Indian Academy of Pediatrics Consensus Guidelines for Probiotic Use in Childhood Diarrhea

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ABSTRACT

Justification: Several probiotic species and strains, single or combined, have been evaluated in childhood diarrheal disorders, and recommendations have ever been changing as newer trials are published. Therefore, there is a need to develop a guideline for Indian children describing the current role of probiotics in clinical practice.

Objectives: To develop a guideline for the use of probiotics in children with diarrhea.

Process: A national consultative group (NCG) was constituted by the Indian Academy of Pediatrics (IAP), consisting of subjects experts. Sub-topics were allotted to various experts as paired groups for detailed review. Members reviewed the international and Indian literature for existing guidelines, systematic reviews, meta-analyses and trials. Thereafter, two virtual structured meetings of the group were held on 2nd and 22nd August, 2020. The management guidelines were formulated by the group and circulated to the participants for comments. The final guidelines were approved by all experts, and adopted by the IAP executive board.

Recommendations: The NCG suggests *Lactobacillus GG* as a conditional recommendation with low-to-moderate level evidence or *Saccharomyces boulardii* as a conditional recommendation with very low-to-low level evidence as adjuvant therapy in acute diarrhea. The NCG also recommends the use of combination probiotics in neonatal necrotizing enterocolitis (NEC), as these reduce the risk of NEC stage II and above, late-onset sepsis, mortality and also time to achieve full feeds. The NCG does not recommend the use of any kind of probiotics in the therapy of acute dysentery, persistent diarrhea, *Clostridium difficile* diarrhea and chronic diarrheal conditions such as celiac disease, diarrhea-predominant irritable bowel syndrome and inflammatory bowel disease in children. Risk of antibiotic-associated diarrhea (AAD) is high with some antibiotics and most of these cases present as mild diarrhea. The NCG recommends probiotics only in special situations of AAD. *L. rhamnoses GG* or *S. boulardii* may be used for the prevention of AAD. VSL#3, a combination probiotic, may be used as an adjuvant in active pouchitis, prevention of recurrences and maintenance of remission in pouchitis.

Keywords: *Antibiotics-associated diarrhea, Celiac disease, Irritable bowel syndrome, Neonate, Necrotizing enterocolitis*.

Diarrhea, accounts for approximately 11% of childhood deaths worldwide [1-3]. The role of probiotics in different subtypes of diarrhea has been discussed in the literature and in various practice guidelines. Eubiosis refers to the harmony and balance of pathogenic bacteria and healthy bacteria in our gut. Dysbiosis is an imbalance of microbes that may result after an acute or chronic insult to the gut leading to perpetuation of the illness. However, the perpetuation may not necessarily be attributable to the pathogenic organisms. Unlike adults with a stable gut flora in a healthy state, the gut colonization is different in children, more so in infants and preterm neonates. The type of delivery,

gestational age, neonatal risk factors, antibiotic use and breast milk exposure are some of the factors determining the quality and quantity of gut microbiota in children. The optimal microbiota balance in various stages of childhood is unknown [4]. Probiotics, when administered in adequate amounts, confer beneficial effects by replacing the pathogenic bacteria with favorable ones, interfering with pathogen attachment, inactivating toxins, having antisecretory effects, reducing the loss of water and electrolytes from the gut, strengthening gut barrier integrity, releasing gut protective metabolites, and having loco-systemic immunomodulatory effects [5]. Despite the promising role, the question as to whether a certain probiotic is the one-stop primary answer or an adjuvant in the therapy of diarrheal diseases and the long-term effects of probiotics in permanent modification of gut microbiota and sustained immunomodulation are unanswered [6].

OBJECTIVE To develop a guideline for the use of probiotics in children with diarrhea.

