of oral tolerance and the baby's cognitive and behavioral process. Everything happening in this period is crucial and decisive for the individual's health well beyond childhood and adolescence[1]. Hence, the importance of the activity of the gastrointestinal tract at the first contact with an antigen. Thus, as intestinal maturation proceeds, there are changes in its permeability that prevent the entry of the corresponding antigens. The physiology of humans depends strongly on a set of microorganisms called microbiota, which includes bacteria, yeasts, fungi, viruses, archaea, and protozoa. These microorganisms are present on the external surface (skin) and the mucous membranes, such as the intestine and vagina. The intestinal flora, today called intestinal microbiota, is the most studied and concentrates the most diversity and abundance of microbial species. It is appropriate to indicate that the term "microbiome" refers to the collection of genes present in the microorganisms of the microbiota and their activity, to the way in which they act among themselves and with the environment.

The colon is said to be a meeting place of two worlds: a world of microbes (the gut microbiota) and a world of immunological cells because over 70% of all human immunological cells live in the intestine, and their differentiation and training depend on the intestinal microbiota. The human microbiota is considered a "diffuse organ" that is acquired at birth and is inherited from the mother (depending on the type of birth and breastfeeding), and also depends on other factors such as the family environment, the environment (including pets present in the home) and the type of complementary feeding1 . The microbiota is established and microbiologically mature in the first 2-3 years of life after birth, although there are reports of the presence of microbial material (nonviable cells, cell walls, DNA) in the placenta, amniotic fluid, fetal membranes, and fetal gastrointestinal tract in healthy, normal pregnancies[2]. The primary maternal microbial contribution to the infant's intestinal colonization happens through its passage along the vaginal canal at birth; therefore, breast milk is the greatest and best source of microorganisms for the conformation of the baby's microbiota[3]. Due to its composition of nutrients, microorganisms, and oligosaccharides (formerly known as bifidogenic factors), breast milk is the best available food for the formation of the intestinal microbiota and the immunological maturation of the intestine[4]. Cesarean delivery and the use of antibiotics are associated with a higher prevalence, throughout childhood, of asthma, juvenile arthritis, inflammatory bowel diseases, immune deficiencies, overweight, obesity, allergies, eczema, and enteric and respiratory infections, among others[5]. The mechanisms that would explain these inflammatory and immunological disorders have to do with colonization of the intestine by proinflammatory microorganisms such as Enterococcus faecalis, Clostridium difficile, Campylobacter, Methanobrevibacter smithii instead of Lactobacillus, Bifidobacterium or Faecalibacterium prausnitzii[1]. Notably, Bifidobacterium is the dominant genus in the intestine of the healthy infant during the first year of life, so although the intestinal microbiota of infants born by vaginal delivery or cesarean section is similar after the first year of life, the dynamics of colonization by Bifidobacterium is different in the first months after delivery, since intestinal colonization by this genus is slower in those born by cesarean section[6]. This difference determines the development of the immune system; thus, cesarean delivery will have a lifelong impact on the risk of developing an immune disease[7].

In a study conducted in China, the longitudinal development, from day 1 to 6 months of life, of infants born by vaginal birth (VB) or cesarean section (CS), fed exclusively with breast milk (BF) or standard infant formula (IF) without prebiotics, was observed. The researchers identified three microbial clusters, dominated by Escherichia/Shigella-Streptococcus (cluster 1), Bifidobacterium-Escherichia/Shigella (cluster 2), and Bifidobacterium (cluster 3). The infants in the PV-LM group showed cluster 3 as predominant by six months of age. Breastfeeding managed to reverse slightly the dysbiosis induced by CP, with no cluster 1 microorganisms observed in this group. In the case of infants fed with IF, regardless of the type of delivery, significant proportions of cluster 3 bifidobacteria were also observed, but in a lower proportion than those in the PV-LM group[8].

