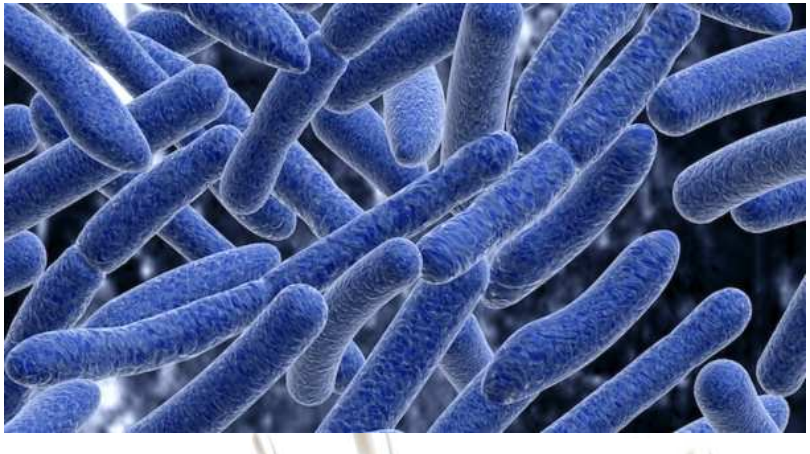
DAL MICROBIOTA AL PROBIOTICO: *BIFIDOBATTERI NELLA PREVENZIONE E NEL TRATTAMENTO*

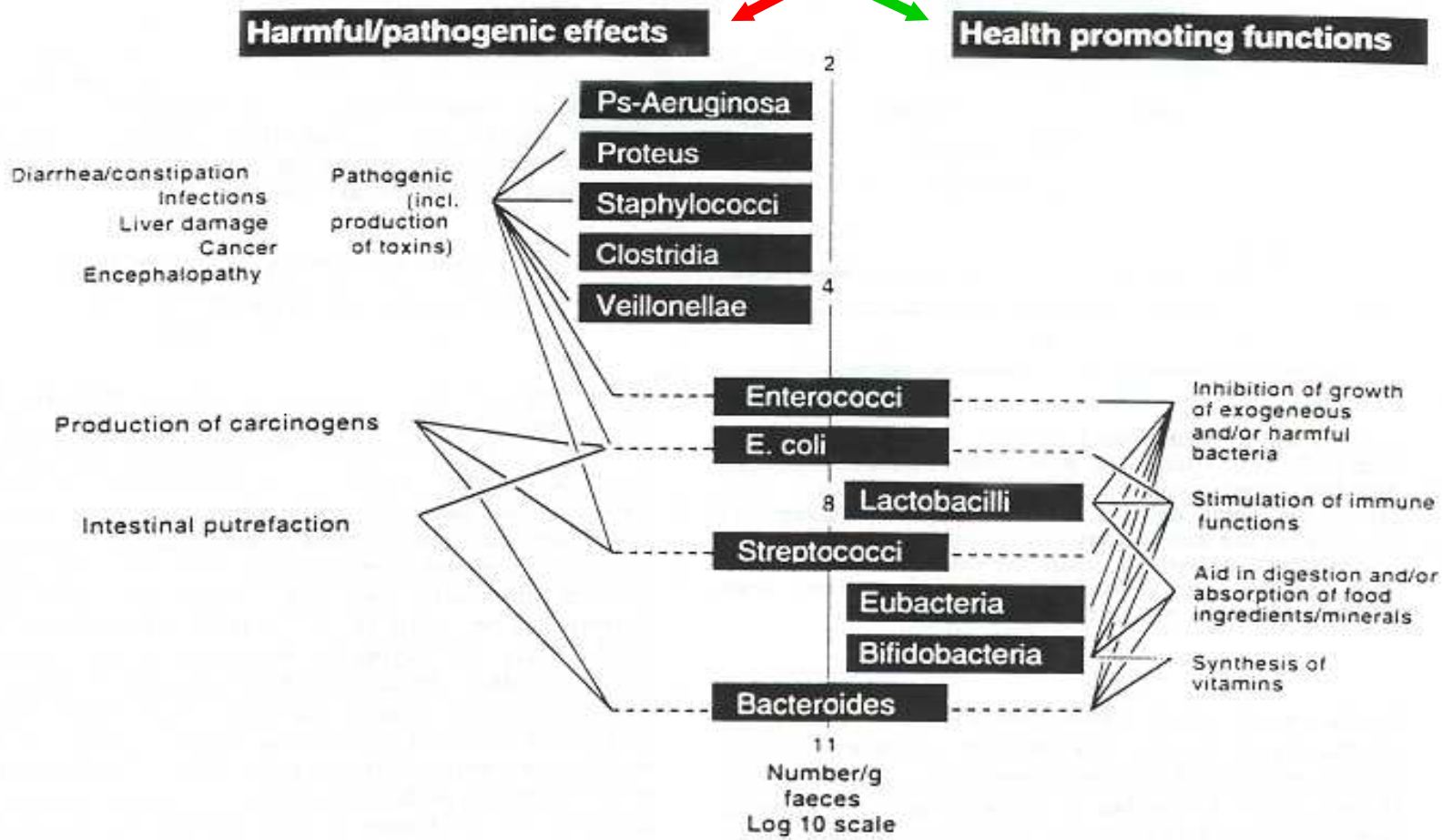
Annamaria Staiano
Dipartimento di Pediatria
Universita' degli Studi di Napoli
"Federico II"

Il Microbiota intestinale

Vi è una crescente evidenza del ruolo fondamentale che la composizione del microbiota intestinale svolge nello sviluppo postnatale del sistema immunitario.



Good and Bad



J Nutr, 1998; 125: 1423

Il microbiota intestinale

- L'interazione del microbiota intestinale con le cellule dell'epitelio intestinale influenza l'equilibrio di processi infiammatori e anti-infiammatori mucosali, che svolgono un ruolo significativo nelle malattie GI e nello stato di salute

- Il microbiota intestinale rappresenta un organo dimenticato che può eseguire molte funzioni fisiologiche e quindi può influenzare profondamente la biologia umana.