PROCESS

A group of national experts, designated by the Indian Academy of Pediatrics (IAP), reviewed the current evidence related to the role of probiotics in childhood diarrhea and formulated practice guidelines. The initial meetings were held on August 2, 2020, and August 23, 2020, on a virtual platform. The key questions addressed included *i*) Do probiotics improve the overall outcome of the diarrheal state? *ii*) Which is/ are the probiotic(s) recommended? *iii*) What dose and duration of the probiotic(s) should be administered? *iv*) Any serious adverse effects of probiotic(s)? *v*) Any deviation from international guidelines? *vi*) Any special recommendations in the Indian context?

Pediatric literature of 15 years till August 2020 Medline indexed publications were deliberated upon, with controversies, if any, being debated upon by the experts. Discontinued probiotic strains were excluded from the discussion. The GRADE approach was used to assess the certainty (i.e., quality) of evidence (CoE) [7] and the strength of recommendations was graded as strong or conditional [8] (**Table I)** A consensus was agreed upon for each of the diarrhea subtypes. A writing group was designated for the manuscript. The draft was sent by email to all experts and their suggestions were incorporated in the final guidelines.

GUIDELINES

Acute Gastroenteritis

Before 2017, recommendations and practice guidelines came from North American and European societies, whereas, heavy caseload with more severity and high mortality were being handled in centers in Asia, Africa and Latin America. (**Box I)** [9-15]. Efficacy of probiotics in acute diarrhea is usually measured in terms of duration of diarrhea, stool volume and duration of hospitalization for caregivers and physicians. Modified Vesikari score (MVS) is used for research purposes to assess response [16]. Two recent double-blind randomized controlled trials (RCT) from North America changed the perception of probiotics in acute diarrhea [17,18]

Lactobacillus rhamanosus GG: The PECARN probiotic study [17] from United States of America, randomized 943 to *L. rhamanosus GG (LGG)* and placebo groups preschool children. The trial used a statistical enrichment design to compensate and represent for those who were likely to benefit (such as longer duration of symptoms) in their cohort. The study failed to show any superiority of the probiotic over placebo in overall MVS or secondary individual components (stool consistency, stool frequency, vomiting frequency, time to vomiting abatement, intravenous fluid requirement, rate of hospital admission, follow-up visits, missed daycare, missed employment hours of caregivers, and household transmission) [17]. A systematic review and meta-analysis on LGG also showed no significant reduction in stool volume (18 RCT, *n*=4208), reduction in diarrhea duration by -0.85 days (95%CI: -1.15 to -0.56) (15 RCT, *n*=3820), daily dose $>10^{10}$ CFU fared better than $<10^{10}$ CFU (15 RCT, *n*=2007), better results in Europe than non-Europe (15 RCT, *n*=2007) and reduction in hospitalization stay by -1.22 days (95%CI: -2.33 to -0.1) (5 RCT, *n*=990) [9]. Another meta-analysis (with a different study inclusion) concluded that LGG had a reduction in diarrhea duration by 24 hours, higher dose $>10^{10}$ CFU fared better than $<10^{10}$ CFU, showed similar performance in Asians and Europeans, was most effective if introduced <72 hours of diarrhea, and had better results in rotaviral diarrhea subset [13]. Based on this information, routine testing for rotaviral diarrhea may not be required as probiotics would have already been instituted before the rotavirus test results are available.

L. rhamanosus R0011 and L. helveticus R0052: The PERC-PROGUT trial randomized 886 preschool children from Canada, with mild diarrhea, to receive *L. rhamnosus* R0011 and *L. helveticus* R0052 combination or placebo. The results were similar to the PECARN study. Probiotic was not superior in terms of MVS or secondary outcomes as compared to placebo. Development of moderate-severe gastroenteritis was 26% in probiotic group vs 25% in placebo [OR 1.06; 95% CI 0.77 to 1.46; *P*=0.72] within 14 days. The two trials (PERC-PROGUT and PECARN) were criticized for inclusion of children with mild severity, those already immunized for rotavirus, stool testing for viral etiology, concurrent antibiotic exposure, questionable viability of probiotics in liquid form, and timing of probiotic administration (after 48 hour of diarrhea onset) [19-22]. The PERC-PROGUT also did not find any significant differences between probiotic vs placebo in viral shedding response for adenovirus, norovirus and rotavirus. No differences were seen in the bacterial and parasitic diarrheal subgroups [23].

