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**SYNBIOTICS IN THE MANAGEMENT OF PEDIATRIC
GASTROINTESTINAL DISORDERS: POSITION PAPER OF THE
ESPGHAN SPECIAL INTEREST GROUP ON GUT MICROBIOTA
AND MODIFICATIONS**

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ABSTRACT

Background: Synbiotics are a mixture comprising of live microorganisms and substrate(s) selectively utilized by host microorganisms that confers a health benefit on the host. There is an increasing number of studies investigating their role in different diseases and disorders.

Aim: The purpose of this manuscript is to provide recommendations for the use of synbiotics in the management of pediatric gastrointestinal disorders. The recommendations are developed by the ESPGHAN Special Interest Group on Gut Microbiota and Modifications.

Methods: From existing literature databases, we searched and appraised all systematic reviews and/or meta-analyses, and subsequently published randomized controlled trials (RCTs) that compared the use of synbiotics, in all delivery vehicles and formulations, at any dose, compared to no synbiotics. Synbiotics which are part of infant formula were not assessed. The recommendations were formulated only if at least two RCTs that used a well-defined synbiotic were available.

Results: Based on the currently available evidence no recommendation can be formulated in favor or against the use of evaluated synbiotic combination in the treatment of acute gastroenteritis, prevention of necrotizing enterocolitis, *Helicobacter pylori* infection, inflammatory bowel disease, functional gastrointestinal disorders and allergy in infants and children.

Conclusion: There is a need for more, well designed RCTs on the role of synbiotics in gastrointestinal disorders with the same outcome measures to enable the inter-studies comparisons.

Key words: acute gastroenteritis, necrotizing enterocolitis, IBD, functional gastrointestinal disorders, H.pylori, prebiotic, probiotic

ACCEPTED

What is known:

- Synbiotics are a mixture comprising of live microorganisms and substrate(s) selectively utilized by host microorganisms that confers a health benefit on the host.
- Number of studies evaluating the effect of different synbiotics is increasing.

What is new:

- Due to lack of data no recommendation for the use of specific synbiotic combination in the prevention of treatment of gastrointestinal diseases can be formulated.
- There is a need for more well-designed studies that would use the same outcome measures for specific clinical indications to allow comparison between studies.

INTRODUCTION

The gastrointestinal (GI) microbiome has been investigated during the last several decades as a potential factor involved in the pathogenesis of many GI diseases. Therefore, targeting the gut microbiota with different strategies could have a possible positive effect in preventing or treating such conditions. These possibilities include different interventions with probiotics, prebiotics, synbiotics, and also with fecal material transplantations. So far, there is an increasing number of publications including guidelines and recommendations for probiotic use. On contrary, although number of studies investigating synbiotics are increasing, systematic reviews and recommendations on their use are still lacking.

Recently, the International Scientific Association for Probiotics and Prebiotics (ISAPP) defined synbiotics as a mixture comprising live microorganisms and substrate(s) selectively utilized by host microorganisms that confers a health benefit on the host (1). The Association also recognized two subsets of synbiotics, complementary and synergistic. According to ISAPP, a synergistic synbiotic is defined as a synbiotic in which the prebiotic substrate is designed to be selectively utilized by the co-administered microorganism(s). In contrast, a complementary synbiotic is a mixture composed of a probiotic strain combined with a prebiotic component, which is designed to target

autochthonous microorganisms (the resident microbiota). Regarding complementary synbiotic, the components must meet minimum criteria for the separate probiotic and prebiotic definitions.

There is an increasing number of studies evaluating efficacy of synbiotics, however, the conclusions are ambiguous. The purpose of this manuscript is to provide recommendations for the use of synbiotics for the management of pediatric GI disorders. These recommendations are developed by the ESPGHAN Special Interest Group (SIG) on Gut Microbiota & Modifications and its Working Group for Pre- and Probiotics (WG).

METHODS

For this review the following databases were searched: Cochrane Database of Systematic Reviews, DARE (Database of Abstracts of Reviews of Effects), CENTRAL (Cochrane Central Register of Controlled Trials), PubMed (National Library of Medicine, includes MEDLINE®) and EMBASE (Biomedical and pharmacological bibliographic database) for systematic reviews and/or meta-analyses, and subsequently published randomized controlled trials (RCTs) that compared the use of synbiotics, in all delivery vehicles and formulations, at any dose, compared to no synbiotic (i.e., placebo or no treatment or other interventions). Studies assessing the effect of synbiotics which is a part of infant formulas were not considered in this review. The reference lists from identified studies and key

review articles, including previously published meta-analyses were also searched. Search was limited to the end of December 2021. Only studies published in English were taken into consideration.

At least two reviewers independently assessed the eligibility of each potentially relevant trial. The data extracted included baseline characteristics, inclusion criteria, experimental and control treatments, setting, dose, outcomes of interest (with definitions if available), adverse events/side effects and funding.

One of the major criteria was that eligible studies describe the synbiotics in a way that microorganism is described by genus, species and strain designations. Consequently, if the strain designation (used by the depositor for the strain) was not given or the probiotic microorganism was not otherwise identifiable, the study was evaluated but not considered when a recommendation was made. In the same manner, study was evaluated but it was not taken into consideration for recommendation if prebiotic was not properly named, and exact dose mentioned.