In the first year of life, under ideal conditions, the brain increases an average of 1 gram per day while the microbiota settles in and begins to diversify at the time of introducing complementary feeding, recommended from 6 months of life, a period until which exclusive breastfeeding is advised, and that complementary feeding be sustained simultaneously until at least two years of age. During the first year of life, the installation and evolution of the microbiota, the maturation of the intestinal immune system, and brain development, with what this implies for the cognitive process, are three events that develop in parallel and are intimately interrelated through the so-called microbiota-intestine-brain axis. In this context, early administration of antibiotics can

cause the definitive elimination of some species of the genus Bifidobacterium[9,10], the most important microbial genus in the first year of life, responsible for guiding the immune maturation process[11]. A recent study associated the administration of antibiotics in the first two years of life with a higher incidence of food allergies, atopic dermatitis, diabetes, overweight, obesity, and celiac disease, among other chronic diseases and developmental disorders, up to the age of 14[12].

Functional gastrointestinal disorders with a focus on infantile colic

Pediatric functional gastrointestinal disorders (FGIDs) comprise a set of chronic or recurrent gastrointestinal symptoms not explained by structural or biochemical abnormalities, with severe consequences on the child's and family's quality of life. Such patients are identified by whether they belong to some of the symptomatic subgroups, which primarily rely on consensus opinions, called Rome Criteria. In 2016, the latest revision of these criteria, IV for adults and III for pediatrics, which diagnose FGIDs, was published[13]. Severity depends on the intensity of intestinal symptoms and other factors: association of gastrointestinal and extraintestinal symptomatology, degree of involvement, forms of perception, and behavior14. Children and adolescents fall into three categories or symptom groups: FGIDs associated with defecation, abdominal pain with vomiting, and aerophagia. The 2016 revision of the Rome IV Criteria[15,16] attempts to incorporate all the evidence available in the last ten years, such as abnormalities in mucosal immune function and the characteristics of the intestinal microbiota.

These new criteria are called "disorders of brain-gut interaction" and consider multifactorial aspects of the problem (clinical and psychosocial). Of course, the "proof of concept" about the role of the microbiota in FGIDs is still not definitively confirmed. However, the phrase "no evidence of organic disease" in the Rome III criteria has been replaced by "after appropriate medical evaluation, the symptoms no longer can be attributed to another medical condition." That gives room to consider new causalities. The overall prevalence of FGIDs in pediatrics is 20-40%, according to Rome III criteria sources in 2016: 10-20% for defecation disorders, 10-20% for abdominal pain-related disorders, and 0.5-4% in vomiting and aerophagia-related disorders[17].

The main FGID in infants is infantile colic, a group of behaviors characterized by prolonged crying, present in up to 25% of infants at six weeks of age18 and associated with an increased risk of recurrent abdominal pain and subsequent allergic disorders in childhood. One study evaluated the relationship between infantile colic and gastrointestinal, allergic, and psychological disorders in populations of 10-year-old children after severe colic19. We observed an association between infantile colic and recurrent abdominal pain ($p = 0.001$) and allergic disorders ($p < 0.05$) such as allergic rhinitis, conjunctivitis, asthmatic bronchitis, pollinosis, atopic eczema, and food allergy, ten years after the colic episodes. Likewise, sleep disorders, restlessness, aggressiveness, and feelings of supremacy were more frequent ($p < 0.05$) in children who suffered colic during early childhood. Thus, susceptibility to recurrent abdominal pain and allergic and psychological disorders in childhood can be increased by infant colic. In this context, severe infantile colic could be the early expression of some of the most common disorders later in childhood[20]. The underlying mechanisms of infantile colic are unclear due, in part, to the lack of animal models that allow reproducibility of the results.

Multiple pathophysiologies, including alterations of the intestinal microbiome, have been proposed as a cause of abdominal pain.

