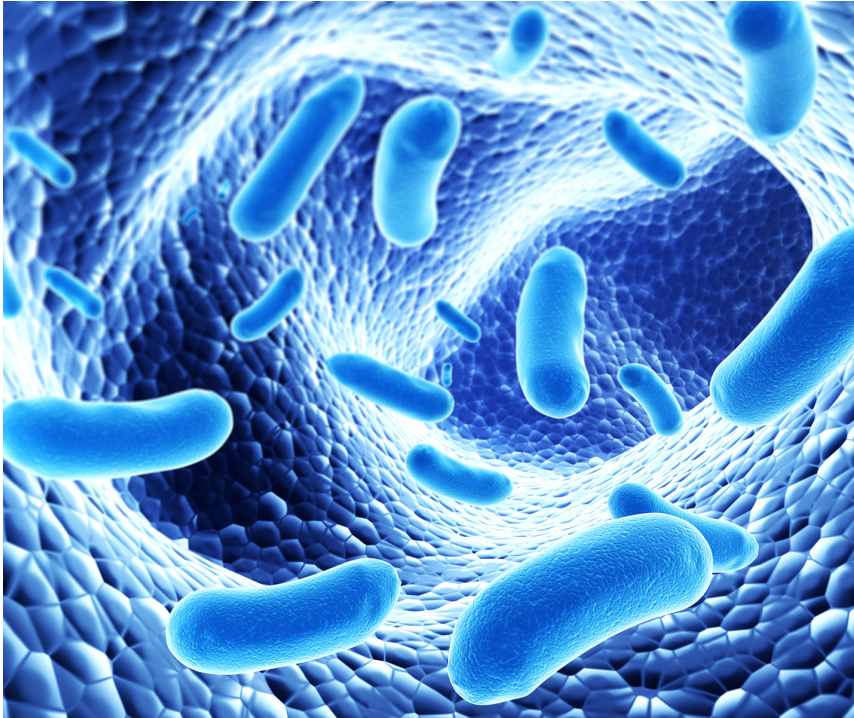


Obrigada por ver esta apresentação
Gostaríamos de recordar-lhe que esta
apresentação é propriedade do autor.

É-lhe fornecida pela Sociedade Portuguesa
de Pediatria no contexto do 19º
Congresso Nacional de Pediatria, para
seu uso pessoal, tal como submetido pelo
autor

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Probióticos na Prematuridade

Ana Rita Prior



**19º Congresso Nacional
de Pediatria**

**24 – 26 out 2018
Centro de Congressos
do Estoril**



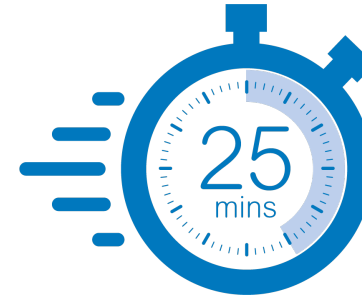
+ Probióticos na Prematuridade

- I. Introdução
- II. Microbiota no pré-termo
- III. Administração de probióticos:
 - a. Revisão bibliográfica
 - Interpretação complexa!
- IV. Questões e controvérsias
- V. E o futuro ?...



+ Probióticos na Prematuridade

- I. Introdução
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 - Interpretação complexa!
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- V. E o futuro ?...





Probióticos: definição



**Food and Agriculture Organization
of the United Nations**



World Health Organization

“Probiotics are life microorganisms, which when administered in adequate amounts confer a health benefit on the host (FAO2001) ¹

¹ Food and A Organization of the United Nations (FAO) , 2001. Health and Nutritional Properties of Probiotics in Food including Powder with with live Lactic Acid Bacteria.

http://www.who.int/foodsafety/publications/fs_management/en/probiotics.pdf





Probióticos: definição



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http://www.who.int/foodsafety/publications/fs_management/en/probiotics.pdf





Probióticos na Prematuridade: porquê?

- Aumento da mortalidade associada a NEC e sepsis
 - Ambas as patologias influenciadas pela microbiota intestinal?
- O microbiota intestinal é influenciável pelas práticas na UCIN
- Administração de Probióticos – uma “escolha”
 - Muitos dados – diferentes opiniões
- Nós (e o pais) queremos saber que estamos a fazer o melhor que podemos...



+ NEC e Sepsis: mortalidade

ORIGINAL ARTICLE

Causes and Timing of Death in Extremely Premature Infants from 2000 through 2011

CONCLUSIONS

We found that from 2000 through 2011, overall mortality declined among extremely premature infants. Deaths related to pulmonary causes, immaturity, infection, and central nervous system injury decreased, while necrotizing enterocolitis–related deaths increased. (Funded by the National Institutes of Health.)

N ENGL J MED 372;4 NEJM.ORG JANUARY 22, 2015



+ NEC e Sepsis: mortalidade

Table 2. Overall Causes of Death among Extremely Premature Infants, 2000–2011.*

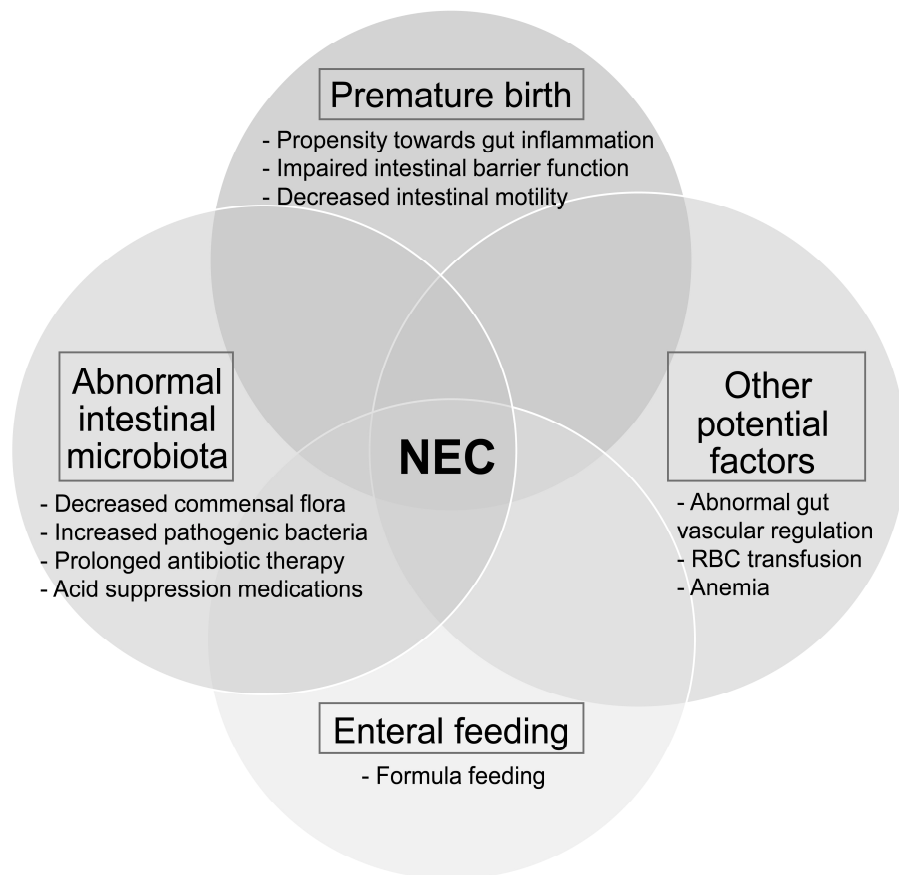
Variable	2000–2003	2004–2007	2008–2011	All Years	P Value
Total live births	7440	7684	7124	22,248	
Total deaths	2043	2193	1839	6075	
Overall mortality rate per 1000 live births (95% CI)†	275 (264–285)‡	285 (275–295)‡	258 (248–268)	273 (267–279)	0.003
Cause-specific mortality rate per 1000 live births (95% CI)†§					
Congenital anomalies	16 (13–19)	14 (12–17)	13 (10–16)	14 (13–16)	0.31
Respiratory distress syndrome	65 (60–71)‡	69 (63–75)‡	56 (51–62)	64 (60–67)	0.02
Bronchopulmonary dysplasia	18 (15–21)	15 (13–18)	12 (10–15)	15 (13–17)	0.09
Pulmonary: respiratory distress syndrome plus bronchopulmonary dysplasia	83 (77–90)‡	84 (78–90)‡	68 (63–74)	79 (75–82)	0.002
Infection	22 (19–25)	24 (21–28)	19 (16–22)	22 (20–24)	0.20
Necrotizing enterocolitis	23 (20–27)‡	29 (25–33)	30 (27–34)	28 (25–30)	0.04
CNS injury¶	7 (5–9)	11 (9–14)	10 (8–13)	9 (8–11)	0.21
Immaturity	86 (80–93)‡	81 (75–88)	81 (75–88)	83 (79–87)	0.04
Other	37 (33–42)	40 (36–45)	34 (30–39)	37 (35–40)	0.20