It was decided to evaluate microbiota strain(s) and prebiotics as a part of synbiotic only. Brand or trade names were not considered, as the same brands may change composition and/or manufacturing practices over time and may have a different composition in different locations. Studies that

evaluated probiotics and prebiotics separately were not included in this review.

The genus of *Lactobacillus* has been recently reclassified into 25 genera, which include 23 novel genera (2). Therefore, throughout the manuscript, the new strain names were used.

The Working group (WG) followed the approach developed earlier (3) and elected to avoid recommendations on the use of the term “synbiotics” in general. Thus, the WG is reporting evidence and recommendations related to a specific synbiotic combination. The recommendations were formulated only if at least two well-designed RCTs that used a given synbiotic were available. If there was only one RCT, regardless of whether it showed a benefit, no recommendation was formulated.

It was planned to use the system developed by the Grading of Recommendations, Assessment Development, and Evaluations Working Group (4), and to categorize the certainty of evidence (quality of the evidence) and the strength of recommendations. Due to lack of evidence this was not performed.

The modified Delphi process was used to establish consensus on the statements. Level of agreement is presented next to the every statement/recommendation.

Table with all identified RCTs are available as supplemental data.

<http://links.lww.com/MPG/C878>

MANAGEMENT OF GASTROINTESTINAL CONDITIONS WITH SYNBIOTICS

INFANTS AND OLDER CHILDREN

Treatment of acute gastroenteritis

Two recent systematic reviews and meta-analyses were published in 2019 (5) and 2021 (6) evaluating probiotics as well as synbiotics. Yang et al. identified 5 RCTs but only 4 included synbiotics (7-10). Meta-analysis from 2021 identified only two quality studies that evaluated synbiotics (9, 10).

Vandenplas et al. (9) evaluated the role of mixture of probiotic bacteria [*Streptococcus (Str.) thermophilus*, 6.5×10^9 ; *Lactobacillus (L.) rhamnosus* and *Lactobacillus (L.) acidophilus* 6.5×10^9 ; *Bifidobacterium (B.) lactis* and *Bifidobacterium (B.) infantis* 6.5×10^9 /colony forming units (CFU)] and fructo-oligosaccharides (FOS) 20 mg compared to placebo and found significant decrease in the synbiotic group in the duration of the diarrhea (median duration was 3 days (IQ 25–75: 2–4 days) vs 4 days (IQ 25–75: 4–5 days); $p < 0.005$) (9). The authors evaluated the effect of the same preparation in another study including 46 children and showed significantly shorter diarrhea duration in the synbiotic group (3.04 ± 1.36 vs. 4.20 ± 1.34

days; $p=0.018$) (11). However, this study was underpowered as calculated sample size was not reached.

A study with another synbiotic combination of *Lacticaseibacillus (L.) paracasei* B21060, 2.5×10^9 CFU, plus 500 mg arabinogalactan, and 700 mg xilooligosaccharides twice daily had also a positive effect; a significantly higher resolution rate of diarrhea at 72 h was found in the synbiotic (67%) compared to the placebo group (40%, $p=0.005$) (10). Furthermore, children in the synbiotic group showed a significant reduction in the duration of diarrhea (90.5 h, 95% CI 78.1–102.9 vs. 109.8 h, 96.0–123.5, $p=0.040$), number of stool outputs (3.3, 95% CI 2.8– 3.8 vs. 2.4, 1.9–2.8, $p=0.005$) and increased stool consistency according to the score (1.3, 95% CI 0.9–1.6 vs. 0.6, 0.4–0.9, $p=0.002$) compared to the placebo group. Two studies performed in Turkey evaluated different compositions of synbiotics (7, 8). *B. lactis* B94 5×10^{10} CFU plus 900 mg inulin showed significant reduction in the duration of diarrhea in comparison to placebo (3.9 ± 1.2 days vs. 5.2 ± 1.3 days, respectively; $p < 0.001$) (8). Another study compared a synbiotic (*Lacticaseibacillus (L.) casei*, *L. rhamnosus*, *Lactiplantibacillus (L.) plantarum*, *B. lactis* at total dose of 4.5×10^9 CFU) and prebiotics such as fructose and galactooligosaccharides (GOS) and polydextrose at dose 1996.57 mg to 15 mg zinc supplementation and no treatment group (7). The duration of diarrhea was significantly reduced in the synbiotic and the zinc

groups compared to the control group (91.0±28.9 hours vs. 114.3±30.9 hours, $p<0.001$; 86.4±30.8 hours vs. 114.3±30.9 hours, $p<0.001$, respectively) with no significant difference between the synbiotic and zinc groups ($p>0.05$). Interestingly, at 72 and 96 hours, the rate of children with diarrhea was lower in the zinc group than in the synbiotic group ($p<0.05$ for both). This study did not mention strains used in synbiotic preparation.

