The Efficacy of The Administration of BifidobacteriumLactis For Treatment of Infantile Colic: A Double-Blind, Randomized, Placebo-Controlled Clinical Trial

Mohammad Ali Pourmirzaiee
Isfahan University of Medical Sciences
Fatemeh Famouri
Isfahan University of Medical Sciences
Akram Kahid
Isfahan University of Medical Sciences
Silva Hovsepian (silvahovsepsecret@gmail.com)
Isfahan University of Medical Sciences
Roya Kelishadi
Isfahan University of Medical Sciences

Research Article

Keywords: Infantile colic, Bifidobacterium Lactis, treatment

Posted Date: August 10th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-730782/v1

Abstract

The aim of this trial was to evaluate the efficacy of Bifidobacterium Lactis administration on infantile colic (IC). In this double blind randomized, placebo-controlled clinical trial, infants with IC diagnosis, exclusive breastfeeding, gestational age more than 37 weeks and birth weight more than 2500 gram were included. The selected infants randomly allocated in the two groups of BBcare group, treated with Bifidobacterium animalis subsp. lactis and the control group treated with placebo, 5 drops per day, for 6 weeks. The mean of crying, vomiting episodes and defecation number at baseline and during follow ups (40 and 60 days after intervention) were compared between the two studied groups. In this study from initially enrolled neonates, 40 and 38 neonates in BB-12 and placebo groups completed the study. In BB-12 group number of defecation, crying and vomiting episodes decreased significantly during intervention till 60 days after probiotic administration($P<0.05$).In the placebo group there was significant decrease for crying and vomiting episodes between baseline and 60 days after intervention($P<0.05$). Between group analyses indicated that there was significant difference between groups regarding mean of crying and vomiting episodes and number of defecation ,60 days after intervention($P<0.05$).

Conclusion: Findings of our study offer compelling signals for the effectiveness of Bifidobacterium animalis subsp. lactis BB-12 in the management of some intestinal problems. These findings could be supportive evidences for the important role of gut microbiota as goal of intervention to improvement in bowel movement and comfortable defecation in IC.

What Is Known?

- The role of probiotics in the treatment of IC has been investigated in several studies.
- Results of a recent systematic review and meta-analysis of clinical trials have demonstrated that probiotics could reduce the crying episodes in infants with IC.

What is new?

- Bifidobacterium Lactis is effective in decreasing the crying and vomiting episodes and number of defecation in infants with IC.
- Our findings could be supportive evidences for the important role of gut microbiota as goal of intervention to improvement in bowel movement and comfortable defecation in IC.

Introduction

Near to 25% of infants suffer by a functional gut disturbance, which named infantile colic (IC). This benign disorder presents between 1–4 months of life and considers as an origin of major distress for the infant, parents and their family [1]. There is no absolute association between IC and kind of feeding, socioeconomic standing and gestational age and occurs equal between both sexes.

Since now, the etiology and pathogenesis of IC remain unknown and is supposed to be multifactorial, but, recent researches proposes that alterations of gastrointestinal microorganisms can donate to the advance of this condition[2].Based on available data, there are reports about a lower variety and constancy of gastrointestinal microbiota in neonates with IC, during the first 2 weeks of life[3].Given the role of dysbiosis in the pathogenesis of IC, some studies have evaluated the effectiveness of different probiotics for the management of this disorder.

Bifidobacterium animalis subsp. lactis, (BB-12®) is an importance probiotic, which really modifies the configuration of the gastrointestinal microbiota and improve the immune system function [4]. These facts probably make BB-12 potentially helpful for the treatment of infantile colic. In a previous trial, a low lactose relatively hydrolysate formula comprising this prebiotic prompted beneficial in decreasing the time of crying in infants with colic [5].

Given that there are not many studies regarding the efficacy of this subsp. of probiotic for treatment of IC. The aim of this trial was to evaluate the efficacy of Bifidobacterium Lactis administration on the infantile gut problems.

Methods

This double blind randomized, placebo-controlled clinical trial was conducted from December 2018 to December 2019 in pediatric department of Amin hospital, affiliated to Isfahan University of Medical Sciences.

The protocol of this study was approved by Regional ethics committee of Isfahan University of Medical Sciences with an ethics code of IR.MUI > MED.REC.1397.298 and research project number of 397266.

The trial was carried in collaboration with a team of general pediatricians and health staffs, working in the Amin clinic in the north area of Isfahan. All inspectors connected in the study attended an inspector conference during which the trial protocol was represented and deliberated, and all explanations and procedures, as well as the important factors in the controlling of IC were shared; including parental training, assurance and concern[6].