Saccharomyces boulardii: Studies with *S. boulardii* had major limitations due to poor designs and limited data outcomes. The analysis was inconclusive regarding stool volume. Reductions were seen in diarrhea duration by mean -1.06 (95% CI -1.32 to -0.79) days (23 RCT, *n*=3450), hospitalization by mean difference -0.85 (95% CI -1.35 to -0.34 days (8 RCT, *n*=999), and reduced diarrhea episodes in the following days (day 2-7) of implementation [10].

L. reuteri DSM 17938: Two systematic reviews were available on *L. reuteri* DSM 17938. The first review in 2016 could not make any logical interpretations due to the extreme heterogeneity in studies

[24]. The second analysis in 2019 included only four studies with 347 patients. The reduction in stool volume was inconclusive. The mean difference in reduction in diarrhea duration was by -0.87 (95% CI -1.43 to -0.31) days and in hospitalization by -0.54 (95% CI -1.09 to 0.0) days [25]. Another RCT also concluded the lack of efficacy of the above probiotics in acute gastroenteritis [26].

L. acidophilus: Chau, et al. [27] randomized *L.acidophilus* vs placebo in 150 school children in Vietnam. The probability of still having diarrhea till 216 hours after implementation was similar in both groups [27].

Bacillus clausii: There is a paucity of well-designed studies regarding *B.clausii* in children. A meta-analysis failed to provide any practical conclusions [28].

Current International recommendations

The European Society of Pediatric Gastroenterology Hepatology and Nutrition (ESPGHAN) made updated recommendations in 2020 [11,12]. They provide weak recommendation for the use of *LGG, S. boulardii* and *L.reuteri* with low-quality of evidence. Use of *B. clausii*, *L. helveticus, or a combination of probiotics is not recommended* in acute diarrhea [11,12]. Based on the moderate quality of evidence, the American Gastroenterology Association (AGA) guidelines (2020) [8,15] do not recommend any probiotic use in children with acute diarrhea (conditional recommendation with moderate quality of evidence).

Indian experience

Unlike their first RCT [29], Basu, et al. [30], in their second RCT, demonstrated a significant reduction in duration of diarrhea, hospital stay and consistency of third-day stools with LGG $>10^{10}$ CFU/day [30]. Another RCT [31] in infantile diarrhea did not demonstrate any benefit of LGG. An RCT [32] in children aged 0.5-5 years showed reduction in diarrhea duration and improvement in consistency by 1 score in the LGG group as compared to controls, with both rotavirus positive and negative groups. In a double-blind RCT by Das, et al. [33], *S. boulardii* had a marginally lesser duration of diarrhea and hospitalization as compared to controls, among children below 5 years. Another double-blind RCT in the same age group showed marginal benefit in the amount of oral rehydration solution (ORS) consumed, faster rehydration and earlier semi-formed stools with *S. boulardii* than placebo [34]. There is a paucity of well-designed studies from India. The summary of NCG recommendations for acute diarrhea is shown in **Table II.**

Necrotizing enterocolitis

Necrotizing enterocolitis **(NEC)** is primarily a disease of premature infants, but may also be seen rarely in 10% of term and near-term babies. The etiology of NEC is multifactorial and there are increasing concerns of abnormal gut microbiota playing a key role in its pathogenesis. The role of probiotics has been evaluated for the prevention and progression to stages 2-3 NEC, late onset sepsis, mortality, and time required until full enteral feeding [35]. The statistical benchmarks required to assess impact of any probiotic are: *i*) reduction of mortality prevalence from 7.5% to 5% (number required for analysis 1465); *ii*) reduction of NEC ≥2 prevalence from 10% to 5% (number required

for analysis 431); and *iii*) reduction of prevalence of late-onset sepsis from 25% to 15% (number required for analysis 247). Prerequisites before administration of probiotics in preterm babies include *i*) certificate of analysis: purity, viability, antibiotic susceptibility *ii*) available microbiological laboratory to detect fungemia; *iii*) inability of any of the probiotics to produce D-lactate; *iv*) absence of antibiotic resistance capability; and *v*) informed consent from parents [35].