N ENGL J MED 372;4 NEJM.ORG JANUARY 22, 2015





NEC: Fatores de risco

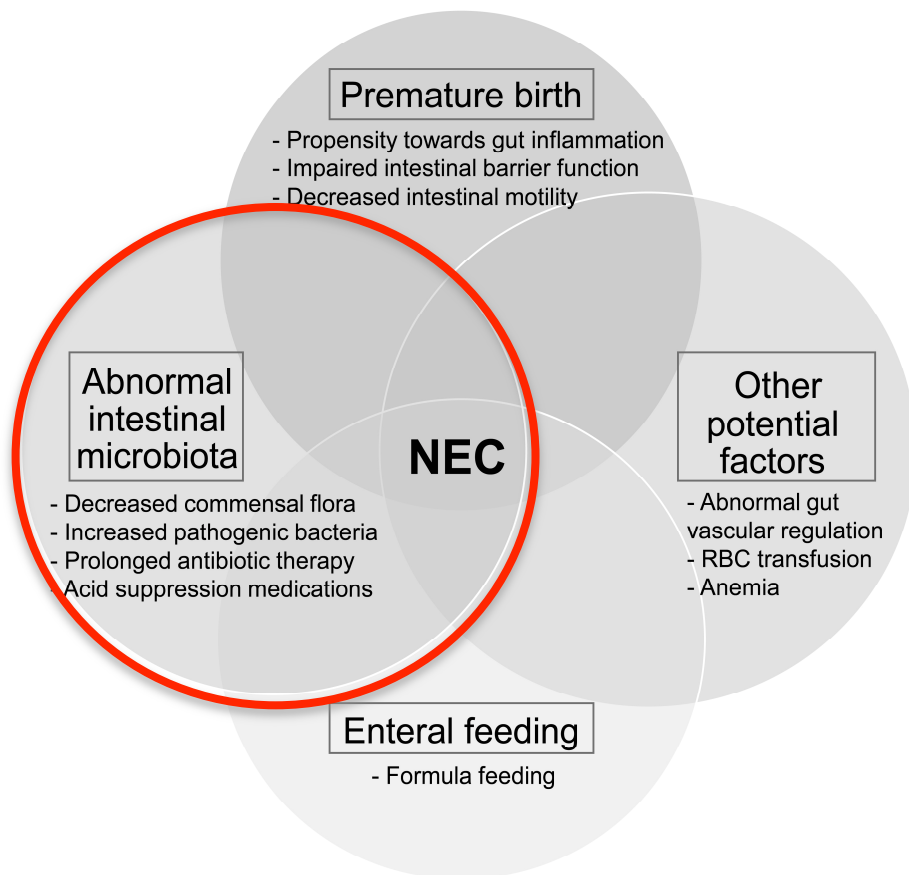


Petel RM and Denning PW
Pediatric Research 2015





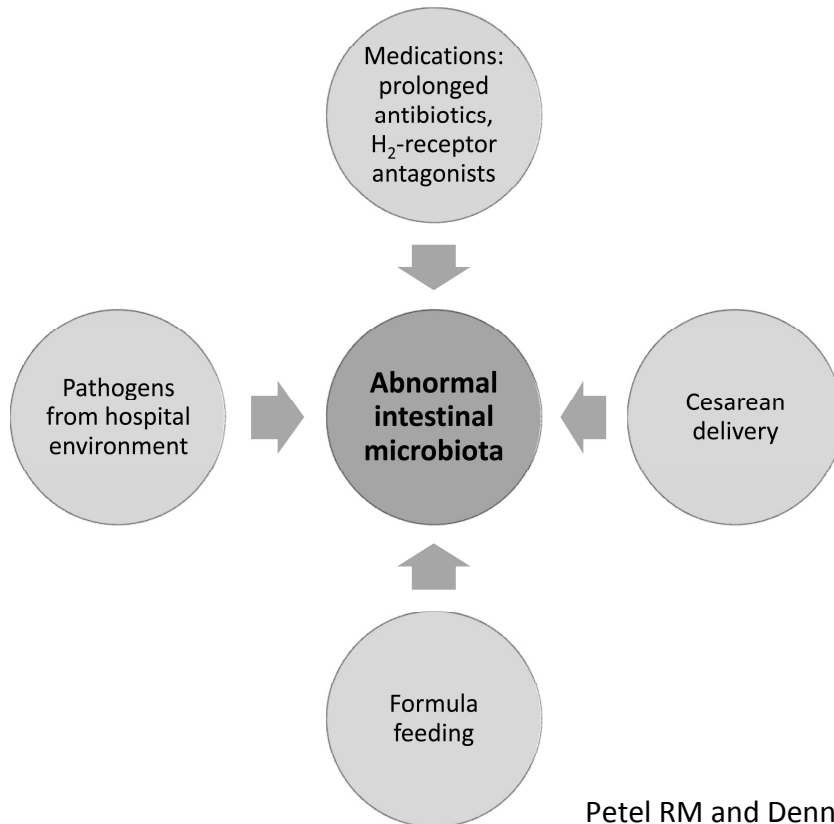
NEC: Fatores de risco



Petel RM and Denning PW
Pediatric Research 2015



+ NEC: microbiota intestinal

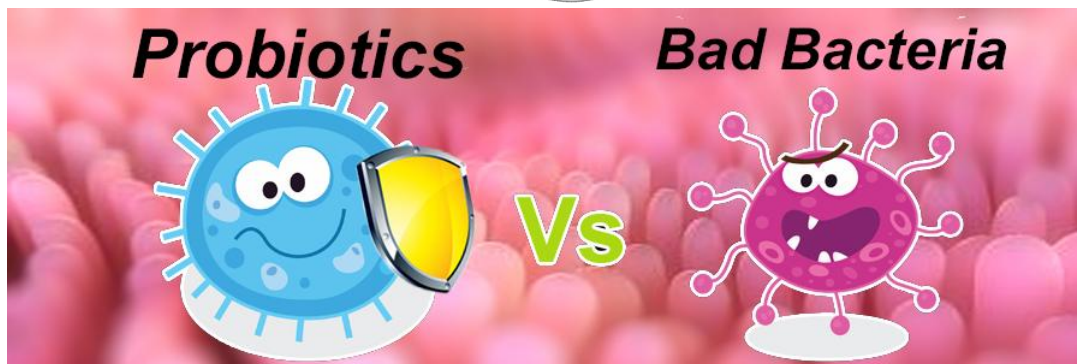


Petel RM and Denning PW
Pediatric Research 2015



+ NEC: microbiota intestinal

Medications:
prolonged
antibiotics,
H₂-receptor
antagonists

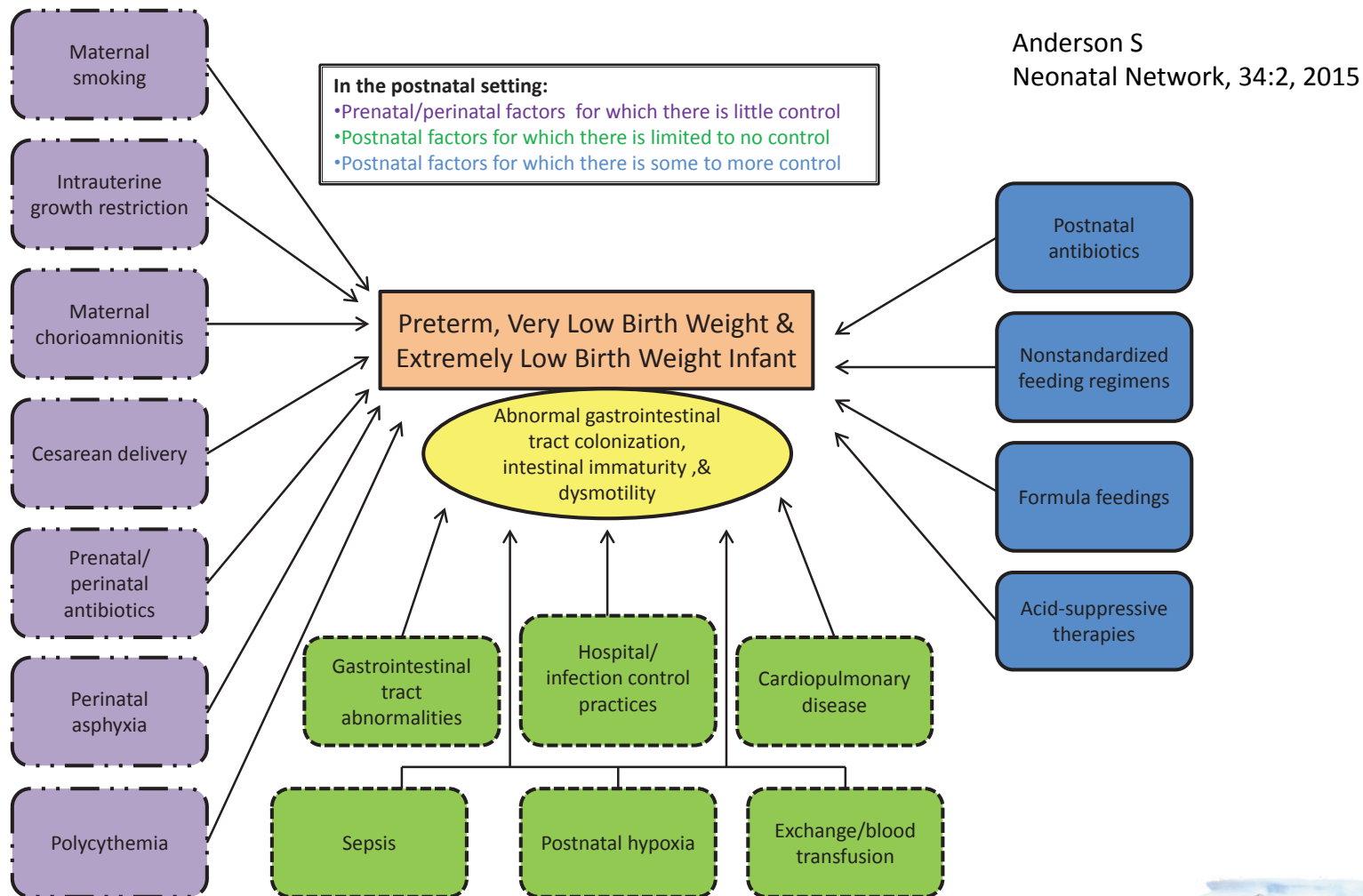


Formula
feeding

Petel RM and Denning PW
Pediatric Research 2015

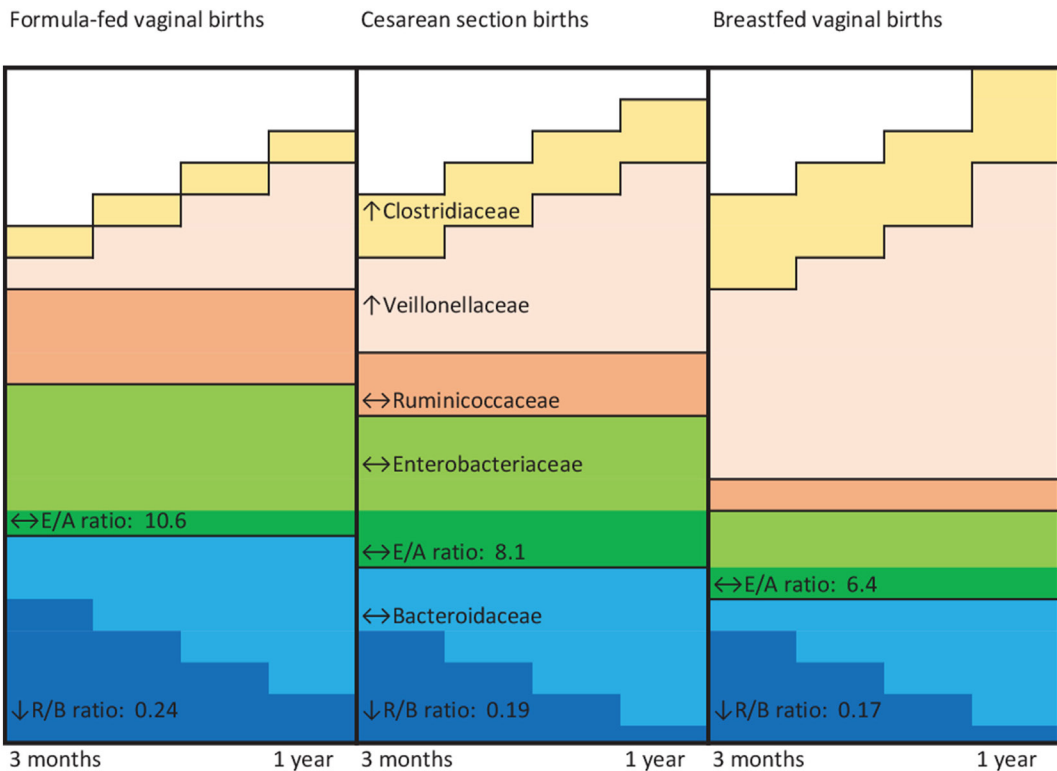


Microbiota intestinal: Termo vs pretermo





Microbiota intestinal: Parto e leite materno



Yasmin et al
Front Pediatr 5:200, 2017

↔ (unchanged)

E/A: Enterobacteriaceae/Alicigenaceae ratio