In this study, all infants aged less than 2 months old who referred to Amin hospital clinic for irritability and exertion during defecation were enrolled using simple sampling (available) method.

Infants with IC diagnosis (crying for more than three hours a day for more than 3 days a week starting from at least one week ago), exclusive breastfeeding, gestational age more than 37 weeks and birth weight more than 2500 gram were included. Those with history of organic causes of crying (such as otitis, UTI, Fisher, esophagitis, and reflux diagnosed by a pediatrician), insufficient weight gain (less than 150 grams per week), and the presence of blood in the stool were not included.

From selected infants those with a history of antibiotic consumption before or during the study and poor parental cooperation were excluded.

The selected infants randomly allocated in the two groups, using random allocation software. One group was treated with BBcare drop(containing Bifidobacterium animalis subsp. lactis made by Zisttakhmir pharmaceutical company) 5 drops per day, as BB-12 group and the other group was treated with placebo drops (containing water and sugar) at 5 drops per day, as placebo group.

Data collection and intervention

Written informed consent was obtained from the parents of all selected infants.

Selected infants examined physically by a pediatrician. Using a questionnaire, demographic, anthropometric, medical history (history of recent infection, diseases and medication use) and clinical findings (vital signs, hydration, growth, nutritional status, otoscopic examination, oral cavity, abdomen, respiratory and lymph node evaluation) were recorded.

Then, infants were followed for a 1-week as a pre-enrolment course. If after this time the diagnosis of IC was established, the infants were randomized to one of the following study groups;

-BB-12 group: parental assurance and teaching plus BB-12 (Bifidobacterium lactis BB-12®, 1 × 109 CFU/daily dose in oil maltodextrin suspension; BBCare Drop, Zisttakhmir company production

-Placebo group: parental assurance and teaching plus placebo (oil maltodextrin suspension). Parents were asked for administration to their infants

5 drops of the allocated study product, once a day, for 30 days directly in the mouth before feeding in the morning.

Instructions for care and upholding the product were offered according to the manufacturer’s suggestion. Study products were made by Zisttakhmir pharmaceutical company. The infant's parents, researcher, staff persons achieving the appraisals and study analysts were blinded to the characteristics of the treatment at every stages, i.e. allocation, interference and statistical investigation. The covering, weight, appearance, taste and smell of the study product and of the placebo were very similar and made sure in the blind conditions. The bottles restraining the placebo or the probiotic were tagged with successive numbers without any mention to the group task. We provided a diary for the infant's parents and asked them to complete it daily with data relating to consumption of daily dose of the trail product, duration and amount of weeping episodes, number of intestinal movements and firmness of infant's stool (according to the Bristol stool scale)[7], duration of sleep, potential adverse events.

During a 6 week period the infants follow up and visited by the trial peditrition for 4 times. Unplanned visits were done if necessary. The pediatricians completed a full examination for the infants at each visit and evaluated and collected data from the trial diary. Compliance was judged by assessing the diary delivered by the parent. We assessed the alterations in dietary behaviors and imaginable influences of maternal nutritional factors, by evaluating data from 7-days diet diary composed during the week before treatment .Moreover, Potential changes in maternal food were measured during the last week of the trial.

All diaries were evaluated by expert dietitians ignorant of the study goals. At each visit, the infants' parents were requested to report any adverse and side effects, unpredicted symptoms or unpredicted events connected or not to the treatment. Parents were educated to avoid any treatment with pre/ probiotic or any herbal and chemical anti-colic drugs during the trial.

Stool consistency was assessed as the number and the proportion of infants with as a minimum one stool sample of each kind per week rendering to the Bristol scale. Regular demographic and anamnestic characteristics were similar matching the two study groups. All infants were from people of middle socioeconomic rank and lived in north areas of Isfahan. All studied infants were breast fed during the whole study period. None of the subjects received any drugs for colic before study entry and throughout the study period. Seven-days food diaries were obtainable from all mothers of the babies included in the trial, and no dietary alterations were detected during the week earlier to treatment.

Statistical analysis

Data analyzed using SPSS version 21 software. Quantitative and qualitative data were reported as mean (SD) and number (percentages), respectively. Comparison of the studied variables in the studied groups and within groups during follow ups were performed using student t-test for quantitative data and Chi-square test for qualitative data. $P < 0.05$ is considered as significant level.

Results

In this study from initially enrolled neonates, 40 and 38 neonates in BB-12 and placebo groups completed the study. Characteristics of the neonates are presented in Table 1.There was not significant differences regarding studied variables in two studied groups (P > 0.05).