International experience

Under standard safety guidelines, ESPGHAN conditionally recommends [35.] *L. rhamnosus GG ATCC 53013* or a combination probiotic *(B. infantis Bb-02, Bifidobacterium lactis Bb-12, and Streptococcus thermophiles TH-4)* for prevention of NEC stage ≥2. No recommendations were made by ESPGHAN regarding probiotics for the reduction in mortality or prevention of late-onset sepsis or on the duration of therapy. In some studies, probiotics were administered for two weeks. However, the majority of the studies administered probiotics for 4-6 weeks or until discharge. No recommendations were made for *L. reuteri* DSM 17938 and *L. acidophilus* NCDO 1748 (ATCC 4356, LA37, or NCIMB 30316) containing regimen as they are partially D-lactate-producing strains, for which there was insufficient safety data available in preterm infants. *S. boulardii* has the potential to cause fungemia in those neonates with central venous catheters, who are critically ill, or in immunocompromised patients [35].

A strain-specific systematic review and network analysis found that the time to enteral feeding was significantly reduced with three different probiotic interventions: *L. reuteri ATCC 55730 or DSM 17938* (3 studies, *n*=626 infants) by 3.3 (range 0.62-6.4) days; for the combination of *B. bifidum, B. infantis, B. longum* and *L. acidophilus* (2 studies, *n*=247 infants) by 4.7 (range 0.70-8.6) days, and for the combination of *B. longum BB536* and *L. rhamnosus GG* (based on 1 study with 94 infants) by 10 (3.6-16) days [36]. A meta-analysis by Bi, et al. [37] (34 studies, *n*= 9161) showed an overall advantage of probiotics in preventing the incidence of necrotizing enterocolitis, gut-associated sepsis, and decrease mortality in preterm infants. AGA guidelines conditionally recommend (moderate-high quality of evidence) the use of a combination of *Lactobacillus* spp. and *Bifidobacterium* spp**.** (*L. rhamnosus* ATCC 53103 and *B. longum* subsp *infantis*; or *L. casei* and *B. breve*; or *L. rhamnosus*, *L. acidophilus*, *L. casei*, *B. longum* subsp *infantis*, *B. bifidum*, and *B. longum* subsp *longum*; or *L. acidophilus* and *B. longum* subsp *infantis*; or *L. acidophilus* and *B. bifidum*; or *L. rhamnosus* ATCC 53103 and *B. longum* Reuter ATCC BAA-999;or *L.acidophilus*, *B.bifidum*, *B.animalis* subsp *lactis*,and *B.longum* subsp *longum*), or *B. animalis* subsp *lactis* (including DSM 15954), or *L. reuteri* (DSM 17938 or ATCC 55730), or *L. rhamnosus* (ATCC 53103 or ATC A07FA or LCR 35) over no and other probiotics in low birthweight preterm babies. Best outcomes have been observed in babies <32 weeks of gestation and weighing <1500 g [38].

Indian experience

Most of the studies have used multi-strain probiotics [39-41] and a few used single-strain probiotics [42]. Probiotic supplementation was continued till discharge, till reaching full feeds, or for a pre-specified duration between 7 and 21 days. Most studies focused on preterm infants <34 weeks while some studies also included infants between 34 and 37 weeks of gestation [39-42]. The meta-analysis by Balasubramanium, et al. [43] showed reduced risk of NEC ≥stage II [RR: (95%CI) 0.36: (0.20, 0.66); *P* =0.0009; 9 RCTs] , late-onset sepsis [RR (95% CI) 0.56: (0.45, 0.71); *P* <0.00001;7 RCT] and mortality [RR: (95% CI) 0.62: (0.41, 0.95), *P*= 0.03; 8 RCTs] in the probiotic group. Probiotics also reduced the time to full feeds [mean difference (MD): −4.09 d (95% CI: −4.52, −3.65); *P*<0.00001; 5 RCTs] and duration of hospital stay [fixed effects model (FEM): MD: −2.00 day (95% CI: −2.46, −1.53); *P*<0.00001;6 RCTs].