R/B: Rikenellaceae/Bacteroidaceae ratio

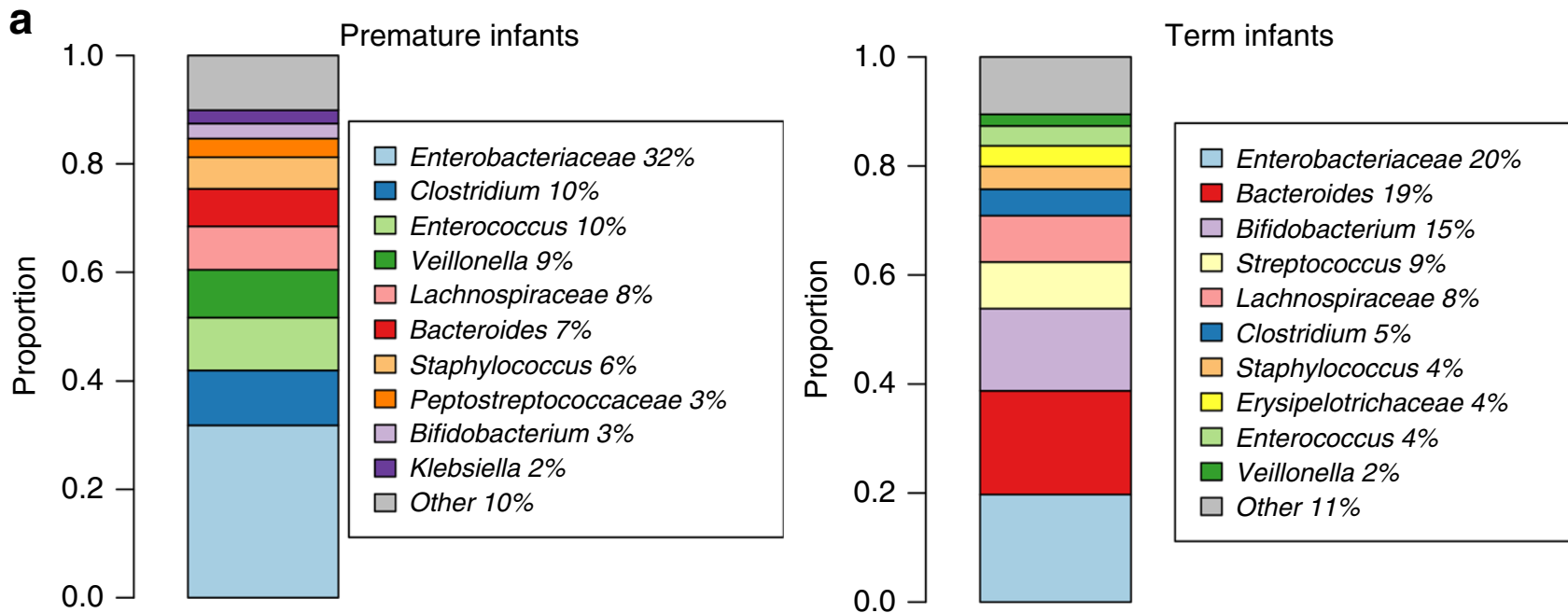
Schematic representation of microbial family changes by perinatal group.





Microbiota intestinal: Termo vs pretermo

Schematic representation of microbial family changes by perinatal group.



The premature infant gut microbiome during the first 6 weeks...

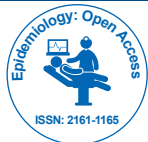
DA Chernikova et al.

Pediatric Research (2018) 84:71 – 79





Microbiota intestinal: o papel dos probióticos



Epidemiology: Open Access

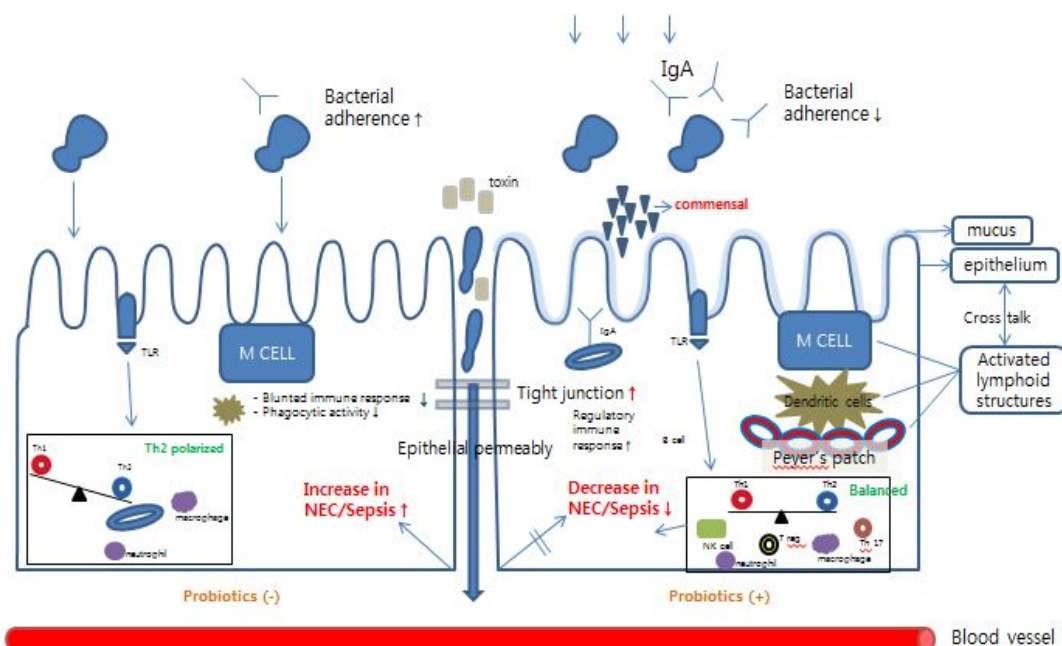
Lee, Epidemiology (Sunnyvale) 2018, 8:1
DOI: 10.4172/2161-1165.1000339

Review

Open Access

An Update on the Gut Microbiome and the Use of Probiotics for Disease Prevention in Preterm Infants

Hyun Ju Lee*



Mechanism of probiotics and the gastrointestinal immune system in newborns.