Table 1
Characterisrics of patients with infantile colic in BB-12 and placebo groups

Variables	BB-12 group n = 40	Placebo group n = 38	P value
Female/Male [n (%)]	16(45%)/24 (55%)	13(48%)/25 (52)	0.127
Normal vaginal delivery [n (%)]	18 (32.5%)	20 (30.5%)	0.731
Gestational age(weeks)*	38.7 (1.2)	38.7(1.1)	0.962
Birth weight(Kg)*	3.4 (0.6)	3.1(0.2)	< 0.001
Age(days)*	25.52(6.42)	23.90(1.97)	0.124
Exposure to passive smoking [n (%)]	10 (22.5)	10(21.4)	0.934
Familial risk for allergy [n (%)]	8(17)	9(15)	0.807
*Mean (SD)			

The mean of crying, vomiting episodes and defecation number at baseline and during follow ups (40 and 60 days after intervention) are presented in Fig. 2–4.Within group analysis indicated that there was significant differences in mean of crying and vomiting episodes and number of defecation in BB-12 group($P < 0.05$ decreased).In placebo group there was significant differences between mean of crying and vomiting episodes($P < 0.05$), but there was not significant difference between mean number of defecation in placebo group($P > 0.05$).

There was significant difference between groups regarding mean of crying and vomiting episodes at baseline ($P < 0.05$). It was higher significantly in BB-12 group then placebo group ($P < 0.05$).

There was not significant differences between groups regarding mean of crying and vomiting episodes and number of defecation, 40 days after intervention (P.0.05).

Between group analyses indicated that there was significant difference between groups regarding mean of crying and vomiting episodes and number of defecation ,60 days after intervention($P < 0.05$).

Frequency of different types of stool consistency are presented in Table 2.In placebo group, the stool consistency have not significant changes during follow up periods ($P > 0.05$).In BB-12 group, the stool consistency changed significantly ($P < 0.05$).

Table 2: The frequency of different forms of stool consistency (according to the Bristol stool scale) at baseline and during follow ups (40 and 60 days after intervention) in BB-12 and placebo groups

Groups/follow ups	Baseline	40 days after intervention	60 days after intervention	P value
BB-12				
- Sausage shaped but lumpy	0(0%)	0(0%)	0(0%)	
- Sausage shaped with cracks	6(15%)	17(42%)	26(65%)	<0.001
- Sausage shaped but soft	25(62%)	22(55%)	11(27%)	
- Fluffy shaped	9(22%)	1(2.5%)	3(7.5%)	
Placebo	0(0%)	1(2.5%)	4(10%)	
- Sausage shaped but lumpy	16(40%)	16(40%)	18(45%)	
- Sausage shaped with cracks	22(55%)	21(52%)	7(17%)	0.680
- Sausage shaped but soft	2(5%)	2(5%)	11(27%)	
- Fluffy shaped				
P-value	0.406	0.974	0.003	
Mann- Whitney				

The anthropometric parameters improved within the regular range from one visit to next visit and they were same in the two groups. There was no any report of antibiotic recommendation

Discussion

In this study we evaluate the effectiveness of BB-12 probiotic strain on the management of IC. Our results indicated that the probiotic have proper effect on crying and vomiting episodes for long term use (60 days after using the probiotic).In BB-12 group number of defecation, crying and vomiting episodes decreased significantly during intervention till 60 days after probiotic administration, whereas in the placebo group there was significant decrease for crying and vomiting episodes between baseline and 60 days after intervention.

The role of probiotics in the treatment of IC has been investigated in several studies [8–18]. Results of a recent systematic review and meta-analysis of clinical trials have demonstrated that probiotics could reduce the crying episodes in infants with IC. They concluded that based on available data we could not concisely assume that probiotics could improve IC by modulation of immune system or modulation of microbiota because most of the reviewed studies reported their data based on associations and recommended to plan further clinical trials base on causations [19].

Most of the trials evaluated the effectiveness of L. reuteri for IC specially in exclusively breastfed infants. However there were also studies which did not show any significant efficacy for L. reuteri [11, 20, 21].

In literature review we found only two studies regarding the effectiveness of BB-12 on IC and its related symptoms including daily average of crying. In the first trial, Vlieger et al. compared a formula supplemented with containing Lactobacillus paracasei, Paracasei *and* Bifidobacteriumanimalis, Lactis with the same formula without the probiotics in infants with IC. Their results did not indicate significant difference in duration of crying and sleeping hours between the two studied groups during the first three months of life [22].