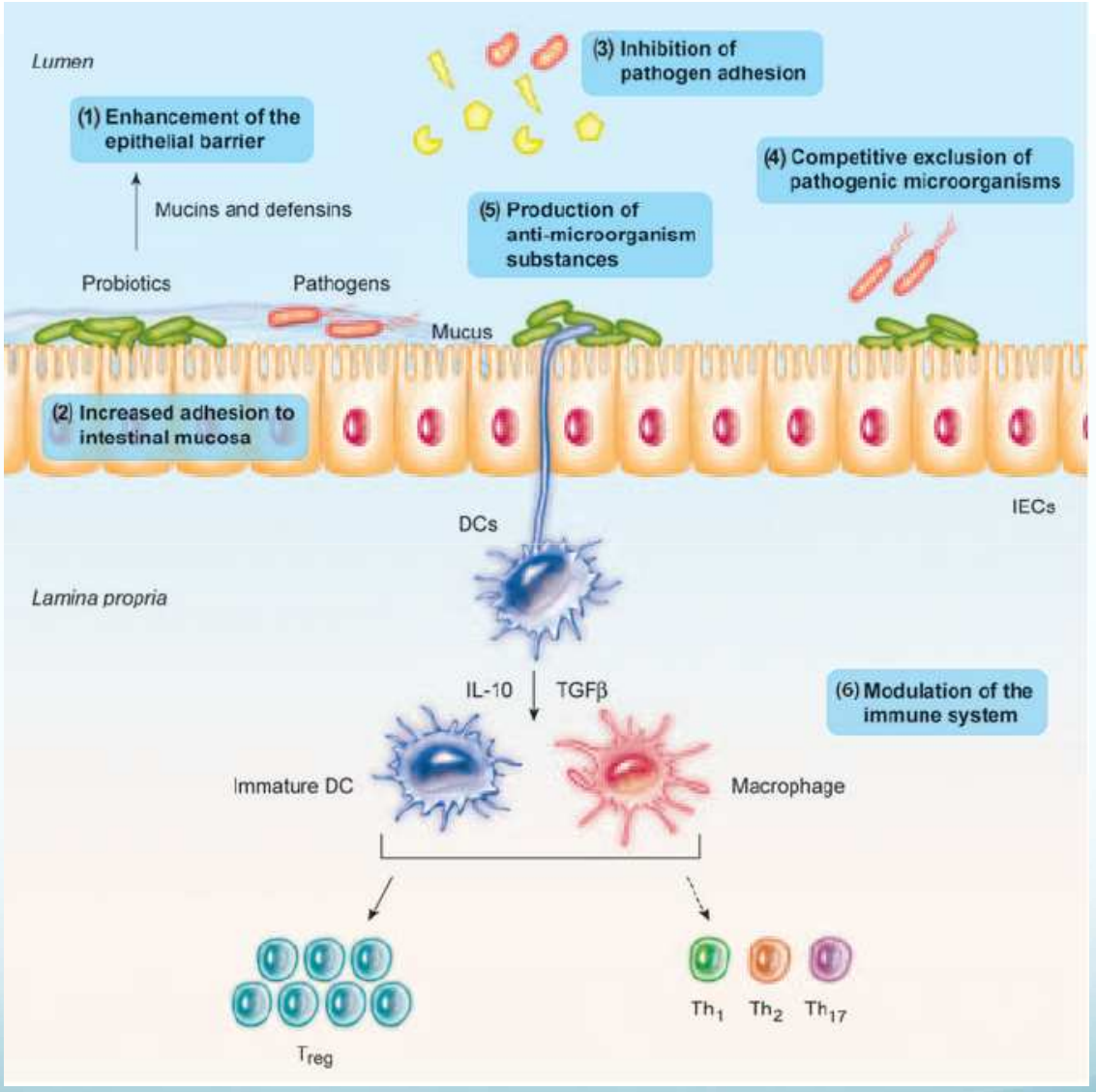
Mazmanian SK et al Nature 2008;453:620-5

Probiotico

- *Probiotico:* integratori orali o prodotti alimentari contenenti un numero sufficiente di microrganismi vitali capaci di modificare la microflora dell'ospite con potenziali effetti benefici sulla salute.

Dan W. Thomas et al. Pediatrics 2010;126;1217

Probiotic Mechanisms of Action



Bermudez-Brito M et al. Ann Nutr Metab. 2012;61(2):160-74

Principali microorganismi utilizzati come probiotici

Batteri

Batteri Lattici

Lactobacillus acidophilus
Lactobacillus bulgaricus
Lactobacillus brevis
Lactobacillus casei
Lactobacillus casei Shirota
Lactobacillus johnsonii
Lactobacillus plantarum
Lactobacillus rhamnosus GG
Lactobacillus reuteri
Lactobacillus salivarius

Bifidobatteri

Bifidobacterium animalis
Bifidobacterium bifidum
Bifidobacterium breve
Bifidobacterium infantis
Bifidobacterium lactis
Bifidobacterium longum

Altri batteri

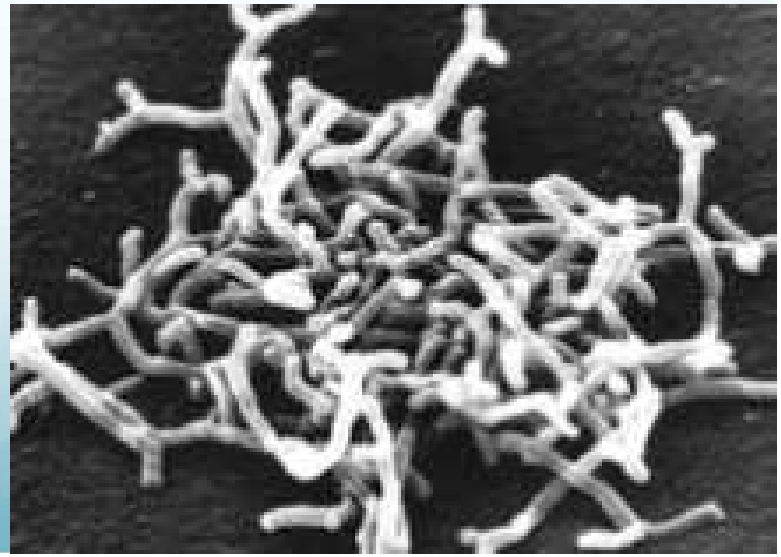
Bacillus cereus
Escherichia coli Nissle 1917
Streptococcus thermophilus