Probióticos

- Múltiplos RCT, metanálises e estudos observacionais (>6500)
- Objectivo principal (benefício para o hospedeiro)
 - NEC, sepsis tardia, mortalidade
 - Tolerância alimentar, tempo para AEE, crescimento, ...
- Compreensão mecanicistica limitada

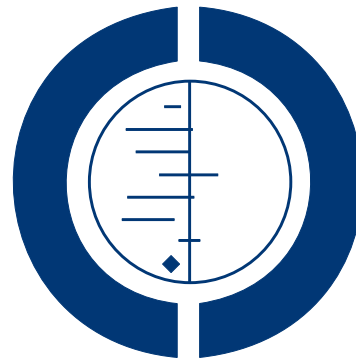




Probióticos na prevenção de NEC no pretermo

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

AlFaleh K, Anabrees J



**THE COCHRANE
COLLABORATION®**

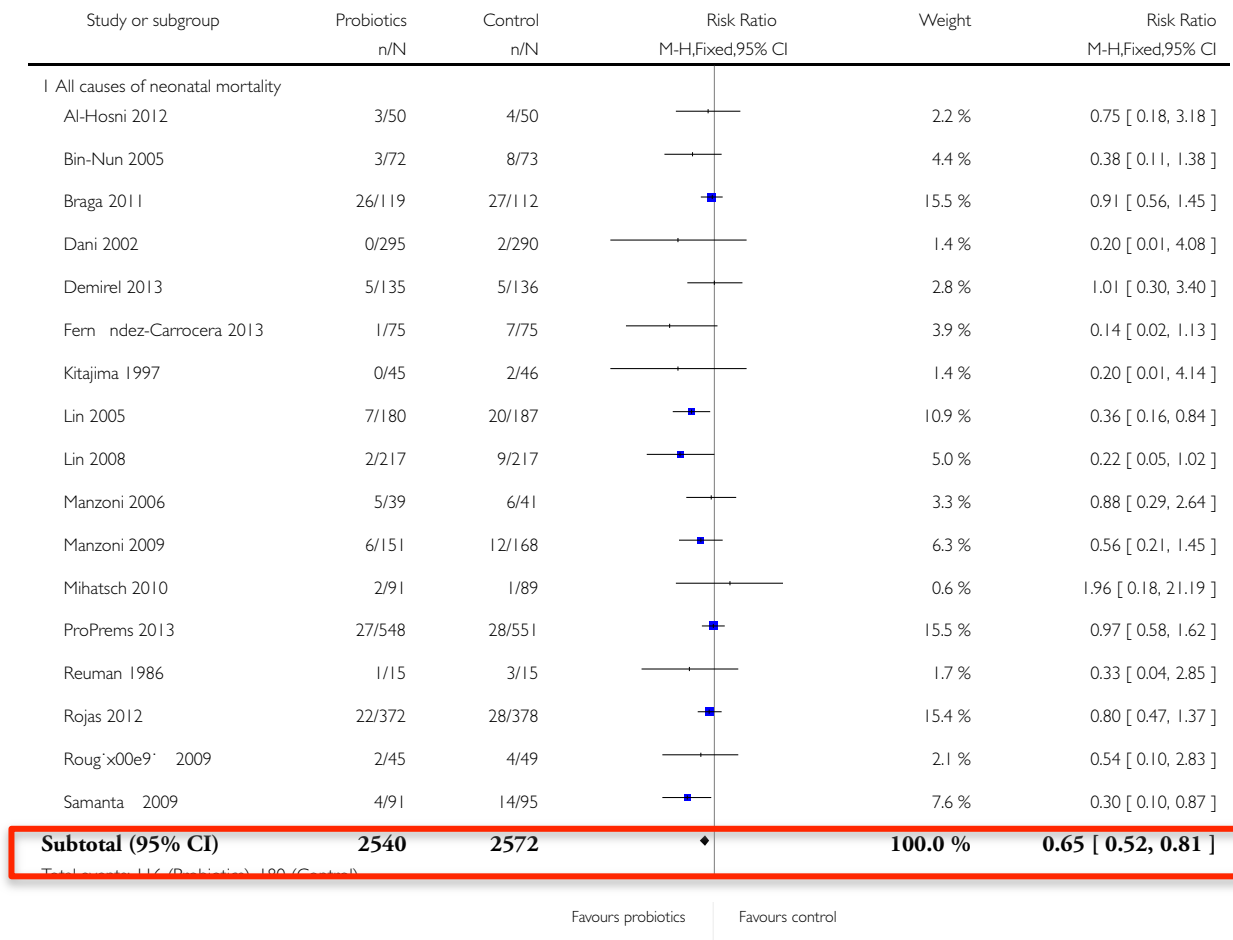
Copyright © 2014 The Cochrane Collaboration. Published by John Wiley & Sons, Ltd.



Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

MORTALIDADE (todas as causas) (n= 5112)

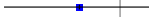
Mortalidade
RR 0,65 (0,52 – 0,81)



Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

MORTALIDADE associada a NEC (n=2755)

2 NEC related mortality

Bin-Nun 2005	0/72	3/73		14.2 %	0.14 [0.01, 2.75]
Dani 2002	0/295	2/290		10.3 %	0.20 [0.01, 4.08]
Kitajima 1997	0/45	2/46		10.1 %	0.20 [0.01, 4.14]
Lin 2008	2/217	3/217		12.3 %	0.67 [0.11, 3.95]
Mihatsch 2010	1/91	0/89		2.1 %	2.93 [0.12, 71.10]
ProPrem 2013	4/548	11/551		44.9 %	0.37 [0.12, 1.14]
Sari 2010	0/110	1/111		6.1 %	0.34 [0.01, 8.17]

Subtotal (95% CI)	1378	1377		100.0 %	0.39 [0.18, 0.82]
--------------------------	-------------	-------------	---	----------------	----------------------------

Total events: 7 (Probiotics), 22 (Control)

Heterogeneity: $\text{Chi}^2 = 2.72$, $\text{df} = 6$ ($P = 0.84$); $I^2 = 0.0\%$

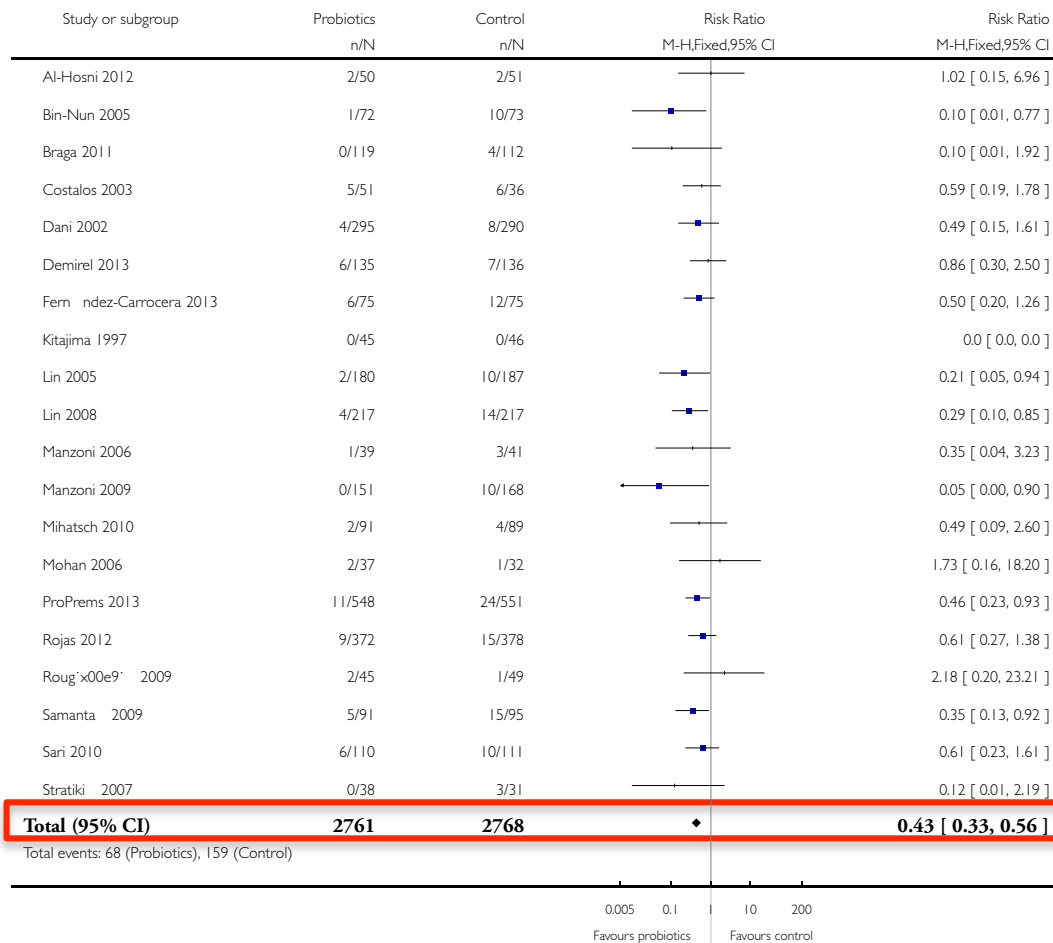
Test for overall effect: $Z = 2.48$ ($P = 0.013$)

0.01 0.1 10 100
Favours probiotics Favours control

Mortalidade associada NEC
RR 0,39 (0,18 – 0,82)

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

NEC grave (II-III, segundo Classificação de Bell) (n= 5529)