In conclusion, only one synbiotic preparation (*Str. thermophilus*, 6.5×10^9 ; *L. rhamnosus* and *L. acidophilus* 6.5×10^9 ; *B. lactis* and *B. infantis* 6.5×10^9 CFU and 20 mg of FOS) was evaluated in two RCTs, however one of those studies (11) was significantly underpowered. Furthermore, difference between groups was approximately one day. Although clinical significance could be questionable, this corresponds to 25 to 30% reduction duration and costs of a very frequent disease of children. In conclusion, there were no two adequate and well-controlled studies at least, evaluating the same synbiotic preparation, so the effectiveness of an intervention could not be established, and no recommendation could be formulated.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation for the treatment of acute gastroenteritis (agreement: 100%).

***Helicobacter (H.) pylori* Infection**

A systematic review and meta-analysis from 2019 (12) identified 6 RCTs that evaluated effect of synbiotic treatment on *H. pylori* eradication rate. Three studies involved pediatric patients (13-15). Two Turkish studies compared the same synbiotic mixture (*B. lactis* B94 at dose 5×10^9 CFU/dose and inulin 900 mg) with triple therapy to triple therapy alone (13, 14) but yield contradictory results. Third study evaluated the effect of *Saccharomyces (S.) boulardii* and inulin (5 g) versus heat killed *L. acidophilus* 10^9 CFU to triple therapy in children colonized with *H. pylori* (15). Although synbiotics were able to clear colonization of *H. pylori* in 12% of children compared to *L. acidophilus* group (6.5%), the difference was not significant.

Two RCTs used the same synbiotic (*B. lactis* B94 at dose 5×10^9 CFU/dose and inulin 900 mg) but yield contradictory results so the effectiveness of an intervention could not be determined, and no recommendation can be formulated.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation for the treatment of *H. pylori* (agreement: 100%).

Inflammatory Bowel Diseases

A recent systematic review evaluated the effect of pro-, pre- and synbiotics in patient with inflammatory bowel diseases (IBD) in all age groups (16). Two studies were identified that included children; however, in the study by Yoshimatsu et al., although children were included, the mean age of participants was 44.8 ± 13.8 and 42.9 ± 15.9 years for synbiotic and placebo group, respectively (17).

The only synbiotic study that included only children and adolescents (up to the age of 21 years) with Crohn's disease in remission (18), compared synbiotic as an active preparation (LGG 10^{10} CFU and inulin 295 mg) to inulin alone (dose 355 mg). There was no placebo control. No significant difference between groups in all outcomes assessed was found. Another small pediatric pilot study included patients with ulcerative colitis (UC) in remission (19). In this study *B. longum* R0175 20×10^9 CFU/day with 15 g/day of inulin was compared to placebo. The study found a significant improvement of quality of life scores in the synbiotic group (phase I $p=0.014$ and phase II $p=0.034$). Severe symptoms occurred in 60% of the controls that experienced disease relapse, oppose to none in synbiotic group ($p=0.032$).

In conclusion, there were no two well designed RCTs at least, that used the same synbiotic preparation in the same population of IBD patients for a

specific health claim, preventing the formulation of a recommendation.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation in children with IBD (agreement: 100%).

Functional Gastrointestinal Disorders

Infantile colic

In a recent study examining the treatment of infantile colic in Germany and Poland, it was shown that almost all pediatricians are using either pro- or synbiotic preparations or pharmacological interventions to treat infantile colic (20). While treatment with probiotics has been extensively studied, scarce information is available on the use of synbiotics. The supplemental table lists all the available randomized controlled trials using synbiotics to treat infantile colic (21, 22). A study from Iran used 1 billion CFU of *L. casei*, *L. rhamnosus*, *Str. thermophilus*, *B. breve*, *L. acidophilus*, *B. infantis*, *L. bulgaricus* and FOS (dose not mentioned) compared to placebo in 45 infants, and found significant reduction in average daily crying time in the synbiotic group at day 7 and day 30, and higher symptom resolution at day 7, but not at day 30 compared to placebo (22). This study did not mention strains used in the synbiotic preparation. An open label randomized study used synbiotic (total of 10^9 CFU of: *L. acidophilus* LA-14, *L. casei* R0215; *L. paracasei* Lpc-3; *L. plantarum* Lp-115; *L. rhamnosus* GG,

Ligilactobacillus (L.) salivarius Ls-33, *B. lactis* B1-04, *B. bifidum* R0071, *B. longum* R0175 and 1.43 g of FOS) in comparison to simethicone (21). Significantly higher responder rates (effect $\geq 50\%$ reduction from baseline) of the multi-strain synbiotic compared to simethicone were found. No significant difference was found for the measure 'reduction of average number of crying phases per day in the last three weeks'.

In conclusion, there were no two well designed RCTs at least, that used same synbiotic preparation hampering a recommendation.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation in infants with infantile colic (agreement: 100%).