Lieviti

Saccharomyces boulardii

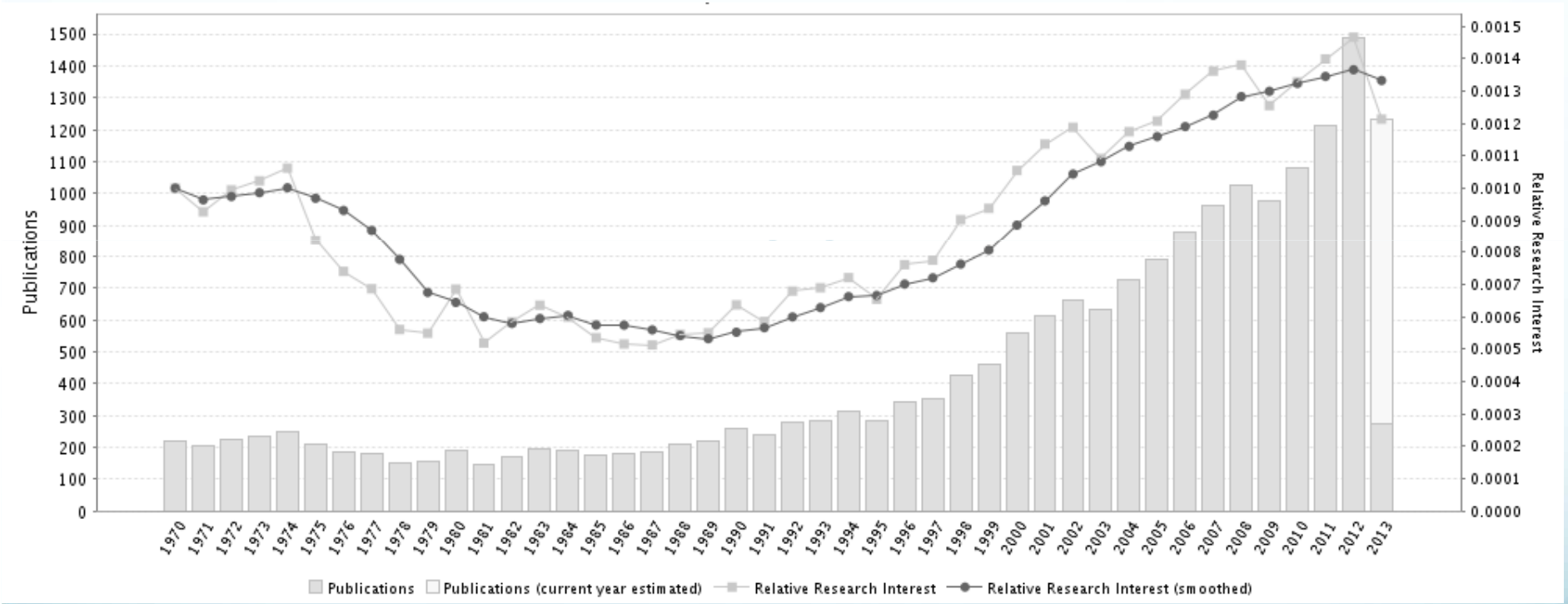
DAL MICROBIOTA AL PROBIOTICO: *BIFIDOBATTERI NELLA PREVENZIONE E NEL TRATTAMENTO*

Lactobacillus acidophilus

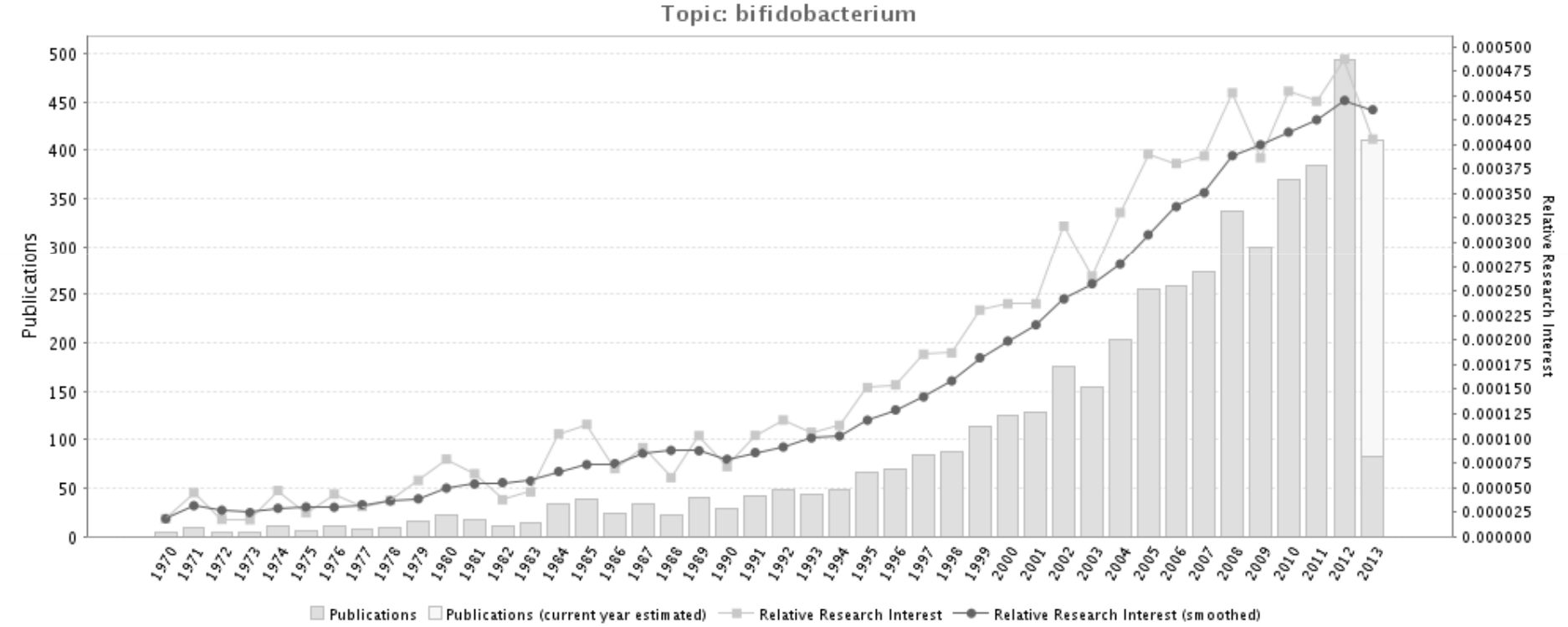


Bifidobacterium longum

Lactobacillus:
N. of Publications from 1970’