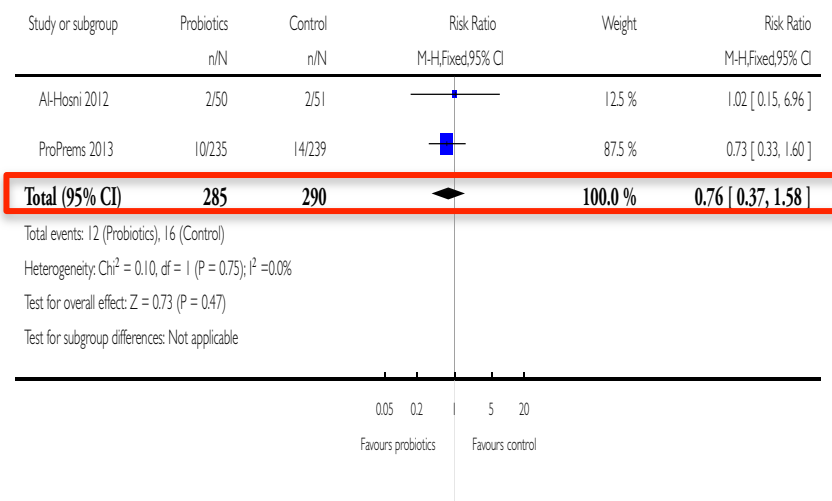
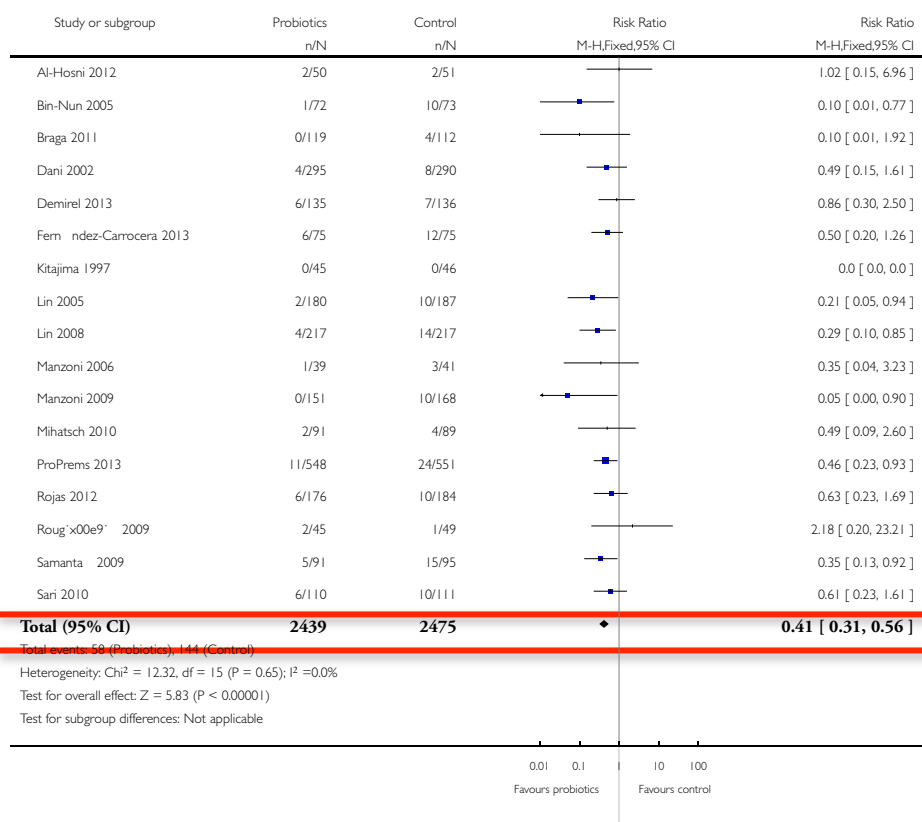
NEC II-III
RR 0,43 (0,33 – 0,56)

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

NEC grade II/III (<1500g versus <1000g)

< 1500 g

< 1000 g



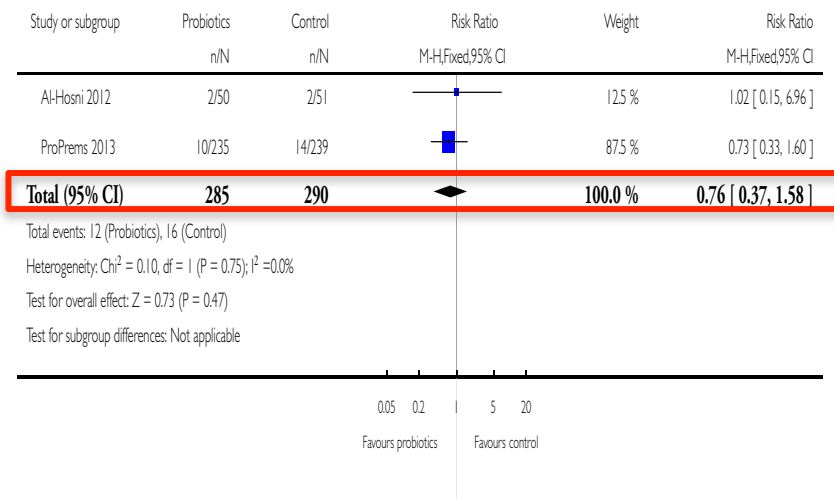
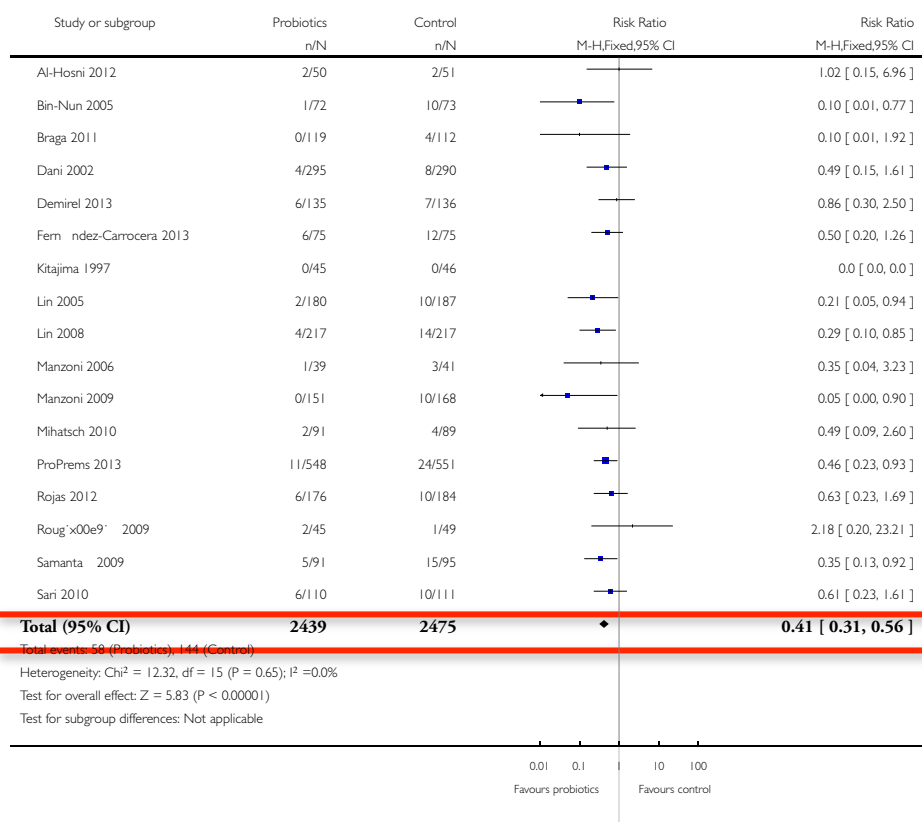
	RR
<1500 g	0,41 (0,31 – 0,56)
<1000 g	0,76 (0,37-1,58)

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

NEC grade II/III (<1500g versus <1000g)