Recently, Nocerino and colleagues reported that using the Bifiidobacterium animalis subsp. lactis BB-12strain in infants with IC result in significant decrease in daily crying episodes. They indicated that its effectiveness was observed at the end of the 2nd week of administration. They also did not report any relapse after intervention. They suggested that the effect could derive from immune and non-immune mechanisms associated with a modulation of gut microbiota structure and function [17].

Our findings were similar to their study. In addition we report its effectiveness on vomiting episodes of neonates with IC. At this time, there is no comprehensive study on the effect of BB-12 on infant vomiting which can be caused by allergy, gastroesophageal reflux, and colic. Based on the high probability role of immune allergic reactions in the gastrointestinal problems including vomiting and the possible effect of probiotics in diminishing this process, there is a strongly need for more research on this issue in the future

The explanation for this inconsistent evidence and various result in previous studies are uncertain, and there is a necessity to explore the causes behind such controversial consequences, principally with accumulating probiotic marketing, variation of strains used, and poorly understanding of colic pathogenesis considering the role of BB-12 on the infantile colic, little study has been done and more studies are needed in the future.

About the role of BB-12 recommendation on daily bowel movements and pattern of infant defecation, our study strongly showed the beneficial effects of this probiotic strain. Previous studies support a clinically relevant benefit of the probiotic strain BB-12®, on defecation frequency in healthy subjects with low defecation frequency and abdominal discomfort [20].A study about the effects of Bifidobacteriumlactis supplementation on colonic transit time and gastrointestinal symptoms in adults with functional constipation conducted by Alvin Ibarra et al. did not strongly show the effectiveness of this probiotic [4]. Another study indicated that BB-12 has a beneficial action on transit time and stool consistency [14]. Probiotics(I.e: BB-12) may intermingle with the immune system in several ways, e.g., by provoking local and systemic antibody production, by aggravating immune cell activity, by modifying signals in epithelial and immune cells, and by stimulation of phenotypic alterations in dendritic cells[23].However, the main mechanism of this effect is still unclear; and a causal relationship between the modulatory effect of probiotics on microbiota and the immune system has not been absolutely confirmed[19].

Previous findings indicate that BB-12 modifies the proliferation of human peripheral blood mononuclear cells and cytokines expression, [20, 24] with protective action against gastrointestinal infections in infants and children. In the setting of IC, these properties could be responsible for a helpful shaping of gut microbiota arrangement. It is recognized that a confident modulation of LL-37, HBD-2, and sIgA expressions into the gut lumen leads in a positive effect on gut micro biota structure and butyrate production [21]. These effects seem mainly significant in IC, where dysbiosis with augmented presence of proteobacteria and reduced presence of Bifidobacteria with declined rate production have been established [25, 26].

The limitations of current study were small number of previous researches for the studies related to this type of probiotic, short term follow up and the inclusion of exclusively breastfeeding infants. It is suggested that future trials can help in well clarifying the action of BB-12 in infant colic and intestinal homeostasis.

The strengths of this study were its well-designed methods (the placebo-controlled, randomized double blind trial), the use of validated procedure for IC diagnosis and the use of a well-defined probiotic strain.

Conclusion

Findings of our study offer compelling signals for the effectiveness of Bifidobacteriumanimalis subsp. lactis BB-12 in the management of some intestinal problems. Though these findings could be supportive evidences for the important role of gut microbiota as goal of intervention to improvement in bowel movement and comfortable defecation in IC, but further studies are needed in this field specially for

evaluation of different types of probiotics in combination with each other's and with different dosages. It is important to emphasize that this study evaluated a particular well-characterized probiotic strain, and that these outcomes cannot be generalized for other probiotic strains.

Declarations

Funding: Isfahan University of Medical Sciences

Conflicts of interest/Competing interests: Authors have no conflict of interest

Ethics approval: Ethics code of IR.MUI> MED.REC.1397.298 and research project number of 397266

Consent to participate: All patients read and confirmed the written consent which was prepared by the authors (The consent include the method and aims of the study and the patients' rights)

Consent for publication: N/A

Availability of data and material: The data are available

Authors' contributions: All authors have contributed in the concept and all stages of the work. All of them have contributed in preparing the first draft of the paper and read and approved the paper.