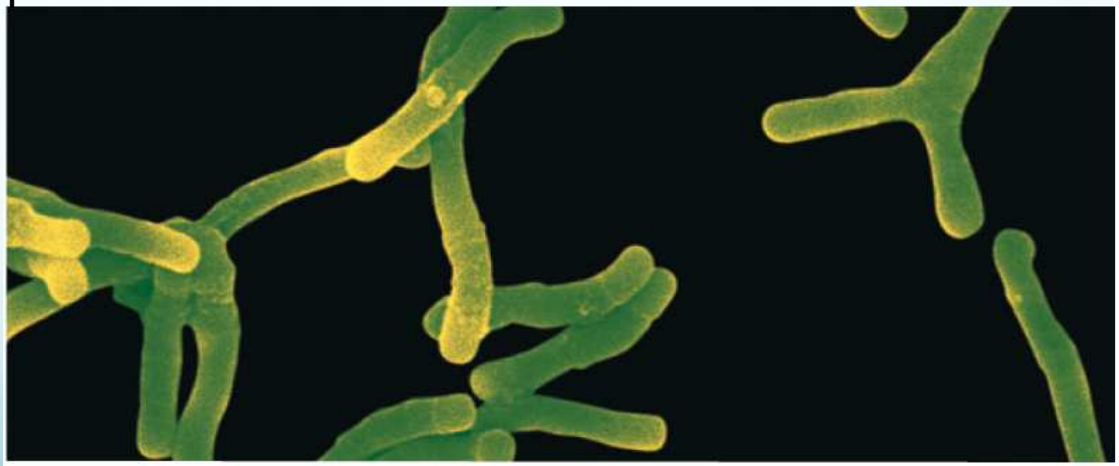


BIFIDOBACTERIUM:
N. of Publications from 1970’



BIFIDOBATTERI

- ✓ I Bifidobatteri sono batteri gram positivi e rigorosamente anaerobici, con un metabolismo fermentativo che produce acetato e lattato come prodotti finali.
- ✓ La forma delle cellule dei Bifidobatteri varia tra le specie, ma in generale si tratta di bastoncelli, sottili, e con estremità leggermente bulbose.
- ✓ Una o entrambe le estremità tendono a biforcarsi. Queste doppie punte danno al batterio il suo nome, dalla parola latina bifidus: 'Diviso in due'.
- ✓ I bifidobatteri sono microrganismi dominanti nel microbiota intestinale del neonato allattato al seno e sono anche importanti membri del microbioma intestinale adulto.



Lee JH, O'Sullivan DJ. Genomic insights into bifidobacteria. Microbiol Mol Biol Rev. 2010;74:378–416.
Biavati, B. et al. Annals of Microbiology 2000; 50: 117-131.
Ventura, M et al. International Journal of Food Microbiology 2007;120: 2-12.

Promoting Bifidobacteria in the human infant intestine: why, how and which one?

- During breast-feeding, gut microbiota become dominated by bifiobacteria
- In formula-fed the microbiota consists of bifidobacteria, E. Coli, and bacteriodes, which is reflected by alterations of stool color, consistency, and odor.
- Bifidobacteria constitutes 60% to 91% and almost 50% of the fecal bacteria of breast-feeding and formula-fed infants, respectively, whereas by adulthood bifidobacteria comprised <5%.
- After weaning, the fecal microbiota resembles the adult microbiota.

Donovan SM. JPGN 2011; 52:648-9

Development and Differences of Intestinal Flora in the Neonatal Period in Breast-Fed and Bottle-Fed Infants

- On day 6, bifidobacteria were the predominant organisms in the stool of breast-fed infants, whereas enterobacteria were the predominant organisms in formula-fed infants, exceeding bifidobacteria by approximately 10:1.

- At 1 month of age, bifidobacteria were the most prevalent organisms in both groups but the number of these organisms in the stool of bottle.fed infants was approximately one tenth that of breast infant

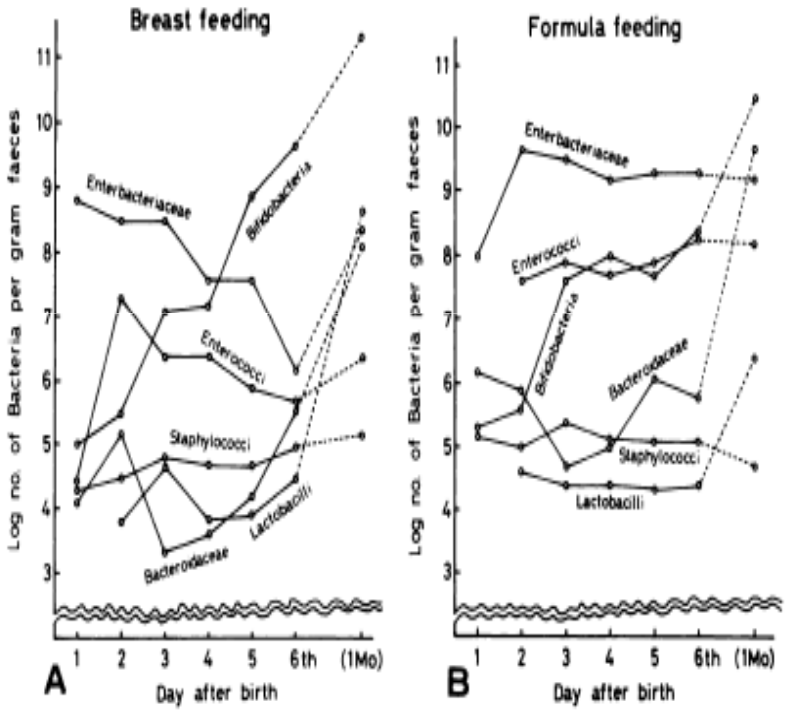
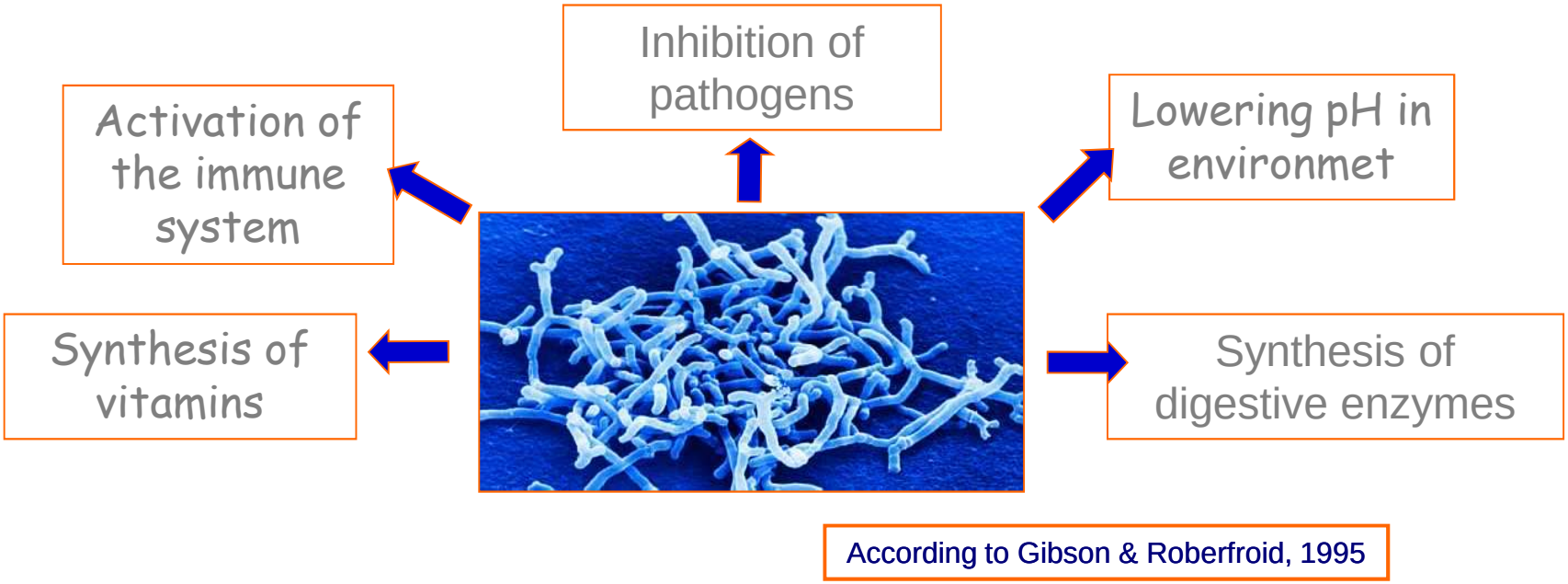