< 1500 g

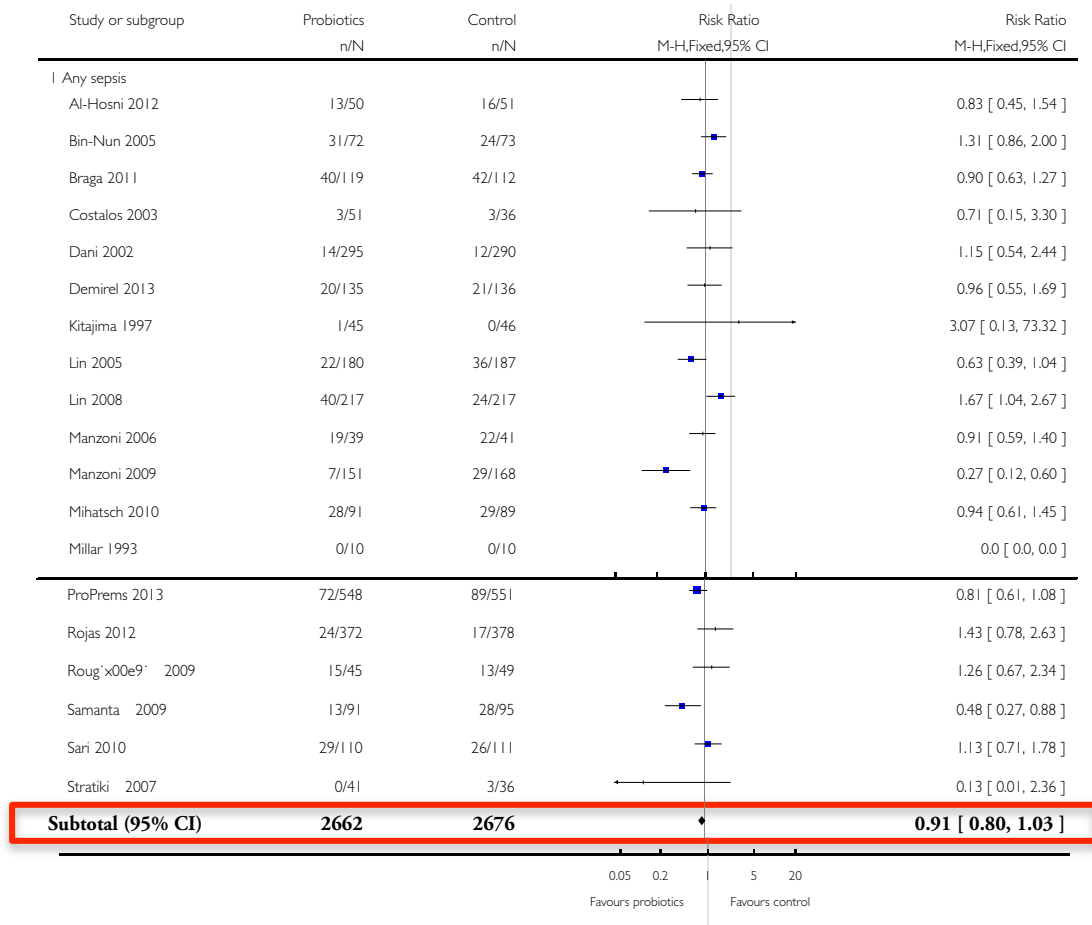
< 1000 g



	RR	N
<1500 g	0,41 (0,31 – 0,56)	4914
<1000 g	0,76 (0,37-1,58)	575

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

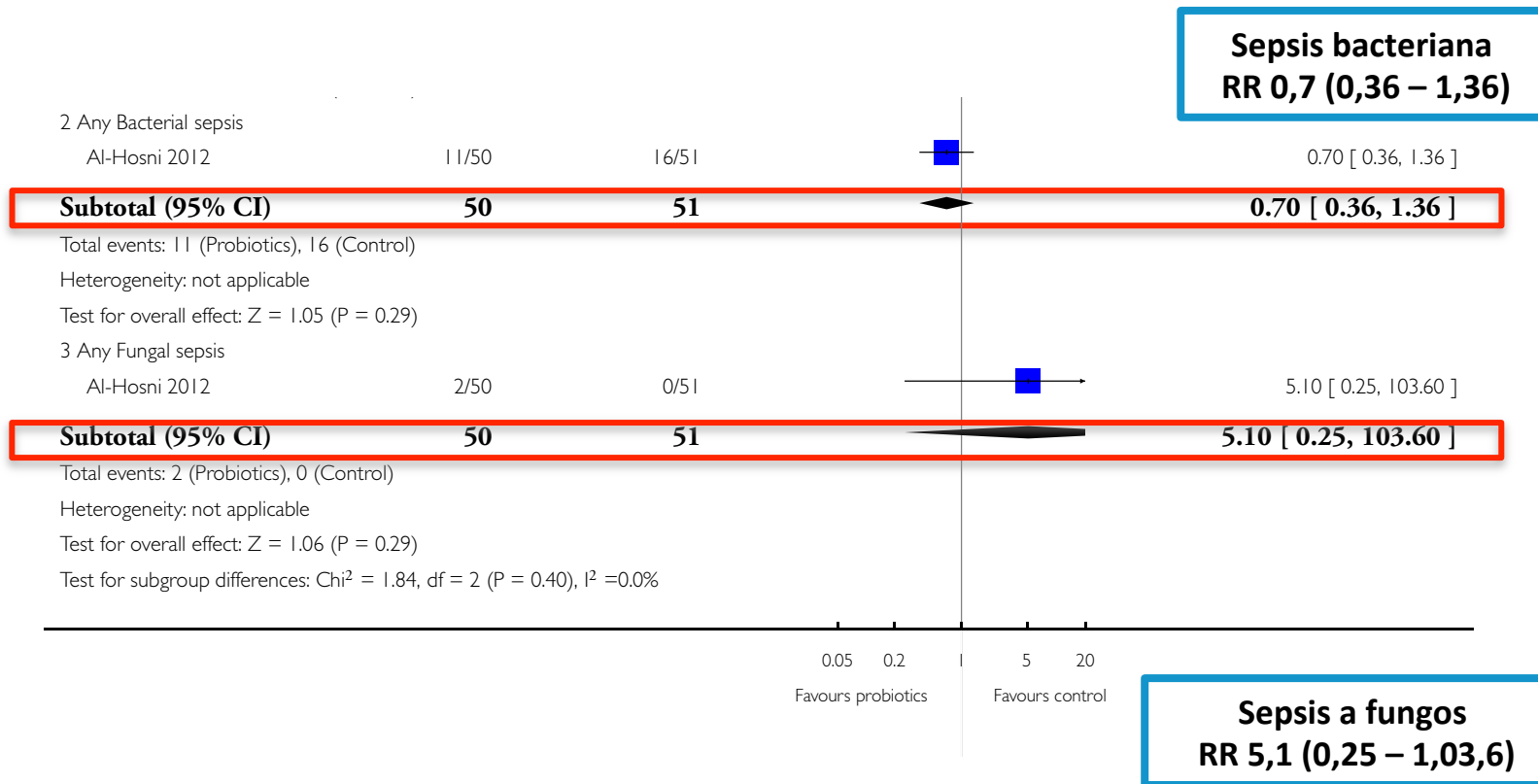
Sepsis (c/ hemocultura positiva)(n= 5338)



Sepsis
RR 0,91 (0,80 – 1,03)

Probiotics for prevention of necrotizing enterocolitis in preterm infants (Review) 2014

Sepsis (c/ hemocultura positiva)



AUTHORS' CONCLUSIONS

Implications for practice

Enteral supplementation of probiotics prevents severe NEC and all cause mortality in preterm infants. Our review strongly supports a change in practice and adoption of probiotics prophylaxis in the management of preterm infants.

Implications for research

More studies are needed to investigate the most effective formulation and dose to be utilized. Parents of preterm infants should be informed of current evidence if further placebo controlled randomized trials are to be conducted.

+ Probióticos e sepsis tardia

Probiotics for Preventing Late-Onset Sepsis in Preterm Neonates

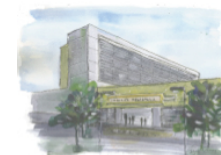
Medicine 95 (8), e2581, 2016

A PRISMA-Compliant Systematic Review and Meta-Analysis of Randomized Controlled Trials

N= 6104

Guo-Qiang Zhang, MSc, Hua-Jian Hu, MD, Chuan-Yang Liu, MSc, Shristi Shakya, MD, and Zhong-Yue Li, MD

Subgroup	Number of Studies	RR (95% CI)
Any sepsis	25	0.83 (0.73, 0.94)
Any bacterial sepsis	11	0.82 (0.71, 0.95)
Any fungal sepsis	6	0.57 (0.41, 0.78)
Birth weight, g		
<2500	25	0.83 (0.73, 0.94)
<1500	19	0.86 (0.75, 0.97)
<1000	3	0.73 (0.45, 1.19)
Probiotic organism		
<i>Lactobacillus</i> species	6	0.72 (0.50, 1.03)
<i>Bifidobacterium</i> species	6	0.78 (0.48, 1.25)
<i>Saccharomyces boulardii</i>	3	0.84 (0.58, 1.22)
Mixture	10	0.85 (0.73, 1.00)





Podemos então parar por aqui ...

- Suporte científico
- Diminuição da mortalidade
- Diminuição da mortalidade associada a NEC
- Diminuição da sepsis tardia





...mas...

- Assimetria dos estudos
- < 1000 g – poucos dados!
- Não inclusão do estudo PIPs (2015)
- Segurança: risco de sepsis a probiótico
- Implicações a longo prazo
- Qual o melhor probiótico? Qual a melhor dose? Qual o melhor “timing”?



+ Probióticos e Prematuridade

Received: 23 September 2018; Accepted: 8 October 2018; Published: 10 October 2018



nutrients



Review

Filling the Gaps: Current Research Directions for a Rational Use of Probiotics in Preterm Infants

Arianna Aceti ^{1,*}, Isadora Beghetti ^{1,†}, Luca Maggio ², Silvia Martini ¹, Giacomo Faldella ¹ and Luigi Corvaglia ¹

- ¹ Neonatal Intensive Care Unit, AOU Bologna, Department of Medical and Surgical Sciences (DIMEC), University of Bologna, 40138 Bologna, Italy; i.beghetti@gmail.com (I.B.); silvia.martini4@gmail.com (S.M.); giacomo.faldella@unibo.it (G.F.); luigi.corvaglia@unibo.it (L.C.)
- ² Department of Woman and Child Health, Obstetric and Neonatology Area, Fondazione Policlinico Universitario A. Gemelli IRCCS, 00168 Rome, Italy; luca.maggio@fastwebnet.it

“ However, despite the enthusiasm towards these products (probiotics) in neonatal medicine, theoretical knowledge and clinical applications still need to be improved.”