Since there is low to moderate level of certainty about the effects of probiotic supplements on the risk of NEC and associated morbidity and mortality (**Table II**), large, high-quality studies are needed. Studies comprising one probiotic with another, single strain versus combination, route of administration (powder/liquid), optimal dose, time of initiation, duration of therapy and quality of enteral feeding (mother/donor/formula milk) are required. Subgroup analyses on <28 weeks gestational age babies, <1000g birthweight babies and term safety issues (immunity, endocrine, metabolic, behavioral) are needed. Demography-based issues (geography, gender, ethnicity, peripartum culture practices) are also a concern.

Acute Dysentery

Dysentery has a 100-fold higher incidence in Asians as compared to developed countries, mostly among children aged 1–4 years living in low and middle-income countries. The Global Enteric Multicenter Study (GEMS) [32], upon analysis of samples with quantitative PCR, found Shigella to be the major pathogen in dysentery (attributable fraction 63.8%), and the second most common pathogen in watery diarrhea (attributable fraction 12.9%) [33]. There is a poor rationale and limited data for the use of probiotics in dysentery in children. Hence the group recommends against the use of any kind of probiotic in acute dysentery (non-*Clostridiodes difficile* causes) (**Table II**).

Antibiotic-associated Diarrhea

Antibiotic-associated diarrhea (AAD) is defined as diarrhea that occurs on exposure to prolonged antibiotics, provided other etiologies have been excluded. The risk of AAD is higher with antibiotics like aminopenicillins without/with clavulanate, cephalosporins, clindamycin, and anti-anerobic agents, whether given orally or through intravenous route. Most AAD presents as mild diarrhea. Rarely they may cause fulminant pseudomembranous colitis where usually, no pathogen is identified. *C. difficile* is the most common incriminating agent in those with underlying diseases such as inflammatory bowel diseases, cystic fibrosis, and neoplasms.

International experience

The ESPGHAN 2016 [46] assessed report 21 RCTs (*n*=3255 children), and the pooled results showed that probiotics as compared to placebo or no intervention reduced the risk of AAD by 52% (21.2% vs 9.1%, respectively; RR 0.48, 95% CI 0.37–0.61). Only two probiotics *(LGG and S. boulardii)* could be assessed with certainty. Compared with placebo, the administration of these probiotics also reduced the risk of *C difficile*-associated diarrhea (4 RCTs, *n* = 938, RR 0.34, 95% CI 0.15–0.76).

The Cochrane systematic review (33 studies; *n*=6352) studied *Bacillus spp., Bifidobacterium spp., Clostridium butyricum*, *Lactobacilli spp.*, *Lactococcus spp., Leuconostoc cremoris*, *Saccharomyces spp.,* or *Streptococcus spp.,* alone or in combination, and concluded was that probiotics reduce the incidence of AAD (NNTB 9, 95% CI 7 to 13) with high doses (≥ 5 billion CFUs/day) being preferable. Thus, they recommended *LGG* or *S. boulardii*; however, with a need for a large well-designed multi-centred randomized trial. They could not conclude the efficacy and safety of the other probiotic agents [47].

In children on antibiotic treatment, AGA guidelines suggest the use of *S. boulardii*; or the two-strain combination of *L. acidophilus CL1285* and *L. casei LBC80R*; or the three-strain combination of *L. acidophilus, L. delbrueckii* subsp *bulgaricus*, and *B. bifidum*; or the four-strain combination of *above three with S. salivarius subsp thermophilus* for prevention of *C. difficile* infection (conditional recommendation, low quality of evidence). AGA also recommends that probiotics can be reasonably avoided in patients having severe illnesses, where the cost-benefit ratio is poor and also in those with a very small risk of *C. difficile* development (particularly in the outpatient settings) [8,15].