Figure. Changes in counts of individual bacterial groups in breast-fed (A) and bottle-fed (B) infants.

Yoshioka H et al. Pediatrics 1983; 72: 317-21

Protective Properties of Bifidobacteria



According to Gibson & Roberfroid, 1995

- Protection against enteral infections in a phase of insufficient immune response (Koletzko et al., 1998; Heine, 1998)
- Induction of oral tolerance towards dietary allergens (Hanson & Telemo, 1997)

Principali campi di applicazione del Bifidobacterium in pediatria

- Allergia
- Dermatite atopica
- Infezioni
- Bifidobatteri nel neonato pretermine
- Disordini Funzionali Gastrointestinali

Principali campi di applicazione del Bifidobacterium in pediatria

ALLERGIA E DERMATITE ATOPICA

Protective effects of breast feeding

Reduced incidence of allergic or atopic diseases

- Studies on the composition of intestinal microflora in children 2-year-old allergic and non-allergic showed that the prevalence of bifidobacteria is lower in allergic children, compared to the number of Staphylococcus aureus and enterobacteria

  Bjorksten B et al. Clin Exp Allergy 1999; 108:342-6

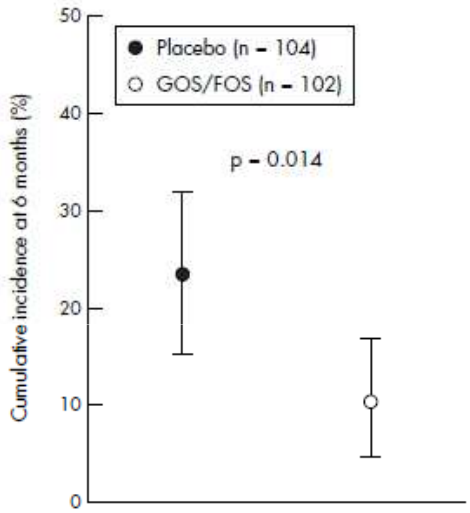
- Infants with allergies often show less colonization with bifidobacteria during the first year of life.

  Bjorksten B et al. J Allergy Clin Immunol 2001; 108:516-20

- In humans, the intake of probiotics, such as lactobacilli and bifidobacteria, can prevent allergic diseases.

  Viljanen M et al. Allergy 2005; 60: 494-500

A mixture of prebiotic oligosaccharides reduces the incidence of atopic dermatitis during the first six months of age



Cumulative incidence of AD at 6 months of age in the group fed a formula supplemented with GOS and FOS or maltodextrins as placebo. Data are expressed as mean (95% CI).

Prebiotic supplements associated with significantly higher number of fecal bifidobacteria compared with controls; no differences in lactobacilli.

Oligosaccharides modulate postnatal immune development by altering bowel flora and have a potential role in primary allergy prevention during infancy.

Moro G et al. Arch Dis Child 2006;91:814–819

Maternal probiotic supplementation during pregnancy and breast-feeding reduces the risk of eczema in the infant

Mothers with allergic disease and atopic sensitization were randomly assigned to receive (1) Lactobacillus rhamnosus LPR and Bifidobacterium longum BL999 (LPR1BL999), (2) L paracasei ST11 and B longum BL999 (ST111BL999), or (3) placebo, beginning 2 months before delivery and during the first 2 months of breast-feeding.

The risk of developing eczema during the first 24 months of life was significantly reduced in infants of mothers receiving LPR1BL999 and ST111BL999

	Placebo	LPR+BL999	ST11+BL999
Eczema			
Ratio (%)	44/62 (71)	21/73 (29)	20/70 (29)
OR (95% CI)*		0.17 (0.08-0.35)	0.16 (0.08-0.35)
P value†		<.001	<.001
Chronically persistent eczema			
Ratio (%)	16/62 (26)	7/73 (10)	4/70 (6)
OR (95% CI)*		0.30 (0.12-0.80)	0.17 (0.12-0.80)
P value†		.016	.003
Skin prick test positive			
Ratio (%)	17/65 (26)	17/76 (22)	19/73 (26)
OR (95% CI)*		0.81 (0.38-1.76)	0.99 (0.46-2.13)
P value†		.60	.99

Prevention regimen with specific probiotics administered to the pregnant and breast-feeding mother, that is, prenatally and postnatally, is safe and effective in reducing the risk of eczema in infants with allergic mothers positive for skin prick test.