Functional abdominal pain disorders

A 2021 systematic review (23) on the different treatments of irritable bowel syndrome (IBS) identified only one trial that evaluated a synbiotic treatment (24). In this trial a synbiotic treatment [*B. lactis* B94 (5×10^9 CFU) and 900 mg inulin] was compared to a probiotic (5×10^9 CFU *B. lactis* B94) or a prebiotic (900 mg inulin) twice daily for 4 weeks, in 71 children with IBS (24). Synbiotic treatment improved belching and abdominal fullness ($p < 0.001$), bloating after meals ($p = 0.004$) and constipation ($p = 0.021$). The synbiotic group had a significantly higher percentage of patients with full recovery than the prebiotic group (39.1% vs. 12.5%, $p = 0.036$).

Administration of synbiotics and probiotics resulted in significant improvements in initial complaints when compared to prebiotics.

Two RCTs were identified that evaluated the role of synbiotics in functional abdominal pain (FAP) (25, 26). Both studies compared a synbiotic preparation to placebo. The study from 2015 (26) showed that the synbiotic (*B. coagulans* Unique IS-2, 1.5×10^8 spore plus FOS, 100 mg) had a higher rate of improvement at week 4 (60% vs. 39.5%, $p=0.044$), but there was no difference between the two groups at week 12 (64.4% vs. 53.4%, $p=0.204$). The more recent study (25) evaluated FOS in combination with seven types of bacteria with no strain determination (*L. casei*, *Str. thermophilus*, *L. acidophilus*, *L. bulgaricus*, *L. rhamnosus*, *B. breve* and *B. infantis*); the dose was also not mentioned. The response rate was higher with the synbiotic than with the placebo, after four weeks (53.1 vs 11.4%; $p<0.001$). Furthermore, the synbiotic had significant superiority to placebo in relieving the duration (4.56 ± 9.12 vs 12 ± 18.59 , min/day, $p=0.04$), frequency (0.31 ± 0.53 vs 1.17 ± 0.7 , episode/week, $p<0.001$) and intensity (2.38 ± 2.29 vs 5.49 ± 1.83 , $p<0.001$) of abdominal pain.

Only 3 RCTs were identified, all using different synbiotic preparations (in one (25) strain determination is missing) and involving a limited number of patients (47 to 88), therefore recommendation could not be formulated.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation in the treatment of functional abdominal pain and irritable bowel syndrome (agreement: 100%).

Functional constipation

Two systematic reviews that included synbiotic treatment in functional constipation, published in 2016 and in 2021 (27, 28), identified only two RCTs. The first was from Khodadad et al. that used a combination of probiotics with no strain specification (*L. casei*, *L. rhamnosus*, *S. thermophilus*, *B. breve*, *L. acidophilus*, *B. infantis* at the dose 1×10^9 CFU/1 sachet), and FOS (dose not mentioned) (29). This study investigated 3 interventions in 97 children: liquid paraffin oil and placebo, synbiotics and placebo, and liquid paraffin oil and synbiotics (29). Treatment success was similar in all groups without any significant difference between them ($p=0.6$), but less seepage of oil was seen in the synbiotic alone group ($p<0.001$). The second one from Baştürk used synbiotic containing *L. casei*, *L. rhamnosus*, *L. plantarum*, *B. lactis* (4×10^9 CFU) and prebiotics at a dose of 1996.57 mg (fiber, polydextrose, FOS and GOS) (30) and found that after 4 weeks of treatment, complete benefit was achieved in 48 (66.7%) and 21 (28.3%) patients in the synbiotic and placebo groups, respectively ($p \leq 0.001$).

Following these systematic reviews, one more RCT was published. It used *Limosilactobacillus (L.) reuteri DSM 17938* and 4 g of agave inulin and had 4 groups: probiotic alone (n=10), prebiotic alone (n=10), placebo (n=10), synbiotic (n=7). The frequency of normal stool tended to increase except the placebo group; only the prebiotic group showed a significant improvement ($p = 0.003$) (31).

In conclusion, only 3 RCTs were identified, all of them using different synbiotic product so no recommendation can be postulated.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation the treatment of constipation (agreement: 100%).

Food allergy prevention

Several recent systematic reviews and one guideline addressed the use of synbiotics in the prevention of allergic diseases (32-35).

Overall, 2 RCTs were identified in those reviews assessing effect of the synbiotic use on allergy prevention (including food allergy); one trial examined synbiotics (*B. bifidum* OLB6378 plus 0.5 g FOS) from birth to six months with or without skincare comparing to no intervention in infants at risk for atopic diseases (AD) (36). The study found that neither the emollient nor the synbiotic, showed any effect on reducing the development of AD and food allergy at 1 year of age. A study from Cabana et al (37)

evaluated the effect of a different synbiotic (*L. rhamnosus* GG (LGG) 10¹⁰ CFU and 225 mg of inulin) in children at risk and reported also no difference in eczema and asthma development. The comparator was only inulin so the effect of LGG, and not of the synbiotic was assessed.

Based on the available evidence the European Academy of Allergy and Clinical

Immunology (EAACI) provides no recommendation for or against synbiotics for pregnant and/or breastfeeding women and/or infants alone or in combination with other approaches to prevent IgE mediated food allergy in infants and young children (35).

Recommendation: No recommendation can be formulated on the use of specific synbiotic preparation in the prevention of food allergies (agreement: 92%).