Savino et al. early studies with traditional culture-dependent microbiological techniques found that colicky infants were more often colonized by proinflammatory gram-negative anaerobic bacteria and less often by lactobacilli when compared to non-colicky infants [21,22].

Several subsequent molecular studies confirmed the enrichment of proinflammatory and gas-producing microbial species, such as those of the phylum Proteobacteria within the stools of colicky infants[23-25]. Often recommended interventions, such as Ethicon and maternal dietary manipulation, produced mixed results[26]. A recent systematic review with meta-analyses on TGIF and microbiota found alterations in microbial diversity, stability, and colonization patterns in colicky infants compared to healthy controls. Likewise, several studies found an increase in pathogenic species of the Proteobacteria phylum and, at the same time, a decrease in beneficial bacteria such as lactobacilli and bifidobacteria[27].

Prebiotics, synbiotics and postbiotics: definitions

The concept of prebiotics appeared in 1995 when Glenn Gibson and Marcel Roberfroid introduced the concept in a publication that was a true paradigm shift[28]. At that time, the so-called "gut flora" was already regarded as potentially playing an active role in the host's health, and there was interest in manipulating their composition towards a positively healthier microbial community. In their work, they sought to promote bacterial genera such as Bifidobacterium, which the scientists perceived as having health-promoting properties. Bifidobacteria are among the dominant bacterial populations in the gastrointestinal tract of humans, and their health benefits result from a complex dynamic interaction between them, with other members of the intestinal microbiota, and with the host[29]. These bacteria were first isolated in the early 20th century by Henry Tissier from the

fecal matter of healthy infants[30]. In this context, these researchers defined prebiotics as non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth and or activity of one or a limited number of bacterial species already residing in the colon. The intake of prebiotics can significantly modulate the colonic microbiota by increasing the number of specific bacteria and thus changing its composition. The non-digestible oligosaccharides, such as fructooligosaccharides (FOS), galactooligosaccharides (GOS), inulin, and others, are examples of prebiotics with proven beneficial effects[31].

Thus, supplementation with prebiotics can change the composition and metabolism of the intestinal microbiota, for example, through the promotion of bifidobacteria, something that several studies have correlated with a higher ratio between Lactobacillus Bifidobacterium and Enterobacteriaceae, and a modulated production of short-chain fatty acids. It is worth noting that breast milk contains more than 200 oligosaccharides (HMOs), the first to which the baby has exposure through breastfeeding, which impacts intestinal colonization, intestinal barrier function, and immunomodulation, among others. HMOs comprise five monomers (glucose, galactose, N-acetylglucosamine, fucose, or sialic acid) linked by different bonds, which give them great structural diversity [29,32]. Many resist infant digestion and pass to the large intestine, where the intestinal bacteria can use them.

Prebiotics such as GOS/FOS stimulate the growth of endogenous bifidobacteria[33] and those administered as probiotics together with prebiotics[34]. Bifidobacteria thus become predominant microorganisms in human feces and drive the maturation process of the immune system[11]. In addition, these prebiotics modulate lipid metabolism, most likely through fermentation products. In 2017, in the context of exponential growth of knowledge about the microbiota thanks to genetic tools such as massive sequencing of microbial DNA (metagenomics), knowing already that lactobacilli and bifidobacteria are not the only beneficial microorganisms naturally resident in the gut and that other microbiotas can be modified by specific prebiotics (such as those of the vagina or skin), a consensus paper was published, under the authorship led again by Prof. Glenn Gibson, now as a member of ISAPP (International Scientific Association for Probiotics and Prebiotics). This Consensus expanded the concept of prebiotics, which we now define as "substrates selectively utilized by host microorganisms that confer a health benefit"[31].