Rautava et al. J Allergy Clin Immunol 2012;130:1355-60

Principali campi di applicazione del Bifidobacterium in pediatria

INFEZIONI

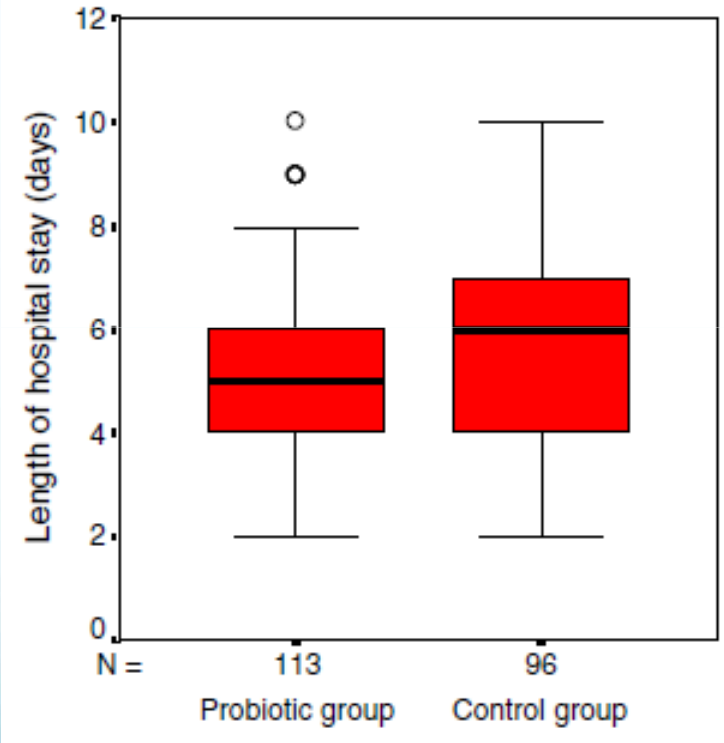
Novel Probiotic *Bifidobacterium longum* subsp. *infantis* CECT 7210 Strain Active against Rotavirus Infections[▽]

- A novel Bifidobacterium longum subsp. infantis strain was isolated from infant feces and selected, based on its capacity to inhibit in vitro rotavirus Wa replication (up to 36.05% infectious foci reduction) and also to protect cells from virus infection (up to 48.50% infectious foci reduction) in both MA-104 and HT-29 cell lines

- Bifidobacterium longum subsp. infantis CECT 7210 can be considered a probiotic able to inhibit rotavirus infection

Muñoz JA et al. Appl Environ Microbiol. 2011 Dec;77(24):8775-83

The effect of a multispecies synbiotic mixture on the duration of diarrhea and length of hospital stay in children with acute diarrhea in Turkey: Single blinded randomized study



- ✓ Prospective randomized multicenter single blinded clinical trial in hospitalized children with acute watery diarrhea.
- ✓ All children were treated with conventional hydration therapy with or without a daily dose of a synbiotic (2.5×109 CFU live bacteria including Lactobacillus acidophilus, Lactobacillus rhamnosus, Bifidobacterium bifidum, Bifidobacterium longum, Enterococcus faecium, and 625 mg fructooligosaccharide) for 5 days.
- ✓ The duration of diarrhea was significantly shorter (~36 h) in children receiving the synbiotic group than the controls

Dinleyici EC et al. Eur J Pediatr. 2012 Dec 14

Principali campi di applicazione
del Bifidobacterium in pediatria

INFEZIONI NEL NEONATO PRETERMINE

Probiotics Reduce the Risk of Necrotizing Enterocolitis in Preterm Infants: A Meta-Analysis

- Nine eligible trials randomizing 1,425 infants were included.
- In a meta-analysis, enteral probiotics supplementation significantly reduced the incidence of severe NEC and mortality
- There was no evidence of significant reduction of nosocomial sepsis or days on total parenteral nutrition.

Conclusion: **Enteral supplementation of probiotics reduces the risk of severe NEC and mortality in preterm infants.**

AlFaleh K. Et al. Neonatology 2010;97:93–99

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review)

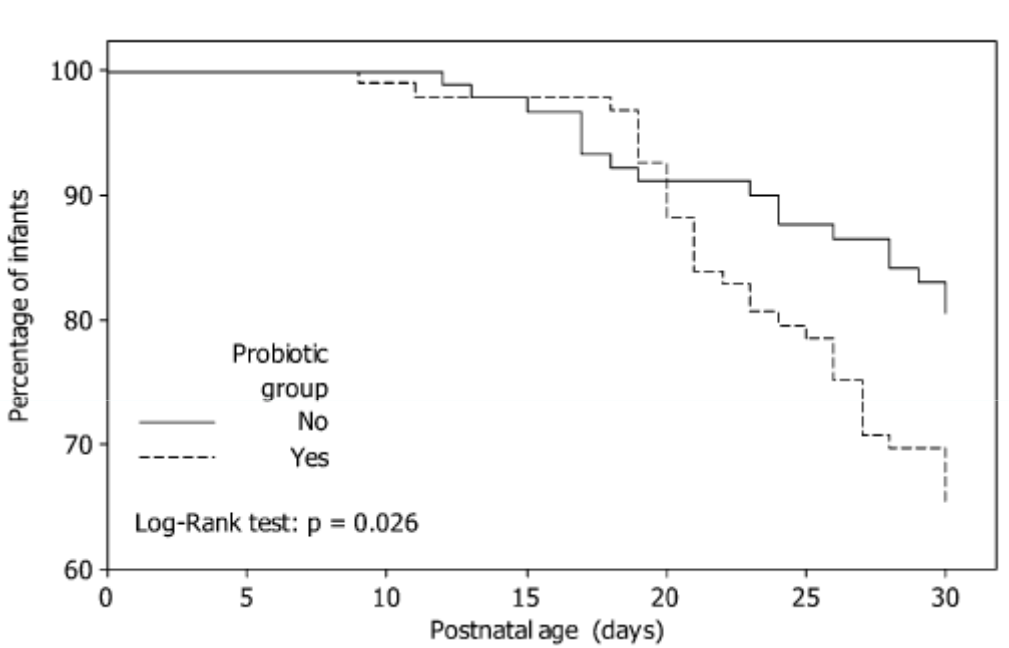
- In a meta-analysis of trial data, enteral probiotics supplementation significantly reduced the incidence of severe NEC (stage II or more) and mortality
- There was no evidence of significant reduction of nosocomial sepsis.
- The included trials reported no systemic infection with the probiotics supplemental organism.