References

1. James-Roberts IS. In Barr RG, St James-Roberts I, Keefe MR, eds (2001) New Evidence on Unexplained Early Infant Crying: Its Origins, Nature and Management. Skillman, New Jersey: Johnson & Johnson Pediatric Institute 2001:5-24
2. Zeevenhooven J, Browne PD, L'Hoir MP, de Weerth C, Benninga MA (2018) Infant colic: mechanisms and management. Nat Rev GastroenterolHepatol 15:479–496
3. Indrio F, Dargenio VN, Giordano P, Francavilla R (2019) Preventing and treating colic. AdvExp Med Biol. 1125:49–56. https://doi. org/10.1007/5584_2018_315
4. Jungersen M, Wind A, Johansen E, Christensen JE, Stuer-Lauridsen B, Eskesen D (2014) The science behind the probiotic strain bifidobacteriumanimalis subsp. lactis BB-12®. Microorganisms. 2:92-110
5. Xinias I, Analitis A, Mavroudi A et al (2017) Innovative dietary intervention answers to baby colic. PediatrGastroenterolHepatolNutr 20:100–106
6. Bellaïche M, Levy M, Jung C (2013) Treatments for infant colic. J Pediatr Gastroenterol Nutr 57:S27–S30
7. -Heaton KW, Radvan J, Cripps H, Mountford RA, Braddon FE, Hughes AO (1992) Defecation frequency and timing, and stool form in the general population: a prospective study. Gut 33:818–824
8. Sung V, Hiscock H, Tang ML et al (2014) Treating infant colic with the probiotic Lactobacillus reuteri: double blind, placebo controlled randomised trial. BMJ 348:2107

9. Chau K, Lau E, Greenberg S et al (2015) Probiotics for infantile colic: a randomized, double-blind, placebo-controlled trial investigating Lactobacillus reuteri DSM 17938. J Pediatr 166:74–78
10. Mi GL, Zhao L, Qiao DD, Kang WQ, Tang MQ, Xu JK (2015) Effectiveness of Lactobacillus reuteri in infantile colic and colicky induced maternal depression: a prospective single blind randomized trial. Antonie Van Leeuwenhoek 107:1547–1553
11. Pärtty A, Lehtonen L, Kalliomäki M, Salminen S, Isolauri E (2015) Probiotic Lactobacillus rhamnosus GG therapy and microbiological programming in infantile colic: a randomized, controlled trial. Pediatr Res 78:470–475
12. Kianifar H, Ahanchian H, Grover Z et al (2014) Synbiotic in the management of infantile colic: a randomised controlled trial. J Paediatr Child Health 50:801–805
13. Urbańska M, Szajewska H (2014) The efficacy of Lactobacillus reuteri DSM 17938 in infants and children: a review of the current evidence. Eur J Pediatr 173:1327–1337
14. Nocerino R, De Filippis F, Cecere G, Marino A, Micillo M, Di Scala C, de Caro C, Calignano A, Bruno C, Paparo L, Iannicelli AM (2020) The therapeutic efficacy of Bifidobacteriumanimalis subsp. lactis BB-12® in infant colic: A randomised, double blind, placebo-controlled trial. Aliment Pharmacol Ther 51(1):110–120
15. Savino F, Cresi F, Pautasso S et al (2004) Intestinal microflora in breastfed colicky and non-colicky infants. ActaPaediatr 93(6):825–829
16. Pärtty A, Lehtonen L, Kalliomäki M, Salminen S, Isolauri E (2015) Probiotic Lactobacillus rhamnosus GG therapy and microbiological programmingin infantile colic: a randomized, controlled trial. Pediatr Res 78(4):470–475
17. Akbarian-Rad Z, Zahedpasha Y, Ahmadpour-Kacho M, Rajabnia R, Tohidi FT (2013) Intestinal lactobacillus species: Is it equal in colicky and non-colicky breastfed infants? Iran J Neonatol 4(2):1–4
18. Savino F, Quartieri A, De Marco A et al (2016) Comparison of formulafed infants with and without colic revealed significant differences in total bacteria, Enterobacteriaceae and faecal ammonia. ActaPaediatr 106(4):573–578
19. Skonieczna-Żydecka K, Janda K, Kaczmarczyk M, Marlicz W, Łoniewski I, Łoniewska B (2020) The Effect of Probiotics on Symptoms, Gut Microbiota and Inflammatory Markers in Infantile Colic: A Systematic Review, Meta-Analysis and Meta-Regression of Randomized Controlled Trials. Journal of clinical medicine 9(4):999
20. Latvala S, Pietilä TE, Veckman V et al (2008) Potentially probiotic bacteria induce efficient maturation but differential cytokine production in human monocyte-derived dendritic cells. World J Gastroenterol 14:5570-5581
21. BerniCanani R, De Filippis F, Nocerino R et al (2017) Specific signatures of the gut microbiota and increased levels of butyrate in children treated with fermented cow's milk containing heat killed Lactobacillus paracasei CBA L74. Appl Environ Microbiol 83:e01206–e1217