This definition expanded to include non-carbohydrate substances (such as phenolic compounds and phytochemicals), applications to body sites other than the gastrointestinal tract, and numerous food categories. It also retained the requirement for selective microbiota-mediated pathways and beneficial health effects substantiated by published efficacy studies. This consensus statement aims to generate an appropriate use of the term "prebiotic", so there is consistency and clarity in scientific publications, marketing of these products, regulatory regulation, and communication to consumers and health professionals. In 2020, ISAPP proposed a consensus definition of "synbiotics",stating that it is a mixture of live microorganisms and substrates selectively utilized by host microorganisms, which exert a beneficial effect when administered in adequate amounts [35]. A first observation is the denomination of "synbiotic" (instead of symbiotic), a neologism that seeks to represent the true meaning of the term, since a prebiotic and a probiotic, administered together, can exert their beneficial effects independently without necessarily establishing a symbiosis as the ecological concept assumes.

In this sense, ISAPP recognizes that there can be complementary synbiotics, which would be a probiotic and a prebiotic administered together, and synergistic synbiotics, involving the administration of a live microorganism and a non-digestible substrate (meaning they do not have individual studies that make it possible to classify them as probiotic and prebiotic).

Finally, and to complete this family of biotics, a new definition of postbiotics[36] was proposed as an alternative to the 2013 definition[37] due to the limitations it presented[36]. Although this terminology is relatively new, it refers to a phenomenon widely recognized in the field of probiotics and functional foods, which is the fact that some microorganisms, even in their non-viable or inactivated forms, and together with their cell fragments, metabolites, or fermentation products, are also capable of exerting some beneficial effects. The term postbiotic is divergent in the sense that - in the scientific literature - it has also been approached with numerous terms, such as heat-killed probiotics, thyndallized probiotics, ghost biotics, and paraprobiotics. This diversity of terms to refer to the same phenomenon is an obstacle when it comes to locating and grouping scientific papers for systematic reviews and meta-analyses to demonstrate their effectiveness, so ISAPP decided to create a new panel of specialists to discuss and propose a consensus definition of postbiotics. This panel suggested that a postbiotic is "a preparation of inanimate (non-viable) microorganisms and/or their components, that confers a health benefit to the host "[38]. These may include different components, such as metabolites, short-chain fatty acids, microbial cell fractions, functional proteins, exopolysaccharides, cell lysates, teichoic acid, peptidoglycan-derived neuropeptides, and or pili-like structures. It is relevant to note that the presence, in the product, of non-viable cells of the strain(s) in question is necessary to meet the definition.

For example, a pure inactivated microbial culture or an inactivated fermented product would qualify as a postbiotic if it has beneficial effects demonstrated by at

least one clinical study, while a cell-free supernatant, vaccines, or phages for phage therapy are outside the scope of this concept. No regulatory body has yet adopted the term postbiotic; however, there are already products on the market, such as infant formulas, that incorporate prebiotics and postbiotics[40] or products based on inactivated lactobacilli for the management of infantile diarrhea[41] or stress[42], indicating once again that regulatory aspects are generally behind technological developments and the frontier of science. It should be noted that postbiotics, being inactivated microorganisms or their cellular fractions, are incapable of reproducing and eventually generating infections in immunosuppressed populations or in which the intestinal barrier is not adequately strengthened, so they would offer possibilities for nutritional intervention in cases where intestinal translocation or worsening of local inflammation continues to be a concern[43]. Likewise, since they are products with non-viable microorganisms, they could have a longer shelf life, do not require a cold chain for logistics, and thus reach geographical regions that have difficulties in ensuring the cold chain necessary for the distribution and adequate storage of the products.

Potential of probiotics, prebiotics, and postbiotics for the nutritional approach to infant colic

Regarding the use of probiotics for the treatment of infant colic, efficacy studies reviewed by meta-analysis suggest their effectiveness, although their implementation needs to be decided by the particular strain[44]. L. reuteri DSM 17938 has several clinical efficiency studies, which have been compiled and analyzed in systematic investigations with meta-analyses[45] that support and promote its use in the context of infantile colic[45]. Some studies have also demonstrated the efficacy of other strains of lactobacilli, such as L. rhamnosus GG[46,47].