AUTHORS' CONCLUSIONS:

- *Enteral supplementation of probiotics prevents severe NEC and all cause mortality in preterm infants.*
- *Our updated review of available evidence supports a change in practice.*
- *More studies are needed to assess efficacy in ELBW infants and assess the most effective formulation and dose to be utilized.*

Alfaleh K et al. Cochrane Database Syst Rev. 2011 Mar 16;(3):CD005496

Efficacy of *Bifidobacterium breve* and *Lactobacillus casei* oral supplementation on necrotizing enterocolitis in very-low-birth-weight preterm infants: a double-blind, randomized, controlled trial[1–3]



Kaplan-Meier survival curves showing the probability of not achieving the complete transition time of orogastric tube feeding to breastfeeding in the probiotics and control groups

- ✓ Oral supplementation of B. breve and L. casei reduced the occurrence of NEC (Bell's stage 2).
- ✓ It was considered that an improvement in intestinal motility might have contributed to this result.

Braga TD et al. Am J Clin Nutr. 2011 Jan;93(1):81-6

Effect of *Bifidobacterium* administration on very-low-birthweight infants

- ✓ **The early administration of B. bifidum to VLBW infants seems effective in promoting growth during the stay in the neonatal intensive care unit without increasing the incidence of morbidity.**
- ✓ **The preferable timing of starting the probiotic supplementation for VLBW infants is at latest less than 48 h after birth.**

Table 5 Start of supplementation and *Bifidobacterium* colonization at 1 week of age

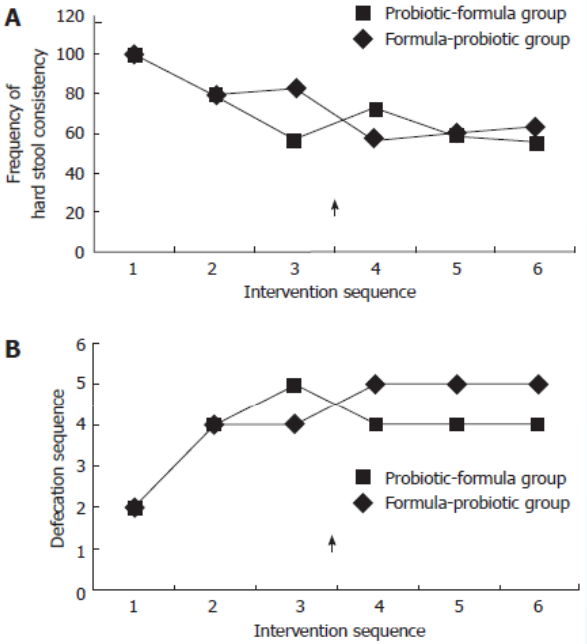
Bifidobacterium	Start of supplementation (hours after birth)				*P*-value
	0–24 ($n = 9$)	24–48 ($n = 9$)	48–72 ($n = 7$)	72- ($n = 11$)	
Logarithm of copy numbers	7.2 ± 0.6	9.7 ± 0.9	8.3 ± 0.8	9.3 ± 1.5	NS
Colonization rate (%)	2/9(22.2)	9/9(100)	5/7(71.4)	9/11(81.8)	<0.05†

Yamasaki C et al. Pediatr Int. 2012 Oct;54(5):651-6.

Principali campi di applicazione
del Bifidobacterium in pediatria

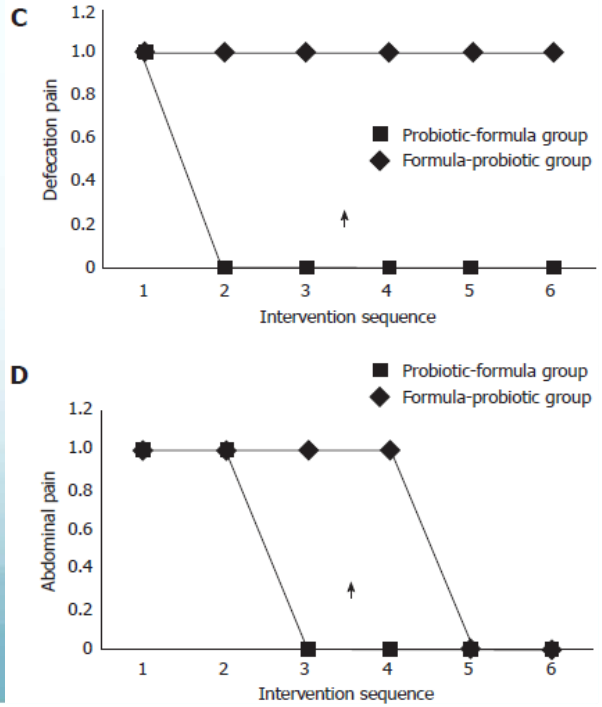
DISORDINI FUNZIONALI GSTROINTESTINALI

Pediatric functional constipation treatment with *Bifidobacterium*-containing yogurt: A crossover, double-blind, controlled trial



❑A crossover double-blind formula-controlled trial was carried out on 59 students (age range: 5-15 years) presenting a functional chronic intestinal constipation diagnostic

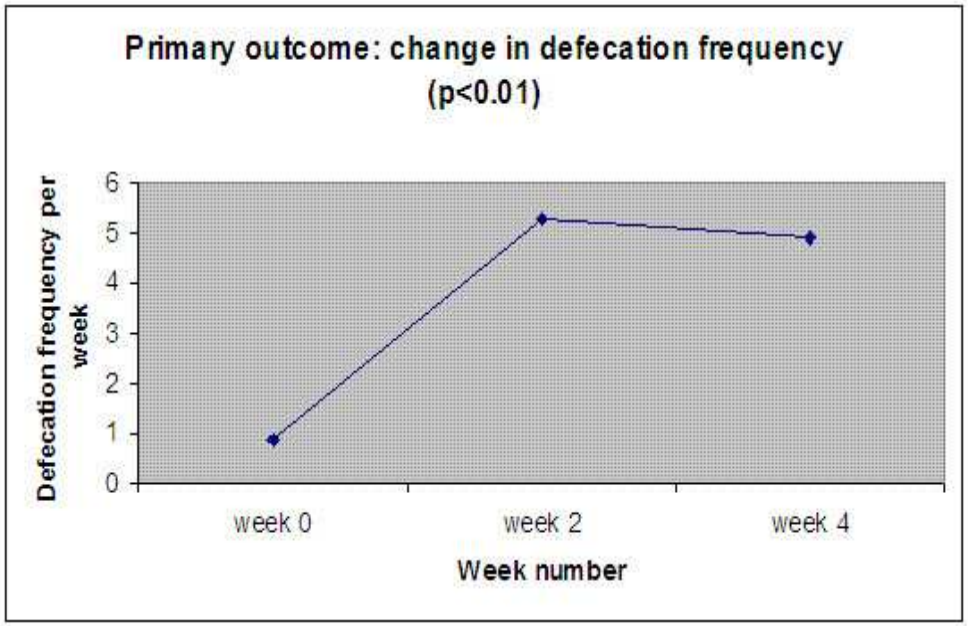
❑ The students were randomized in two groups to receive a goat yogurt supplemented with 109 colony forming unit/mL Bifidobacterium longum (B. longum) (probiotic) daily or only the yogurt for a period of 5 wk (formula).