PRETERM INFANTS AND NEONATES

Synbiotics in preterm infants

Recently, the combined use of prebiotics and probiotics, was recommended to optimize the effect of probiotics on premature infant's health based on an up-to-date network meta-analysis (38), although this was not done in a strain-specific manner.

Our systematic review of the literature found 5 studies using GOS, FOS, or long chain fructans (inulin) together with probiotics in preterm infants

(39-43).

All the studies used different synbiotics which prevents a meta-analysis of the data. Besides, all are underpowered, precluding any sound conclusions or recommendations on the effects of synbiotics on outcomes such as necrotizing enterocolitis (NEC). Small-sized studies increase the risk of making recommendations based on type 1 errors (false positive). Taking three of the most important related outcomes in preterm infants into consideration (late onset sepsis, stage ≥ 2 NEC and mortality), ESPGHAN previously estimated sample sizes per group of at least 247, 431, and 1465, respectively, to be required in RCTs (44). Recommendations based on small studies require downgrading of certainty due to imprecision.

There was only one RCT available which studied whether adding prebiotic (900 mg inulin) improves the effects of a probiotic (*B. lactis* B94, 5×10^9 CFUs) on proven sepsis, NEC or mortality (42). In this study, no beneficial effect on NEC was demonstrated, by adding a prebiotic to the probiotic. Furthermore, the number of included infants per group was low. The remaining RCTs (70-108 infants per intervention group) studied different multispecies synbiotics which all contained both Bifidobacteria and Lactobacilli together with FOS and/or GOS (39-41, 43). Unfortunately, in most studies the specific strain numbers were not reported in the original manuscripts hampering firm conclusions. The average gestational age in

several of the studies was above 30 weeks, so that complication rates are frequently much lower. Mortality was prevented by two of the products (39, 40), whereas two products (41) did not have an effect on mortality.

The first of these trials (39) studied a probiotic mixture (*L. rhamnosus* (4.1×10^8 CFU) + *L. casei* (8.2×10^8 CFU) + *L. plantarum* (4.1×10^8 CFU) + *Bifidobacterium (B.) animalis* (4.1×10^8 CFU) (NBL probiotic®) together with 383 mg of FOS and 100 mg of GOS. The study was not blinded, used alternate 2:1 randomization and did not have a placebo.

The second trial (40) studied a mixture of *L. acidophilus* (7.5×10^7 CFU/kg/d), *B. longum* (3.75×10^7 CFU/kg/d), *Bifidobacterium (B.) bifidum* (3.75×10^7 CFU/kg/d), and *Str. thermophilus* (3.75×10^7 CFU/kg/d) plus FOS 25mg/kg/d. The study was not blinded, randomization is unclear and there was no placebo.

The third trial (41) studied a mixture of *L. acidophilus* (1.4×10^9 CFU/d), *B. longum* (8×10^8 CFU/d), *L. rhamnosus* (8×10^8 CFU/d), *L. plantaris* (6×10^8 CFU/d), *L. casei* (6×10^8 CFU/d), *Lactobacillus (L.) bulgaricus* (6×10^8 CFU/d), *B. infantis* (6×10^8 CFU/d) and *B. breve* (6×10^8 CFU/d) plus FOS (200 mg/d). The study was not blinded and there was no use of placebo (41).

The final trial studied a mixture of *L. rhamnosus* (8.2×10^8 CFU/d), *L. plantarum* (4.1×10^8 CFU/d), *L. casei* (4.1×10^8 CFU/d), and *B. lactis* (4.1×10^8 CFU/d) plus FOS (383 mg/d), GOS (100 mg/d), BLF (2 mg/d),

and vitamins (25 mg/d of vitamin C, 8 mg/d of vitamin E, 0.5 mg/d of vitamin B1, B2 and B6). Again, there was no use of placebo. One of the investigators and the breast milk team were not blinded. There was no significant effect on NEC, late onset sepsis or mortality (43).

In conclusion, there are no firm data showing that the addition of a prebiotic improves the effect of a probiotic in preterm infants on NEC or mortality. Existing data on different multispecies synbiotics need to be reconfirmed by adequately powered and well-designed RCTs. Given the conflicting data on safety, efficacy of probiotic preparations in preterm infants and the potential for harm in a highly vulnerable population, current evidence does not support the administration of any of the considerably less studied synbiotics (45). The contribution of the prebiotic components of these products to the hypothesized effects of the probiotic strains are unknown.

Synbiotics in prevention of NEC in newborns with cyanotic congenital heart disease

Infants with congenital heart disease are at an increased risk of developing NEC (46). Potential preventive strategies such as the application of pro- and synbiotics have recently been reviewed (47). In 100 randomized newborns with cyanotic congenital heart disease (CCHD), synbiotic therapy (*B. lactis* B94, 5×10^9 CFU plus inulin 900 mg) prevented NEC (10% vs. 0%, $p=0.02$) and reduced mortality (28% vs 10%, $p = 0.04$) (48).