On the other hand, some prebiotics, such as the GOS:FOS mixture in a 9:1 ratio, have also shown a beneficial effect on the modulation of the intestinal microbiota, softening the stool and reducing colic symptoms [48-50]. The combination of prebiotics and postbiotics from a fermentative process, followed by thermal inactivation by spray drying of the strains used for fermentation, also demonstrated efficacy in the treatment of infant colic[50,51]. These probiotics, prebiotics, and postbiotics are marketed as ready-to-drink liquid food supplements (in drops), as powders for reconstitution, or also included in infant formulas.

In any case, the best nutritional choice for the prevention and or treatment of infant colic remains breast milk[52].

CONCLUSION

The combination of several factors such as term and vaginal delivery, exclusive breastfeeding until six months of age and sustained until two years of age, the interaction with the environment, and the non-use (as far as possible) of antibiotics and antacids during the first two years of life, allow the development and evolution of an ideal microbiota, dominated first by bifidobacteria, which then diversifies towards two years of age. All this enables immunological training and the development of oral tolerance, with significant beneficial effects at a cognitive and behavioral level, through the microbiota-gut-brain axis, and thus the prevention of GIFT and chronic non-communicable diseases.

On the other hand, beyond treatment of symptoms, dysbiosis regulation in FGIDs (mainly infant colic) through the use of probiotics, prebiotics, and postbiotics is a promising and scientifically supported therapeutic possibility. It is now clear that the gut microbiome is a central player in the pathophysiology of FGIDs through its effects on host physiological processes, although the precise mechanisms underlying microbial regulation are poorly elucidated and remain a very active area of research. The presentation of mild gastrointestinal symptoms and functional etiology is very usual in infants, and the best nutritional recommendation remains human milk. Even so, when breastfeeding is not possible for any reason, food supplements or infant formulas with probiotics, prebiotics, or postbiotics present scientific evidence to consider their implementation.

Conflict of interests: Gabriel Vinderola is engaged in technology liaison activities (training, consulting, lecturing in scientific meetings, quality control, and product development) with dairy and biotic industries. He is currently a member of the board of directors of the International Scientific Association of Probiotics and Prebiotics (ISAPP).

REFERENCES

1. Milani C, Duranti S, Bottacini F, et al. The first microbial colonizers of the human gut: composition, activities, and health implications of the infant gut microbiota. Microbiol Mol Biol Rev. 2017;81(4):e00036-17. https://doi.org/10.1128/MMBR.00036-17.
2. Kuperman AA, Koren O. Antibiotic use during pregnancy: how bad is it? BMC Med. 2016;14(1):91. https://doi.org/10.1186/s12916-016-0636-0.
3. Lyons KE, Ryan CA, Dempsey EM, et al. Breast milk, a source of beneficial microbes and associated benefits for infant health. Nutrients. 2020;12(4):1039. https://doi.org/10.3390/nu12041039.
4. Le Doare K, Holder B, Bassett A, et al. Mother's milk: a purposeful contribution to the development of the infant microbiota and immunity. Front Immunol. 2018;9:361. https://doi.org/10.3389/fimmu.2018.00361.