+ Probióticos e NEC: revisão sistemática e metanálise (2015)

Aceti et al. *Italian Journal of Pediatrics* (2015) 41:89
DOI 10.1186/s13052-015-0199-2



REVIEW

Open Access

N = 6605



Probiotics for prevention of necrotizing enterocolitis in preterm infants: systematic review and meta-analysis

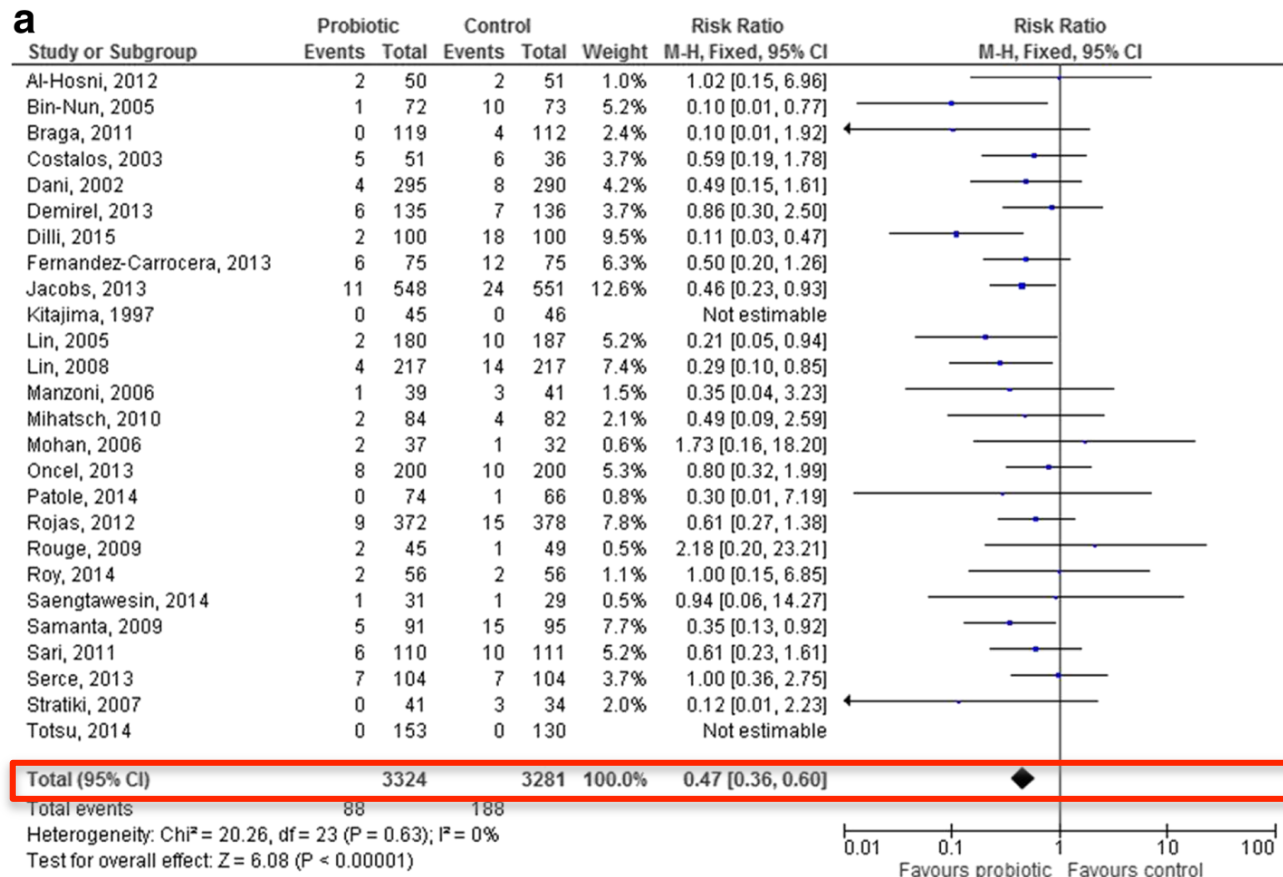
Arianna Aceti^{1*}, Davide Gori², Giovanni Barone³, Maria Luisa Callegari⁴, Antonio Di Mauro⁵, Maria Pia Fantini², Flavia Indrio⁵, Luca Maggio³, Fabio Meneghin⁶, Lorenzo Morelli⁴, Gianvincenzo Zuccotti⁶, Luigi Corvaglia¹ and on behalf of the Italian Society of Neonatology

Probiotics prevented NEC in preterm infants (RR 0.47 [95 % CI 0.36–0.60], $p < 0.00001$). Strain-specific sub-meta-analyses showed a significant effect for *Bifidobacteria* (RR 0.24 [95 % CI 0.10–0.54], $p = 0.0006$) and for probiotic mixtures (RR 0.39 [95 % CI 0.27–0.56], $p < 0.00001$). Probiotics prevented NEC in very-low-birth-weight infants (RR 0.48 [95 % CI 0.37–0.62], $p < 0.00001$); there were insufficient data for extremely-low-birth-weight infants. The majority of studies presented severe or moderate microbiological flaws.

Probiotics had an overall preventive effect on NEC in preterm infants. However, there are still insufficient data on the specific probiotic strain to be used and on the effect of probiotics in high-risk populations such as extremely-low-birth-weight infants, before a widespread use of these products can be recommended.



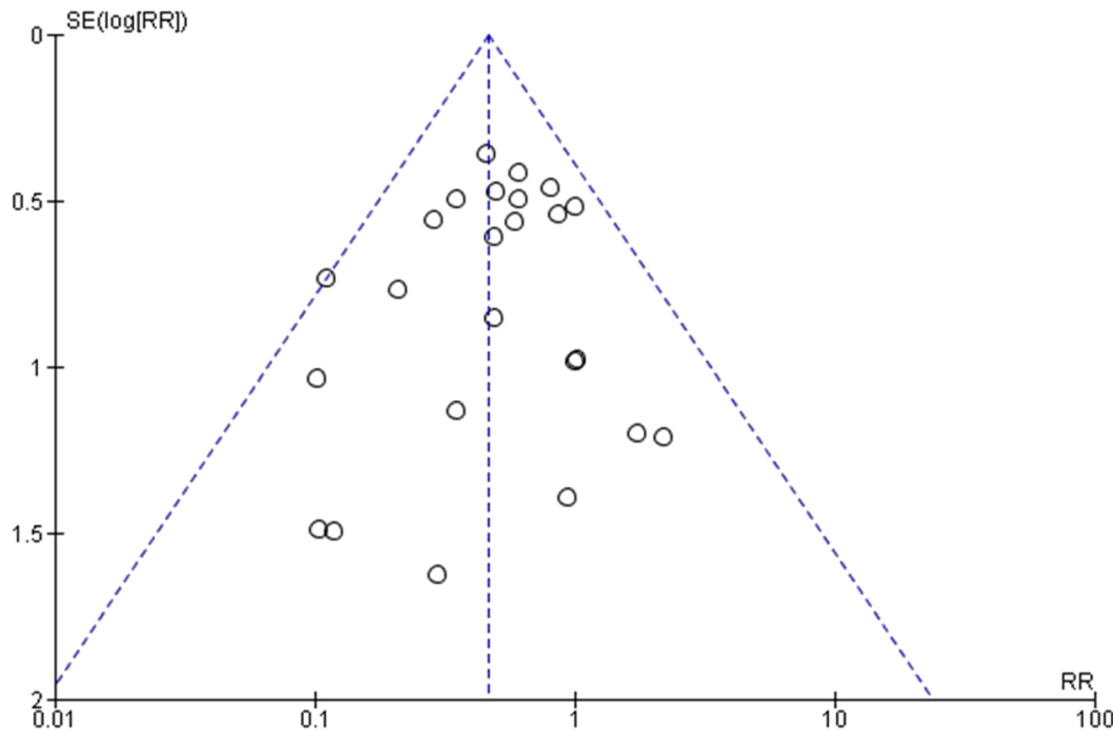
+ Probióticos e NEC: revisão sistemática e metanálise (Aceti 2015)



N = 6605



+ Probióticos e NEC: revisão sistemática e metanálise (Aceti 2015)



N = 6605

(5912 < 1500g; (25-50 % < 1000 g)

Sem assimetrias

Thus, the proposal made by the authors of the recent Cochrane review of a “change in practice” in the use of probiotics in preterm infants [11] might require further investigation

+ PIPs

Costeloe K, Bowler U, Brocklehurst P, Hardy P, Heal P, Juszczak E, *et al.* A randomised controlled trial of the probiotic *Bifidobacterium breve* BBG-001 in preterm babies to prevent sepsis, necrotising enterocolitis and death: the Probiotics in Preterm infantS (PiPS) trial. *Health Technol Assess* 2016;**20**(66).

Objective: To test the use of the probiotic *Bifidobacterium breve* strain BBG-001 to prevent NEC, late-onset sepsis and death in preterm babies while monitoring probiotic colonisation of participants.

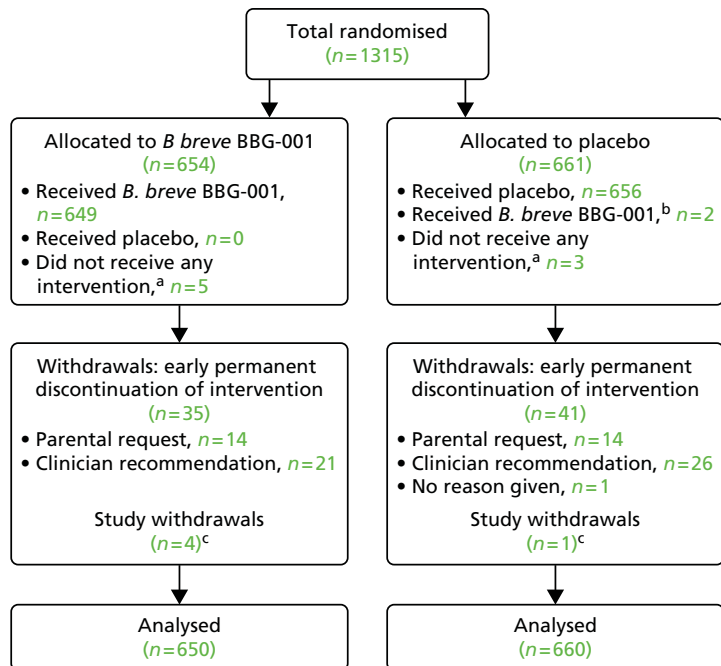
Design: Double-blind, randomised, placebo-controlled trial.