It is unknown whether inulin contributed to the observed effect. As already discussed in preterm infants the same research group has shown that adding inulin to *B. lactis* B94 had no effect (42). The etiology of NEC in infants with CCHD is certainly different from preterm infants and largely depends on the type of CCHD (46). Due to the large variability of CCHD, the different types were not evenly distributed between the placebo and the synbiotic group (e.g. all infants with hypoplastic left heart syndrome ended up in the placebo group) (48). Therefore, the authors regard their exciting data as preliminary and asked for further studies.

Recommendation: No recommendation can be formulated on the use of any specific synbiotic preparation for the prevention of NEC in preterm infants and newborns with cyanotic congenital heart disease (agreement: 100%).

ADVERSE EVENTS OR SIDE EFFECTS

From all included trials, six did not report on the side effects or adverse events (11, 14, 15, 39, 40, 48). Others, except for two studies (26, 31), reported that there were no side effects or/nor adverse events during the intervention. Garcia Contreras et al found no difference between synbiotic preparation (*L.reuteri* DSM 17938 and 4 g of agave inulin), probiotic alone, prebiotic alone and placebo in flatulence and abdominal distension that

were equally present in all 4 groups (31). While Saneian et al found that synbiotic group (*B. coagulans* Unique IS-2, 1.5×10^8 spore plus FOS, 100 mg) experienced more dry mouth than the placebo group but no difference in other possible side effects (26).

Discussion and conclusions

This review revealed insufficient evidence to provide recommendations in favor or against the use of synbiotics in pediatric GI diseases. The specific indications addressed here have been investigated by only limited number of studies, ranging from 2 RCTs per indication (infantile colic and IBD) to 5 RCTs (acute gastroenteritis). There are only two indications where two same synbiotic preparations were used. One is acute gastroenteritis where combination of strains (*Str. thermophilus*, 6.5×10^9 ; *L. rhamnosus* and *L. acidophilus* 6.5×10^9 ; *B. lactis* and *B. infantis* 6.5×10^9 CFU) with 20 mg of FOS that was tested in two RCTs (9, 11) performed by the same authors. However, one of the studies (11) was significantly underpowered, therefore no firm conclusion could be made. Another indication is the eradication of *H. pylori* where synbiotic combination (*B. lactis* B94 at dose 5×10^9 CFU/dose and inulin 900 mg) was used together with triple therapy and compared to triple therapy alone in two RCTs (13, 14). However, these studies yield contradictory results.

Furthermore, studies often included limited number of patients, had significant methodological biases (allocation concealment and/or blinding methods not reported, lack of comparator, bias in reporting), scarcely reported on the adverse events, and reported different outcomes.

Comparison of studies was further limited by the synbiotic preparation used, where dose effect was not assessed; only limited number of studies used the same synbiotic preparation for a specific clinical indication and, even more limited, used the same amount of live bacteria and prebiotic in the preparation. Also, very frequently the specific strain designation, in adjunction to the reported species, was not reported in the original manuscripts.

All of the above means that available studies would not fulfill newly stringent recommendations for RCTs evaluating the effect of synbiotics proposed by ISAAP (1). According to ISAAP, studies on a “synergistic synbiotic” that compare the synbiotic to the control can provide supportive evidence, but do not constitute direct evidence of the synergistic effect. Instead, a study including the combination, the substrate alone, the live microorganisms alone, and a control should be conducted. For the “complementary synbiotic” a two-arm parallel or crossover design was proposed.

In conclusion, due to the lack of data, no recommendation in favor or against the use of specific synbiotic combination in children with different gastrointestinal conditions could be formulated. There is a need for more, well designed RCTs that would follow the above suggested recommendations for study design and would use the same outcomes measures, making the inter-study comparisons possible.

DISCLAIMER

ESPGHAN is not responsible for the practices of physicians and provides guidelines and position papers as indicators of best practice only. Diagnosis and treatment are at the discretion of physicians

REFERENCES

1. 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.
2. Zheng J, Wittouck S, Salvetti E, et al. A taxonomic note on the genus *Lactobacillus*: Description of 23 novel genera, emended description of the genus *Lactobacillus* Beijerinck 1901, and union of *Lactobacillaceae* and *Leuconostocaceae*. *Int J Syst Evol Microbiol* 2020;70(4):2782-858.
3. Szajewska H, Guarino A, Hojsak I, et al. Use of probiotics for management of acute gastroenteritis: a position paper by the ESPGHAN Working Group for Probiotics and Prebiotics. *J Pediatr Gastroenterol Nutr* 2014;58(4):531-9.
4. Guyatt GH, Oxman AD, Vist GE, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. *BMJ* 2008;336(7650):924-6.
5. Yang B, Lu P, Li MX, et al. A meta-analysis of the effects of probiotics and synbiotics in children with acute diarrhea. *Medicine (Baltimore)* 2019;98(37):e16618.
6. Vassilopoulou L, Spyromitrou-Xioufi P, Ladomenou F. Effectiveness of probiotics and synbiotics in reducing duration of acute

infectious diarrhea in pediatric patients in developed countries: a systematic review and meta-analysis. *Eur J Pediatr* 2021;180(9):2907-20.