5. Tamburini S, Shen N, Wu HC, et al. The microbiome in early life: implications for health outcomes. Nat Med. 2016;22(7):713-722. https://doi.org/10.1038/nm.4142.
6. Reyman M, van Houten MA, van Baarle D, et al. Impact of delivery mode-associated gut microbiota dynamics on health in the first year of life. Nat Commun. 2019;10(1):4997. https://doi.org/10.1038/s41467-019-13014-7. Errata en: Nat Commun. 2019;10(1):5352. https://doi.org/10.1038/s41467-019-13373-1.
7. Francino MP. Birth mode-related differences in gut microbiota colonization and immune system development. Ann Nutr Metab. 2018;73 Suppl 3:12-16. https://doi.org/10.1159/000490842.
8. Guo C, Zhou Q, Li M, et al. Breastfeeding restored the gut microbiota in caesarean section infants and lowered the infection risk in early life. BMC Pediatr. 2020;20(1):532. https://doi.org/10.1186/s12887-020-02433-x.
9. Palleja A, Mikkelsen KH, Forslund SK, et al. Recovery of gut microbiota of healthy adults following antibiotic exposure. Nat Microbiol. 2018;3(11):1255-1256. https://doi.org/10.1038/s41564-018-0257-9.
10. McDonnell L, Gilkes A, Ashworth M, et al. Association between antibiotics and gut microbiome dysbiosis in children: systematic review and meta-analysis. Gut Microbes. 2021;13(1):1-18. https://doi.org/10.1080/19490976.2020.1870402.
11. Henrick BM, Rodriguez L, Lakshmikanth T, et al. Bifidobacteria-mediated immune system imprinting early in life. Cell. 2021;184(15):3884-3898. https://doi.org/10.1016/j.cell.2021.05.030.
12. Aversa Z, Atkinson EJ, Schafer MJ, et al. Association of infant antibiotic exposure with childhood health outcomes. Mayo Clin Proc. 2021;96(1):66-77. https://doi.org/10.1016/j.mayocp.2020.07.019.
13. Zeevenhooven J, Koppen IJ, Benninga MA. The new Rome IV criteria for functional gastrointestinal disorders in infants and toddlers. Pediatr Gastroenterol Hepatol Nutr. 2017;20(1):1-13. https://doi.org/10.5223/pghn.2017.20.1.1.
14. Mearin F, Ciriza C, Mínguez M, et al. Guía de práctica clínica: síndrome del intestino irritable con estreñimiento y estreñimiento funcional en adultos. Tratamiento (Parte 2). Med Gen Fam. 2017;6(2):69-85. https://doi.org/10.24038/mgyf.2017.015.
15. Vázquez Frias R. Actualidades del síndrome de intestino irritable en pediatría. Rev Gastrohnup. 2009;11(1):52-60.
16. Dorn SD, Kaptchuk TJ, Park JB, et al. A meta-analysis of the placebo response in complementary and alternative medicine trials of irritable bowel syndrome. Neurogastroenterol Motil. 2007;19(8):630-637. https://doi.org/10.1111/j.1365-2982.2007.00937.x.
17. Huertas-Ceballos A, Logan S, Bennett C, et al. Dietary interventions for recurrent abdominal pain (RAP) and irritable bowel syndrome (IBS) in childhood. Cochrane Database Syst Rev. 2008;(1):CD003019. https://doi.org/10.1002/14651858.CD003019.pub2. Actualizada en: Cochrane Database Syst Rev. 2009;(1):CD003019.
18. Wolke D, Bilgin A, Samara M. Systematic review and meta-analysis: fussing and crying durations and prevalence of colic in infants. J Pediatr. 2017;185:55-61.e4. https://doi.org/10.1016/j.jpeds.2017.02.020.
19. Savino F, Castagno E, Bretto R, et al. A prospective 10-year study on children who had severe infantile colic. Acta Paediatr. 2007;94(449):129-132. https://doi.org/:10.1111/J.1651-2227.2005.TB02169.X.
20. Savino F. Focus on infantile colic. Acta Pædiatrica. 2007;96(9):1259-1264. https://doi.org/10.1111/J.1651-2227.2007.00428.X.
21. Savino F, Cresi F, Pautasso S, et al. Intestinal microflora in breastfed colicky and non-colicky infants. Acta Paediatr. 2004;93(6):825-829.
22. Savino F, Bailo E, Oggero R, et al. Bacterial counts of intestinal Lactobacillus species in infants with colic. Pediatr Allergy Immunol. 2005;16(1):72-75. https://doi.org/10.1111/j.1399-3038.2005.00207.x.
23. Savino F, Cordisco L, Tarasco V, et al. Molecular identification of coliform bacteria from colicky breastfed infants. Acta Paediatr. 2009;98(10):1582-1588. https://doi.org/10.1111/j.1651-2227.2009.01419.x.
24. Rhoads JM, Fatheree NY, Norori J, et al. Altered fecal microflora and increased fecal calprotectin in infants with colic. J Pediatr. 2009;155(6):823-828.e1. https://doi.org/10.1016/j.jpeds.2009.05.012.
25. de Weerth C, Fuentes S, Puylaert P, et al. Intestinal microbiota of infants with colic: development and specific signatures. Pediatrics. 2013;131(2):e550-558. https://doi.org/10.1542/peds.2012-1449.