22. Vlieger AM, Robroch A, van Buuren S et al (2009) Tolerance and safety of Lactobacillus paracasei ssp. paracasei in combination with Bifidobacteriumanimalis ssp. lactis in a prebioticcontaining infant formula: a randomised controlled trial. Br J Nutr 102(6):869–875
23. Ibarra A, Latreille-Barbier M, Donazzolo Y, Pelletier X, Ouwehand AC (2018) Effects of 28-day Bifidobacteriumanimalis subsp. lactis HN019 supplementation on colonic transit time and gastrointestinal symptoms in adults with functional constipation: a double-blind, randomized, placebo-controlled, and dose-ranging trial. Gut Microbes 9(3):236–251
24. Lopez P, Gueimonde M, Margolles A, Suarez A (2010) Distinct Bifidobacterium strains drive different immune responses in vitro. Int J Food Microbiol 138:157–165
25. Savino F, Quartieri A, De Marco A et al (2017) Comparison of formulafed infants with and without colic revealed significant differences in total bacteria, Enterobacteriaceae and faecal ammonia. ActaPaediatr 106:573–578
26. Savino F, Cordisco L, Tarasco V, Calabrese R, Palumeri E, Matteuzzi D (2009) Molecular identification of coliform bacteria from colicky breastfed infants. ActaPaediatr 98:1582–1588