7. Yazar AS, Guven S, Dinleyici EC. Effects of zinc or synbiotic on the duration of diarrhea in children with acute infectious diarrhea. *Turk J Gastroenterol* 2016;27(6):537-40.

8. Islek A, Sayar E, Yilmaz A, et al. The role of *Bifidobacterium lactis* B94 plus inulin in the treatment of acute infectious diarrhea in children. *Turk J Gastroenterol* 2014;25(6):628-33.

9. Vandenplas Y, De Hert SG, group PR-s. Randomised clinical trial: the synbiotic food supplement Probiotal vs. placebo for acute gastroenteritis in children. *Aliment Pharmacol Ther* 2011;34(8):862-7.

10. Passariello A, Terrin G, Cecere G, et al. Randomised clinical trial: efficacy of a new synbiotic formulation containing *Lactobacillus paracasei* B21060 plus arabinogalactan and xilooligosaccharides in children with acute diarrhoea. *Aliment Pharmacol Ther* 2012;35(7):782-8.

11. Kluijfhout S, Trieu TV, Vandenplas Y. Efficacy of the Probiotic Probiotal Confirmed in Acute Gastroenteritis. *Pediatr Gastroenterol Hepatol Nutr* 2020;23(5):464-71.

12. Pourmasoumi M, Najafgholizadeh A, Hadi A, et al. The effect of synbiotics in improving *Helicobacter pylori* eradication: A systematic review and meta-analysis. *Complement Ther Med* 2019;43:36-43.

13. Ustundag GH, Altuntas H, Soysal YD, et al. The Effects of Synbiotic "Bifidobacterium lactis B94 plus Inulin" Addition on Standard Triple Therapy of Helicobacter pylori Eradication in Children. *Can J Gastroenterol Hepatol* 2017;2017:8130596.
14. N Sirvan B, K Usta M, U Kizilkan N, et al. Are Synbiotics added to the Standard Therapy to eradicate Helicobacter pylori in Children Beneficial? A Randomized Controlled Study. *Euroasian J Hepatogastroenterol* 2017;7(1):17-22.
15. Gotteland M, Poliak L, Cruchet S, et al. Effect of regular ingestion of Saccharomyces boulardii plus inulin or Lactobacillus acidophilus LB in children colonized by Helicobacter pylori. *Acta Paediatr* 2005;94(12):1747-1751.
16. Zhang XF, Guan XX, Tang YJ, et al. Clinical effects and gut microbiota changes of using probiotics, prebiotics or synbiotics in inflammatory bowel disease: a systematic review and meta-analysis. *Eur J Nutr* 2021;60(5):2855-2875.
17. Yoshimatsu Y, Yamada A, Furukawa R, et al. Effectiveness of probiotic therapy for the prevention of relapse in patients with inactive ulcerative colitis. *World J Gastroenterol* 2015;21(19):5985-5994.
18. Bousvaros A, Guandalini S, Baldassano RN, et al. A randomized, double-blind trial of Lactobacillus GG versus placebo in addition to

standard maintenance therapy for children with Crohn's disease. *Inflamm Bowel Dis* 2005;11(9):833-839.

19. Haskey N, Dahl WJ. Synbiotic therapy improves quality of life and reduces symptoms in paediatric ulcerative colitis. *Infant Child Adolesc Nutr*. 2009;1:88–93.

20. Sommermeyer H, Krauss H, Checinska-Maciejewska Z, et al. Infantile Colic-The Perspective of German and Polish Pediatricians in 2020. *Int J Environ Res Public Health* 2020;17(19).

21. Piatek J, Bernatek M, Krauss H, et al. Effects of a nine-strain bacterial synbiotic compared to simethicone in colicky babies - an open-label randomised study. *Benef Microbes* 2021;12(3):249-257.

22. Kianifar H, Ahanchian H, Grover Z, et al. Synbiotic in the management of infantile colic: a randomised controlled trial. *J Paediatr Child Health* 2014;50(10):801-805.

23. Levy EI, De Geyter C, Ouald Chaib A, et al. How to manage irritable bowel syndrome in children. *Acta Paediatr* 2022;111(1):24-34

24. Basturk A, Artan R, Yilmaz A. Efficacy of synbiotic, probiotic, and prebiotic treatments for irritable bowel syndrome in children: A randomized controlled trial. *Turk J Gastroenterol* 2016;27(5):439-443.

25. Gholizadeh A, Mehrabani S, Esmaili Dooki M, et al. Effect of a synbiotic on functional abdominal pain in childhood. *Caspian J Intern Med* 2021;12(2):194-199.
26. Saneian H, Pourmoghaddas Z, Roohafza H, et al. Synbiotic containing *Bacillus coagulans* and fructo-oligosaccharides for functional abdominal pain in children. *Gastroenterol Hepatol Bed Bench* 2015;8(1):56-65.
27. Koppen IJ, Benninga MA, Tabbers MM. Is There A Role for Pre-, Pro- and Synbiotics in the Treatment of Functional Constipation in Children? A Systematic Review. *J Pediatr Gastroenterol Nutr* 2016;63 Suppl 1:S27-35.
28. Wegh CAM, Baaleman DF, Tabbers MM, et al. Nonpharmacologic Treatment for Children with Functional Constipation: A Systematic Review and Meta-analysis. *J Pediatr* 2022; 240:136-149.e5.
29. Khodadad A, Sabbaghian M. Role of synbiotics in the treatment of childhood constipation: a double-blind randomized placebo controlled trial. *Iran J Pediatr* 2010;20(4):387-392.
30. Basturk A, Artan R, Atalay A, et al. Investigation of the efficacy of synbiotics in the treatment of functional constipation in children: a randomized double-blind placebo-controlled study. *Turk J Gastroenterol* 2017;28(5):388-393.