26. Harb T, Matsuyama M, David M, et al. Infant colic - what works: a systematic review of interventions for breast-fed infants. J Pediatr Gastroenterol Nutr. 2016;62(5):668-686. https://doi.org/10.1097/MPG.0000000000001075.
27. Hofman D, Kudla U, Miqdady M, et al. Faecal microbiota in infants and young children with functional gastrointestinal disorders: a systematic review. Nutrients. 2022;14(5):974. https://doi.org/10.3390/nu14050974.
28. Gibson GR, Roberfroid MB. Dietary modulation of the human colonic microbiota: introducing the concept of prebiotics. J Nutr. 1995;125(6):1401-1412. https://doi.org/10.1093/jn/125.6.1401.
29. Hidalgo-Cantabrana C, Delgado S, Ruiz L, et al. Bifidobacteria and their health-promoting effects. Microbiol Spectr. 2017;5(3). https://doi.org/10.1128/microbiolspec.BAD-0010-2016.
30. Tissier H. Traitement des infections intestinales par la méthode de la flore bácterienne de l'intestine. CR Soc Biol. 1906;60:359-361.
31. Gibson GR, Hutkins R, Sanders ME, et al. Expert consensus document: the International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat Rev Gastroenterol Hepatol. 2017;14(8):491-502. https://doi.org/10.1038/nrgastro.2017.75.
32. Cheng L, Akkerman R, Kong C, et al. More than sugar in the milk: human milk oligosaccharides as essential bioactive molecules in breast milk and current insight in beneficial effects. Crit Rev Food Sci Nutr. 2021;61(7):1184-1200. https://doi.org/10.1080/10408398.2020.1754756.
33. Skórka A, Pieścik-Lech M, Kołodziej M, Szajewska H. Infant formulae supplemented with prebiotics: Are they better than unsupplemented formulae? An updated systematic review. Br J Nutr. 2018;119(7):810-825. https://doi.org/10.1017/S0007114518000120.
34. Kosuwon P, Lao-Araya M, Uthaisangsook S, et al. A synbiotic mixture of scGOS/lcFOS and Bifidobacterium breve M-16V increases faecal Bifidobacterium in healthy young children. Benef Microbes. 2018;9(4):541-552. https://doi.org/10.3920/BM2017.0110.
35. Swanson KS, Gibson GR, Hutkins R, et al. The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of synbiotics. Nat Rev Gastroenterol Hepatol. 2020;17(11):687-701. https://doi.org/10.1038/s41575-020-0344-2.
36. Vinderola G, Sanders ME, Salminen S. The concept of postbiotics. Foods. 2022;11(8):1077. https://doi.org/10.3390/foods11081077.
37. Tsilingiri K, Rescigno M. Postbiotics: what else? Benef Microbes. 2013;4(1):101-107. https://doi.org/10.3920/BM2012.0046.
38. Salminen S, Collado MC, Endo A, et al. The International Scientific Association of Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of postbiotics. Nat Rev Gastroenterol Hepatol. 2021;18(9):649-667. https://doi.org/10.1038/s41575-021-00440-6.
39. Wegh CA, Geerlings SY, Knol J, et al. Postbiotics and their potential applications in early life nutrition and beyond. Int J Mol Sci. 2019;20:4673. https://doi.org/10.3390/ijms20194673.
40. Salminen S, Stahl B, Vinderola G, et al. Infant formula supplemented with biotics: current knowledge and future perspectives. Nutrients. 2020;12(7):1952. https://doi.org/10.3390/nu12071952.
41. Remes Troche MJ, Coss Adame E, Valdovinos Díaz AM, et al. Lactobacillus acidophilus LB: a useful pharmabiotic for the treatment of digestive disorders. Therap Adv Gastroenterol. 2020;13:1756284820971201. https://doi.org/10.1177/1756284820971201.
42. Nishida K, Sawada D, Kuwano Y, et al. Health benefits of Lactobacillus gasseri CP2305 tablets in young adults exposed to chronic stress: a randomized, double-blind, placebo-controlled study. Nutrients. 2019;11(8):1859. https://doi.org/10.3390/nu11081859.
43. Morniroli D, Vizzari G, Consales A, et al. Postbiotic supplementation for children and newborn's health. Nutrients. 2021;13(3):781. https://doi.org/10.3390/nu13030781.
44. Hjern A, Lindblom K, Reuter A, et al. A systematic review of prevention and treatment of infantile colic. Acte Paediatr. 2020;109(9):1733-1744. https://doi.org/10.1111/apa.15247.
45. Gutiérrez-Castrellón P, Indrio F, Bolio-Galvis A, et al. Efficacy of Lactobacillus reuteri DSM 17938 for infantile colic: Systematic review with network meta-analysis. Medicine (Baltimore). 2017;96(51):e9375. https://doi.org/10.1097/MD.0000000000009375.
46. Fatheree NY, Liu Y, Ferris M, et al. Hypoallergenic formula with Lactobacillus rhamnosus GG for babies with colic: a pilot study of recruitment, retention, and fecal biomarkers. World J Gastrointest Pathophysiol. 2016;7(1):160. https://doi.org/10.4291/WJGP.V7.I1.160.