Figures

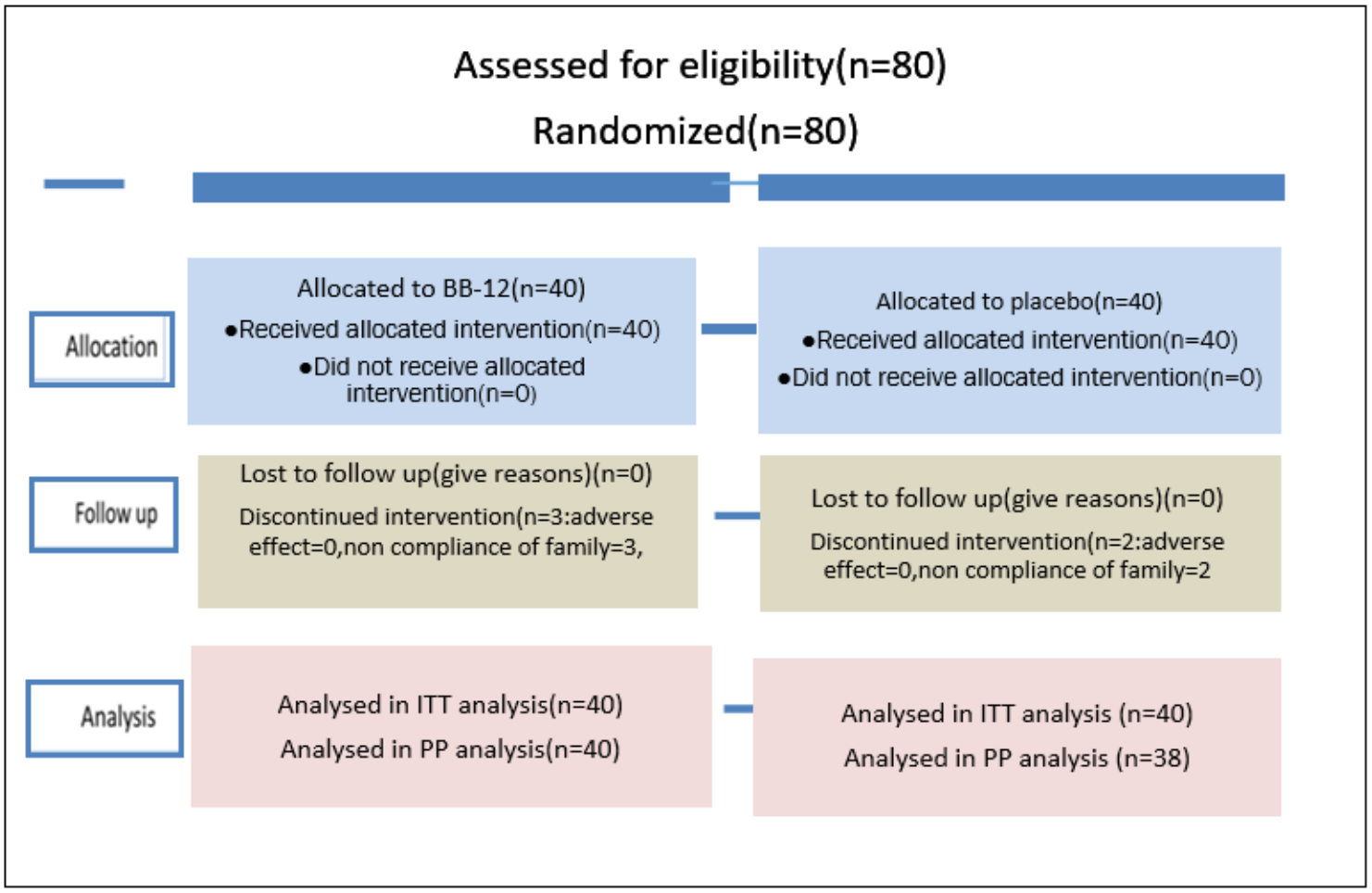

Figure 1

The consort diagram of the study

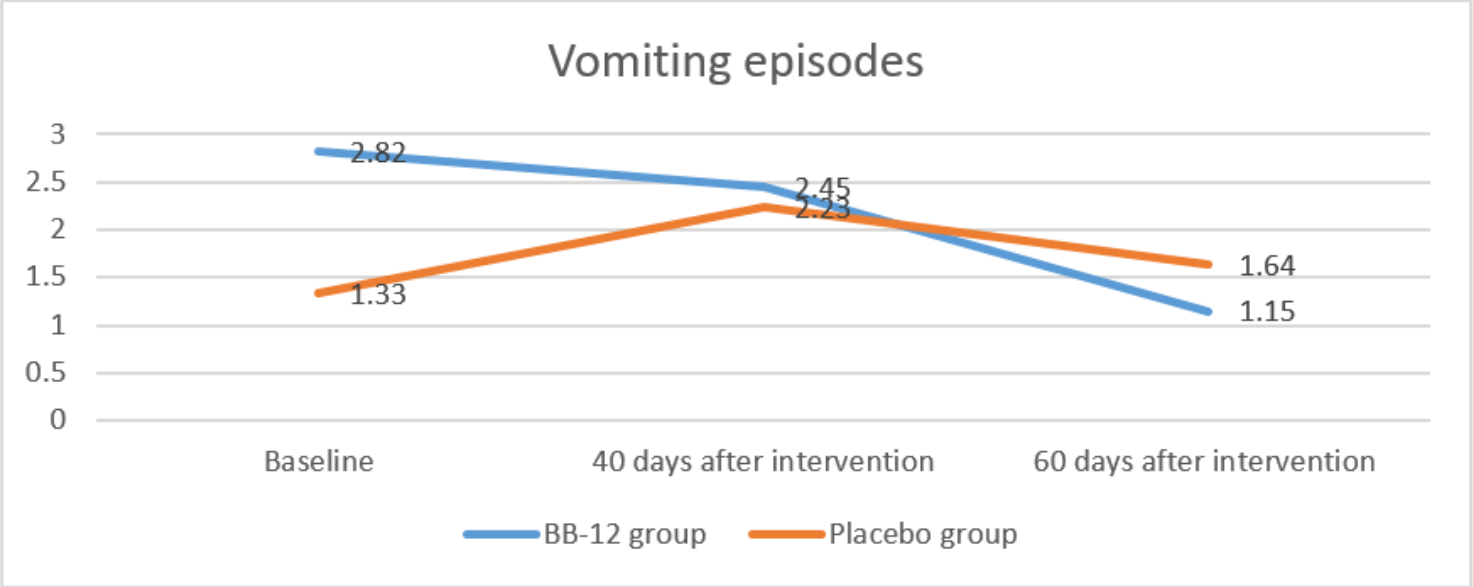

Figure 2

Mean of vomiting episodes number at baseline and 40 and 60 days after intervention in BB-12 and placebo groups