31. Garcia Contreras AA, Vasquez Garibay EM, Sanchez Ramirez CA, et al. Lactobacillus reuteri DSM 17938 and Agave Inulin in Children with Cerebral Palsy and Chronic Constipation: A Double-Blind Randomized Placebo Controlled Clinical Trial. *Nutrients* 2020;12(10).
32. Meirlaen L, Levy EI, Vandenplas Y. Prevention and Management with Pro-, Pre and Synbiotics in Children with Asthma and Allergic Rhinitis: A Narrative Review. *Nutrients* 2021;13(3).
33. Vandenplas Y, Meyer R, Chouraqui JP, et al. The role of milk feeds and other dietary supplementary interventions in preventing allergic disease in infants: Fact or fiction? *Clin Nutr* 2021;40(2):358-71.
34. de Silva D, Halken S, Singh C, et al. Preventing food allergy in infancy and childhood: Systematic review of randomised controlled trials. *Pediatr Allergy Immunol* 2020;31(7):813-826.
35. Halken S, Muraro A, de Silva D, et al. EAACI guideline: Preventing the development of food allergy in infants and young children (2020 update). *Pediatr Allergy Immunol* 2021;32(5):843-858.
36. Dissanayake E, Tani Y, Nagai K, et al. Skin Care and Synbiotics for Prevention of Atopic Dermatitis or Food Allergy in Newborn Infants: A 2 x 2 Factorial, Randomized, Non-Treatment Controlled Trial. *Int Arch Allergy Immunol* 2019;180(3):202-211.

37. Cabana MD, McKean M, Caughey AB, et al. Early Probiotic Supplementation for Eczema and Asthma Prevention: A Randomized Controlled Trial. *Pediatrics* 2017;140(3).
38. Chi C, Li C, Buys N, et al. Effects of Probiotics in Preterm Infants: A Network Meta-analysis. *Pediatrics* 2021;147(1).
39. Guney-Varal I, Koksall N, Ozkan H, et al. The effect of early administration of combined multi-strain and multi-species probiotics on gastrointestinal morbidities and mortality in preterm infants: A randomized controlled trial in a tertiary care unit. *Turk J Pediatr* 2017;59(1):13-19.
40. Sreenivasa B, Sunil Kumar P, Suresh Babu MT, et al. Role of synbiotics in the prevention of necrotizing enterocolitis in preterm neonates: a randomized controlled trial. *Int J Contemp Pediatr* 2015;2(2):127-130.
41. Nandhini LP, Biswal N, Adhisivam B, et al. Synbiotics for decreasing incidence of necrotizing enterocolitis among preterm neonates - a randomized controlled trial. *J Matern Fetal Neonatal Med* 2016;29(5):821-825.
42. Dilli D, Aydin B, Fettah ND, et al. The pro-pre-save study: effects of probiotics and prebiotics alone or combined on necrotizing enterocolitis in very low birth weight infants. *J Pediatr* 2015;166(3):545-51 e1.
43. Serce Pehlevan O, Benzer D, Gursoy T, et al. Synbiotics use for preventing sepsis and necrotizing enterocolitis in very low birth weight

neonates: a randomized controlled trial. *Clin Exp Pediatr* 2020;63(6):226-231.

44. van den Akker CHP, van Goudoever JB, Shamir R, et al. Probiotics and Preterm Infants: A Position Paper by the European Society for Paediatric Gastroenterology Hepatology and Nutrition Committee on Nutrition and the European Society for Paediatric Gastroenterology Hepatology and Nutrition Working Group for Probiotics and Prebiotics. *J Pediatr Gastroenterol Nutr* 2020;70(5):664-680.

45. Poindexter B, Committee On F, Newborn. Use of Probiotics in Preterm Infants. *Pediatrics* 2021;147(6).

46. McElhinney DB, Hedrick HL, Bush DM, et al. Necrotizing enterocolitis in neonates with congenital heart disease: risk factors and outcomes. *Pediatrics* 2000;106(5):1080-1087.

47. Kelleher ST, McMahon CJ, James A. Necrotizing Enterocolitis in Children with Congenital Heart Disease: A Literature Review. *Pediatr Cardiol* 2021;42(8):1688-1699.

48. Dilli D, Aydin B, Zenciroglu A, et al. Treatment outcomes of infants with cyanotic congenital heart disease treated with synbiotics. *Pediatrics* 2013;132(4):e932-938.

Supplemental data. Table with studies included in the systematic review.

ACCEPTED