C. *difficile*-associated diarrhea

The Cochrane review (31 trials, *n*=8672 adults and children) suggested that probiotics reduce the risk of CDAD by 60%. Trials with a baseline CDAD risk of 0-2% and 3.5% did not show any difference in risk but trials enrolling participants with a baseline risk of >5% for developing CDAD demonstrated a large (70%) risk reduction. Regarding the detection of *C. difficile* in the stools, pooled complete case results from 15 trials (1214 participants) did not show a reduction in infection rates. No major adverse events were reported. The review concluded that probiotics are effective for preventing CDAD (NNTB = 42 patients, 95% CI 32 to 58), especially in those with CDAD baseline risk >5% (NNTB = 12; moderate certainty evidence). Of all the probiotic agents, *S. boulardii* CNCM 1745 >5 billion CFU/day was found useful [48]. AGA does not recommend the use of probiotics in CDAD due to a considerable knowledge gap [8,15].

Indian scenario

There is a paucity of data in India regarding AAD in children. A study from Bhopal [49], showed that 64% under-5 children with a diagnosis of acute gastroenteritis from outpatient services received antimicrobials [49]. Another study [50] from a tertiary referral hospital in Eastern India showed that

80% of 265 children received 535 antimicrobial agents, of which 85% were based on empirical decisions. Of these 2.2% developed loose stools as adverse events [50]. The cost vs benefit of preventive use of probiotics in AAD cannot be entirely gauged. *C. difficile* is present in stool culture of 15% of pediatric acute diarrhea, of which only 2.9% are toxigenic [50]. It is toxigenic *C. difficile* that assumes importance in pathogenesis of AAD.

Persistent Diarrhea

There is limited data on the use of probiotics for persistent diarrhea in children [4,8]. There is a suggestion that *S. boulardii* and LGG decrease the duration and frequency of loose stools along with reduction of hospital stay in children with persistent diarrhea [51,52]. In a Cochrane meta-analysis (4 trials; n = 464) [53], it was found that probiotics reduced the duration of persistent diarrhea (MD (95% CI) 4.02 (4.61 to 3.43 days); 2 trials, n=324). Stool frequency was reduced with probiotics in two trials. One trial reported a shorter hospital stay [53]. In clinical practice, it is noted that most children with acute diarrhea may have already received probiotics before they enter the persistent phase. NCG does not recommend the use of probiotics in the management of persistent diarrhea **(Table II)**.

Chronic diarrhea With Underlying Gastrointestinal Conditions

The role of probiotics in children with such conditions is mostly extrapolated from adult studies.

International experience

AGA does not recommend any probiotic in irritable bowel syndrome-diarrhea (IBS-D), induction or remission of Crohn's disease, induction or remission of ulcerative colitis (UC), due to lack of adequately powered studies [8,15]. ECCO-ESPGHAN guidelines [54] do not recommend the use of probiotics in the management of Crohn's disease. Probiotic agents (e.g., VSL#3, *E. coli* Nissle 917) can be considered in mild UC as adjuvant therapy or in those intolerant to mesalamine) [54,55]. In celiac disease, multiple attempts have been made to use probiotics as an alternative management strategy but none have been able to significantly alter the epithelial recovery, intestinal permeability and microbial signature. Lifelong gluten-free diet still continues to be the only proven therapy in celiac disease [56,57]. NCG does not recommend the use of probiotics in chronic diarrhea with underlying gastrointestinal conditions (**Table II**).