47. Pärtty A, Lehtonen L, Kalliomäki M, et al. Probiotic Lactobacillus rhamnosus GG therapy and microbiological programming in infantile colic: a randomized, controlled trial. Pediatr Res. 2015;78(4):470-475. https://doi.org/10.1038/pr.2015.127.
48. Savino F, Palumeri E, Castagno E, et al. Reduction of crying episodes owing to infantile colic: A randomized controlled study on the efficacy of a new infant formula. Eur J Clin Nutr. 2006;60(11):1304-1310. https://doi.org/10.1038/sj.ejcn.1602457.
49. Savino F, Cresi F, Maccario S, et al. "Minor" feeding problems during the first months of life: effect of a partially hydrolysed milk formula containing fructo- and galacto-oligosaccharides. Acta Paediatr Suppl. 2003;91(441):86-90. https://doi.org/10.1111/j.1651-2227.2003.tb00653.x.
50. Vandenplas Y, Ludwig T, Bouritius H, et al. Randomised controlled trial demonstrates that fermented infant formula with short-chain galacto-oligosaccharides and long-chain fructo-oligosaccharides reduces the incidence of infantile colic. Acta Paediatr. 2017;106(7):1150-1158. https://doi.org/10.1111/apa.13844.
51. Rodriguez-Herrera A, Mulder K, Bouritius H, et al. Gastrointestinal tolerance, growth and safety of a partly fermented formula with specific prebiotics in healthy infants: a double-blind, randomized, controlled trial. Nutrients. 2019;11(7):1530. https://doi.org/10.3390/nu11071530.
52. Toca M, Sosa P, Aprigliano G, et al. Manejo de los trastornos funcionales digestivos más frecuentes en lactantes sanos. Arch Argent Pediatr. 2015;113(6):568-575. https://doi.org/10.5546/aap.2015.568.