Afterwards, the groups were intercrossed for another 5 wk. An improvement in defecation frequency and abdominal pain was observed using both supplemented and non-supplemented yogurt, but an additional improvement with B. longum supplementation was obtained.

Paula VP Guerra *World J Gastroenterol 2011 14; 17(34): 3916-3921*

Is *Bifidobacterium breve* effective in the treatment of childhood constipation? Results from a pilot study



- Twenty children (75% male, mean age 7.4) were included in this pilot study.
- The defecation frequency per week significantly increased from 0.9 (0-2) at baseline to 4.9 (0-21) in week 4 ($p < 0.01$).
- The mean stool consistency score increased from 2.6 (2-4) at baseline to 3.5 (1-6) in week 4 ($p = 0.03$).
- The number of faecal incontinence episodes per week significantly decreased from 9.0 (0-35) at baseline to 1.5 (0-7) in week 4 ($p < 0.01$).
- Abdominal pain episodes per week significantly decreased from 4.2 (0-7) at baseline to 1.9 (0-7) in week 4 ($p = 0.01$).

Conclusion

Bifidobacterium breve is effective in increasing stool frequency in children with functional constipation. Furthermore it has a positive effect with respect to stool consistency, decreasing the number of faecal incontinence episodes and in diminishing abdominal pain.

Tabbers MM et al. Nutr J. 2011 Feb 23;10:19.

Compositional Development of *Bifidobacterium* and *Lactobacillus* Microbiota Is Linked with Crying and Fussing in Early Infancy

- *Behavioral patterns of 89 infants during the 7th and 12th week of life were recorded in parental diaries.*
- *Infants' gut microbiota profiles were investigated by several molecular assays during the first six months of life.*

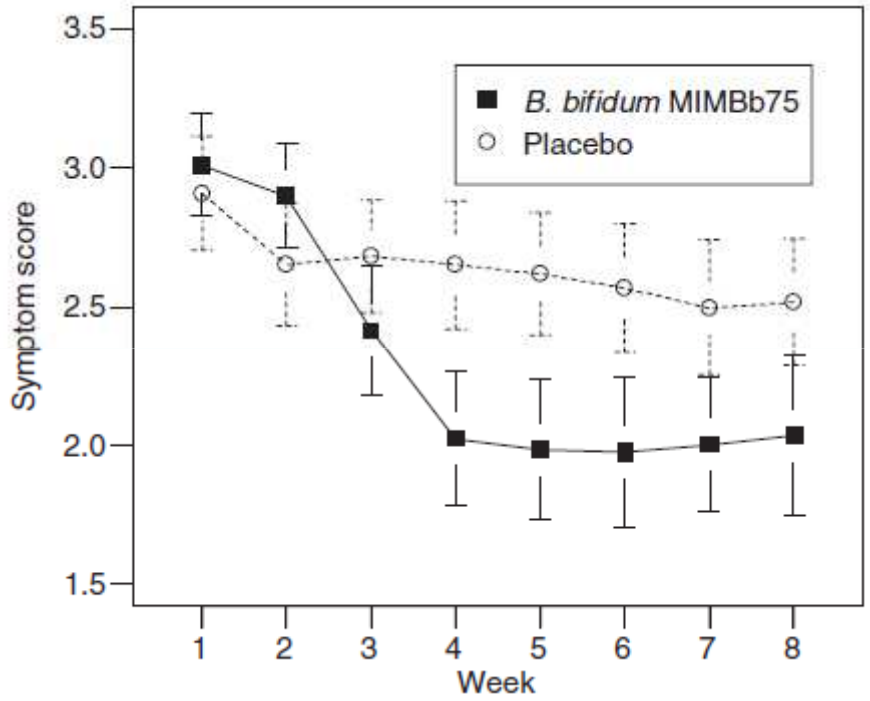
	7th week	12th week
Fussing*	65 (0–255)	31 (0–319)
Other cry†	30 (0–124)	15 (0–154)
Colic-type cry‡	6 (0–118)	2 (0–69)
Total distress	106 (0–478)	58 (0–448)

The amount (minutes/day; median with range) of fussing and crying, and total distress reported by parents during the 7th and 12th weeks of life

- *Bifidobacterium and Lactobacillus appear to protect against crying and fussing.*
- *Identification of specific strains with optimal protective properties would benefit at-risk infants.*

Partty A et al. PLoS One. 2012;7(3):e32495

Randomised clinical trial: *Bifidobacterium bifidum* MIMBb75 significantly alleviates irritable bowel syndrome and improves quality of life – a double-blind, placebo-controlled study



✓A total of 122 patients were randomised to receive either placebo (N = 62) or Bifidobacterium bifidum MIMBb75 (N = 60) once a day for 4 weeks

Comparison of effects of placebo and B. bifidum MIMBb75 on global IBS symptoms (by SGA, recorded on a 0–6 scale) on a weekly basis. Significant improvement of global IBS symptoms in the bifidobacteria group vs. placebo.

S. Guglielmetti et al. Aliment Pharmacol Ther 2011; 33: 1123–1132

DAL MICROBIOTA AL PROBIOTICO: *BIFIDOBATTERI NELLA PREVENZIONE E NEL TRATTAMENTO*

Conclusioni

- Bifidobatteri: probiotici promettenti, sia nella prevenzione che nel trattamento di molti disordini pediatrici:

  - ***Allergia***
  - ***Infezioni***
  - ***Disordini Funzionali Gastrointestinali***
  - ***…..MICI***
  - ***……ulteriori studi clinici sono necessari!!!!!***