Pouchitis

Pouchitis is a problem in those who have undergone colectomy with an ileal pouch-anal anastomosis (IPAA), in various disease conditions such as ulcerative colitis, IBD-undifferentiated or polyposis coli. Probiotics may be considered in active pouchitis, primary prevention of pouchitis or recurrence of pouchitis. There is some evidence regarding the efficacy of VSL#3 [a mixture of eight probiotic strains: *Streptococcus thermophilus, Bifidobacterium breve, B. longum, B. infantis, Lactobacillus acidophilus, L. plantarum, L. paracasei, L. delbrueckii (subsp. bulgaricus*)] in preventing the initial episode and further relapses of pouchitis. The Asia-Pacific [58] guideline states that in pouchitis,

VSL#3 may be considered based on the evaluation of individual cases. Due to the lack of literature in children, doses of VSL#3 as per a previous pediatric study in IBD [59] may be adopted for its use in this disease state also. Doses of 450 billion bacteria/day in 4-6 years of age (weighing 17-23 kg), 900 billion in 7-11 years of age (weighing 24-33 kg), 1350 billion in 11-14 years of age (weighing 34-53 kg) and 1800 billion in 15-17 years of age (weighing 54-66 kg) [59]. In adults and children with pouchitis, AGA recommends a 8-strain combination of [*L. paracasei* DSM 24733, *L. plantarum* DSM 24733, *L. acidophilus* DSM 24733, *L delbrueckii subsp bulgaricus* DSM 24733, *B. longum* DSM 24733, *B. breve*, DSM 24733, *B. longum* subsp *infantis* DSM 24733, and *S. salivarius* subsp *thermophilus* DSM 24731] over no or other probiotics (conditional recommendation, low quality of evidence) [8,15]. Cochrane meta-analysis concluded that it was uncertain if probiotics had an advantage over placebo in all the above three settings [60]. NCG recommendations for pouchitis are given in **Table II.**

Probiotics Currently Available in India

A recent publication [61] has comprehensively summarized the composition and laboratory correlation of the commercially available probiotics in India till recently. Significant differences were noted in some of the brand preparations and the next-generation sequencing results with regard to the probiotic organism. In some of the probiotics, new strains were found on cultures that were not a part of the original label [61]. They also highlighted the differences in the label concentrations and the in vitro viable cell count on culture indirectly signifying the actual bioavailability [61]. In combination probiotics such as VSL#3, there are other issues regarding the recommended concentration and marketed concentration. Hence the prescription of the probiotic should be carefully written keeping in mind the rationale, actual species and strain, marketed concentration, mode of delivery (sachet, capsule, syrup or vial), projected bioavailability and shelf life. Among the recommended single-strain probiotics in this guideline, LGG and *S. boulardii* are marketed in India. With regards to NEC in preterms, an exclusive combination of *Lactobacillus* spp and *Bifidobacterium* spp in the recommended concentrations are not available yet in India. It is cautioned that some of the marketed combination probiotics contain *S. boulardii*. VSL#3 is available in India at lower concentrations than the international standards. In view of the above lacunae, pharmaceutical manufacturers in India should take appropriate steps in accordance with the scientific guidelines.

CONCLUSIONS

Probiotics in India should be used judiciously, after obtaining complete information regarding the strains present, CFU, shelf life and storage requirements. The decision to use a probiotic should be in line with scientific evidence keeping in mind the cost-benefit ratio.

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experience; DS: literature review, intellectual inputs and editing of manuscript; GVB, BJP: conceptualization and intellectual inputs. All authors approved the final version of manuscript.

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Table I Quality of Evidence and Strength of Recommendations Used for these Guidelines [1,2]

Category	***Inference***
Quality of evidence	
High quality	Further research is unlikely to change our confidence in the estimate of effect
Moderate quality	Further research is likely to have an important impact on our confidence in the estimate of effect and may change the estimate
Low quality	Further research is very likely to have an important impact on our confidence in the estimate of effect and is likely to change the estimate
Very low quality	Any estimate of effect is uncertain
Strength of recommendation	
Strong	Most individuals should receive the recommended course of action. Formal decision aids are not likely to be needed to help individuals make decisions consistent with their values and preferences
Conditional	Different choices will be appropriate for different patients. Decision aids may be useful in helping individuals in making decisions consistent with their values and preferences. Clinicians should expect to spend more time with patients when working towards a decision.