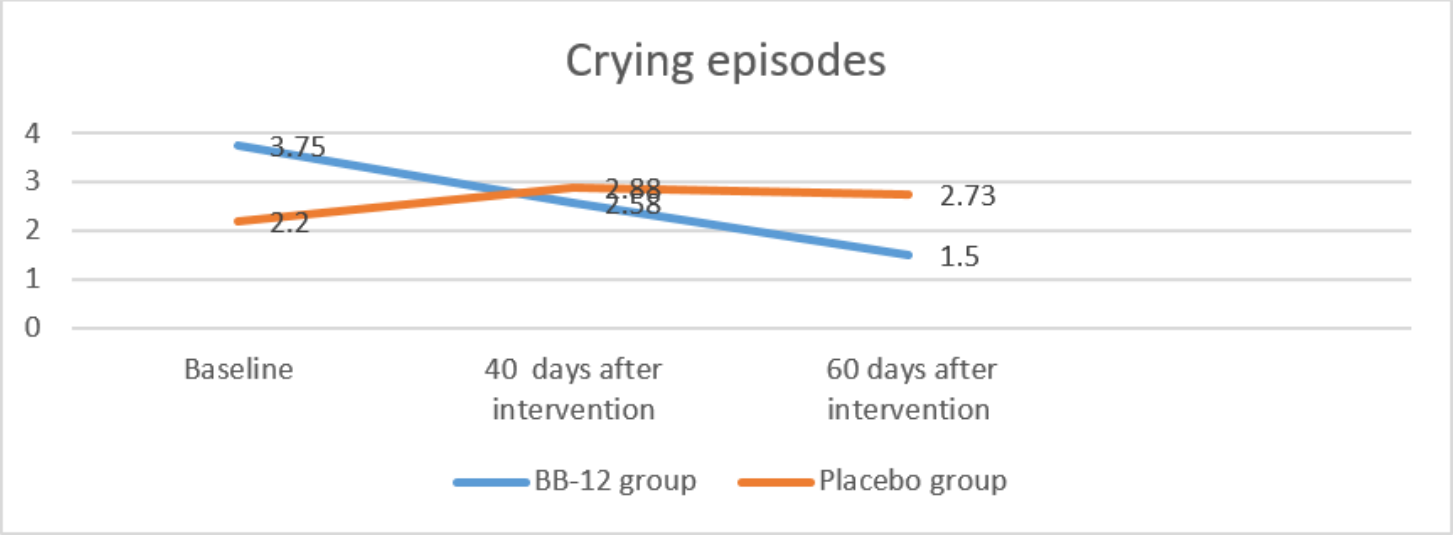

Figure 3

Mean of crying episodes number at baseline and 40 and 60 days after intervention in BB-12 and placebo groups

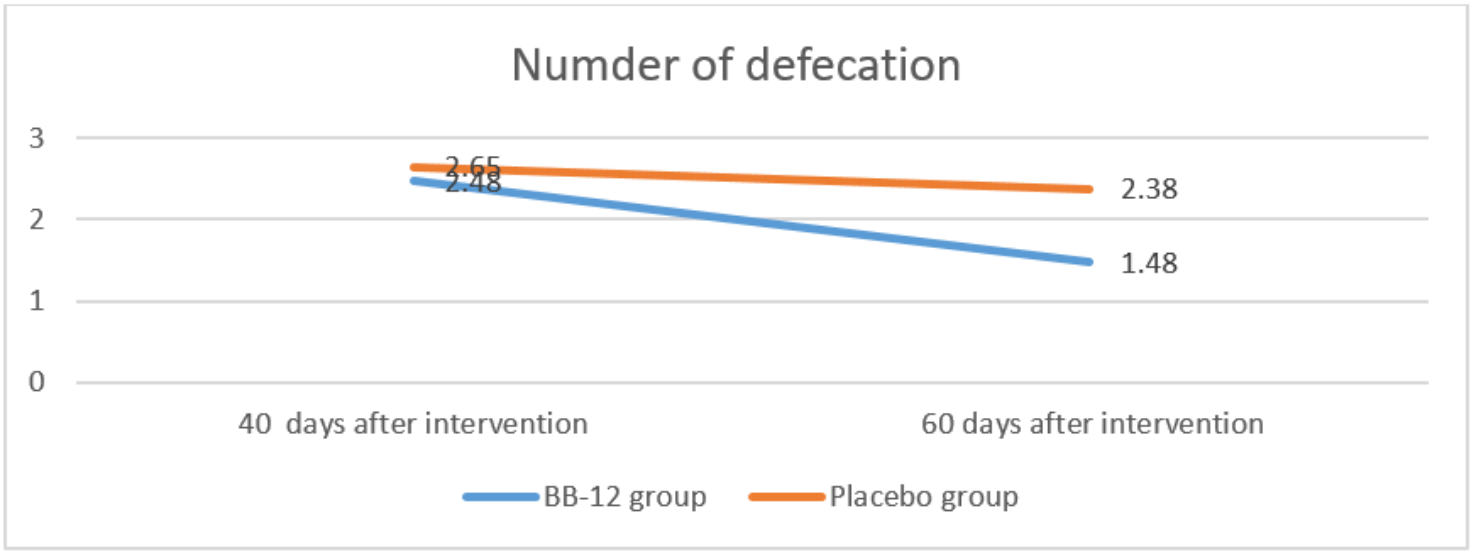

Figure 4

Mean number of defecation at baseline and 40 and 60 days after intervention in BB-12 and placebo groups