Table II Summary of National Consultative Group Recommendations in Various Disease Conditions of Diarrhea

Condition/Recommendations	*Strength of recommendation*	*Quality of evidence*
Acute Diarrhea *Lactobacillus rhamanosus GG* $\geq 10^{10}$ CFU/d, typically 5-7 d or *Saccharomyces boulardii* 250 to 750 mg/d, for 5-7 d Above probiotics can be used as adjuvant therapy in acute diarrhea. The marginal cost-benefit advantage must be carefully considered before prescribing them in impoverished conditions in India. No recommendations can be made for *Lactobacillus reuteri* DSM 17938, *Lactobacillus acidophilus* and *Bacillus clausii.* No recommendations can be made for combination probiotics	Conditional Conditional	Low to moderate level Very low- to-low level
Prevention of NEC Combination of *Lactobacillus* spp. 1 x10^9 to 6 x10^9 CFU and *Bifidobacterium spp.* 3.0 to 3.5 x 10^8 CFU for 4 to 6 weeks or up until discharge or L. rhamnosus GG ATCC 53013 1 x10^9 to 6 x10^9 CFU for 4 to 6 weeks or up until discharge Probiotics seem to be useful in reducing risk of NEC ≥ stage II, with reduction of late onset sepsis, mortality and also reduce the time to full feeds (Indian data). There is moderate-high quality of evidence in preterm (less than 37 weeks gestational age), low-birth weight using probiotic combination of *Lactobacillus* spp. and *Bifidobacterium* spp. Conditionally recommends L. rhamnosus GG ATCC 53013 as a single probiotic in situations of non-availability of suitable combination of Lactobacillus spp. and Bifidobacterium spp. Probiotics are	Conditional Conditional	Moderate-high In preterm (less than 37 weeks gestational age) and low- birth weight babies Low for prevention of NEC stage ≥2

more effective in human milk fed babies.		
Antibiotic associated diarrhea (AAD) NCG does not recommend routine use of probiotics for the prevention of AAD in immunocompetent children being treated with antibiotics. However probiotics may be prescribed for prevention of AAD in certain special situations where the risk of AAD is very high or if AAD has the propensity to result in prolonged morbidity or mortality. In children with prior history of AAD (two or more separate episodes), *either Lactobacillus rhamnoses GG or S. boulardii* may be used In children where there is an expected high risk of morbidity / mortality due to AAD (e.g., hospitalized sick children, underlying chronic diseases), *Lactobacillus rhamnoses GG* only should be used. Whenever used, the probiotic should be prescribed for the entire duration of antibiotic therapy. *C. difficile* diarrhea is not common in Indian children (personal experience of NCG), and does not recommend probiotics for the specific purpose of preventing CDAD.	Conditional Conditional	Low to moderate Low to moderate
Pouchitis In active pouchitis probiotics may be used as an adjuvant therapy Probiotics seem to be beneficial for prevention of recurrences and also for maintenance of remission in pouchitis Probiotic used may be either VSL#3 or a mix of strains as suggested by ACG guideline. Doses in children are given in the text under pouchitis.	Conditional Conditional	Low quality Moderate
NCG: A national consultative group; AAG: American Gastroenterological Association		

No recommendations for acute dysentery, persistent diarrhea, Celiac disease, Diarrhea predominant irritable bowel syndrome, Inflammatory bowel disease, as there is a lack of published evidence.

Box I Reasons for Heterogeneity of Various Probiotic Studies [9-15]

Patient factors
Setting: Low income vs high income countries (Human Development Index)
Severity of diarrhea
Immunization status
Nutrition status
Viral vs. bacterial vs unidentified etiology
Intervention factors
Day of intervention in acute diarrhea
Single vs combination probiotic
Strain and dose of bacteria
Duration of therapy
Type of trial (randomized vs. open label)
Methodology issues