Participants: Babies born between 23 and 30 weeks' gestation and randomised within 48 hours of birth. Exclusions included life-threatening or any gastrointestinal malformation detected within 48 hours of birth and no realistic chance of survival.

Interventions: Active intervention: 1 ml of *B. breve* BBG-001 in one-eighth-strength infant formula Neocate® (Nutricia Ltd, Trowbridge, UK), (6.7×10^7 to 6.7×10^9 colony-forming units) per dose administered enterally. Placebo: 1 ml of one-eighth-strength infant formula Neocate. Started as soon as practicable and continued daily until 36 weeks' postmenstrual age.



+ PIPs



	<i>Bifidobacterium breve</i> BBG-001 probiotic (n=650)	Placebo (n=660)	Adjusted* risk ratio (95% CI)
Necrotising enterocolitis†	61 (9%)	66 (10%)	0.93 (0.68–1.27)
Sepsis‡	73 (11%)	77 (12%)	0.97 (0.73–1.29)
Death before discharge home§	54 (8%)	56 (9%)	0.93 (0.67–1.30)

Data are n (%), unless otherwise indicated. *Adjusted for sex, gestational age at birth, and randomisation within 24 h of birth. Adjustment by centre was excluded because the model did not converge. Allowances for correlations between multiple births are accounted for. †Necrotising enterocolitis (Bell stage 2 or 3).^{9,10} ‡Sepsis is defined as bloodstream infection with non-skin commensals after 72 h postnatal age and before 46 weeks' postmenstrual age. §Includes three infants who remained on paediatric wards at the time of analysis and are included as survivors; all were later discharged home.

Table 2: Primary outcomes

Limitations: Cross-colonisation of the placebo arm could have reduced statistical power and confounded results; analyses suggest that this did not happen.

Conclusions: This is the largest trial to date of a probiotic intervention. It shows no evidence of benefit and does not support routine use of probiotics for preterm infants.



ProPREMS versus PIPs

ProPREMS

Australia, n= 1099

Mais leite materno
Menor incidência de NEC

Mistura de probióticos

Resultado principal:
Sepsis – sem efeito

Resultado secundário
NEC – com redução significativa

PIPS

Reino Unido, n= 1310

Menos Leite materno
Maior incidência de NEC

Um único probiótico

Resultado principal:
NEC, sepsis morte – sem efeito



+ Probióticos: segurança

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 22, No. 9, September 2016

***Bifidobacterium longum* Subspecies *infantis* Bacteremia in 3 Extremely Preterm Infants Receiving Probiotics**

**Eirin Esaiassen, Pauline Cavanagh, Erik Hjerde,
Gunnar S. Simonsen, Ragnhild Støen,
Claus Klingenberg**

Author affiliations: University Hospital of North Norway, Tromsø, Norway (E. Esaiassen, P. Cavanagh, G.S. Simonsen, C. Klingenberg); Arctic University of Norway, Tromsø (E. Esaiassen, E. Hjerde, G.S. Simonsen, C. Klingenberg); St. Olavs Hospital, Trondheim, Norway (R. Støen); Norwegian University of Science and Technology, Trondheim (R. Støen)

DOI: <http://dx.doi.org/10.3201/eid2209.160033>

Table. Characteristics of 3 extremely preterm infants with *Bifidobacterium longum* subspecies *infantis* bacteremia, 2015*

Characteristic	Patient 1	Patient 2	Patient 3
NICU	A	B	A
Sex	M	M	F
Date of onset	Apr	Jul	Sep
Gestational age, wk	24	23	24
Birth weight, g	730	500	697
Mode of delivery	Vaginal	Vaginal	Caesarean section
Apgar score at 1, 5, and 10 min after birth	4, 5, 5	Unknown, 0, 4	2, 2, 3
Reason for prematurity	Preterm rupture of membranes, maternal infection	Sudden preterm rupture of membranes, delivery not attended by healthcare personnel	Placental abruption
Age at onset of sepsis, d	8	12	46
Maximum CRP level, mg/L, ≤ 48 h of symptom onset	147	25	242
Age at discharge, wk	40	41	43
Weight at discharge, kg	3.3	3.4	3.3
Bacterial culture medium and conditions	BacT/ALERT, † aerobic, 36°C	BACTEC Plus, † aerobic, 35°C	BacT/ALERT, † aerobic, 36°C
Bacterial growth in blood culture, d	2	3	2

*Patients were given ½ to 1 capsule/day of oral probiotics (Infloran; Laboratório Farmacêutico **Specialità Igienico Terapeutiche**, Mede, Italy) that contained 10^9 *Lactobacillus acidophilus* (ATCC 4356) and 10^9 *B. longum* subspecies *infantis* (ATCC 15697). MICs (mg/L) for antimicrobial drugs tested were 0.016 for meropenem, 0.032 for ampicillin, 0.064 for penicillin, 0.064 for piperacillin/tazobactam, 0.250 for cefotaxime, 0.250 for clindamycin, 0.250 for vancomycin, and 4.000 for ciprofloxacin. All bacterial strains were inherently resistant to aminoglycosides. ATCC, American Type Culture Collection (Manassas, VA, USA); CRP, C-reactive protein; NICU, neonatal intensive care unit. †bioMérieux (Marcy l'Étoile, France).



+ Probióticos: segurança

- Sepsis a probióticos
 - Descritos case reports; não descrito em nenhum RCT,...
 - Não reportada em nenhuma metanálise como efeito adverso
 - O risco existe...
- Transmissão de resistência aos antibióticos ?
- Suplementos alimentares versus medicamento- verificação de qualidade e segurança



+ Qual o melhor probiótico? Qual a melhor dose? Qual o melhor “timing”?

- O que é o “melhor”?
 - Diferentes resultados /objectivos
 - Qual ? (NEC, sepsis, alimentação entérica exclusiva, ...)
- Outros factores associados
 - Gravidez, UCINs,
 - Leite materno
 - Política de antibióticos
 - Outras práticas UCIN,...





Commentary

Para-probiotics for Preterm Neonates—The Next Frontier

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+ Comentários finais

■ Prevenção NEC

“ Correct probiotic given to “right” babies at “right” time may help, but...”

- Probióticos?? – necessário mais estudos!
 - Maximizar a utilização de leite materno
 - Minimizar a utilização/duração dos antibióticos
 - Estabelecimento de protocolos de nutrição
 - Conhecimento da situação local
- Incentivar a investigação:



+ Probióticos na prematuridade



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+ Bibliografia

- M. Hosny et al. *Human Microbiome Journal* 2017;4:14–19
- Costeloe et al. *J Ped* 2018;1:1-6
- Battersby C, et al. *Arch Dis Child Fetal Neonatal Ed* 2018;0:F1–F8. doi:10.1136/archdischild-2017-313880
- Cavallaro G, et al. *Scientific Reports* 2017. 7: 13014 | DOI:10.1038/s41598-017-13465-2
- Olsen R, et al. *Neonatology* 2016;109:105–112
- Aceti et al. *Italian Journal of Pediatrics* 2015;41:89
- AlFaleh K, Anabrees J, Bassler D, Al-Kharfi T. Probiotics for prevention of necrotizing enterocolitis in preterm infants. *Cochrane Database of Systematic Reviews* 2011, Issue 3. Art. No.: CD005496. DOI: 10.1002/14651858.CD005496.pub3.
- Lee, *Epidemiology (Sunnyvale)* 2018, 8:1. DOI: 10.4172/2161-1165.1000339
- Sun, et al. *Adv Nutr* 2017;8:749–63; doi: <https://doi.org/10.3945/an.116.014605>.
- Anderson, S. Probiotics for Preterm Infants: A Premature or Overdue Necrotizing Enterocolitis Prevention Strategy? *NEONATAL NETWORK* 2015; 34(2) 83-101
- Deshpande G, et al. *BMJ Open* 2017;7:e017638. doi:10.1136/bmjopen-2017-017638
- Martin A, et al. *BMJ Open* 2018;8:e023044. doi:10.1136/bmjopen-2018-023044
- Jacobs SE, et al. *BMJ Paediatrics Open* 2017;1:e000176. doi:10.1136/bmjpo-2017-000176
- Totsu S, et al. *BMJ Paediatrics Open* 2018;2:e000256. doi:10.1136/bmjpo-2018-000256
- AlFaleh K, Anabrees J. Probiotics for prevention of necrotizing enterocolitis in preterm infants. *Cochrane Database of Systematic Reviews* 2014, Issue 4. Art. No.: CD005496. DOI: 10.1002/14651858.CD005496.pub4.
- Gengaimuthu K. The Cross Contamination (Cross Colonization) Phenomenon of Probiotic Use in Neonatal Intensive Care Units: Putative Mechanisms and Clinical and Research Implications. *Cureus* 10(5): e2691. DOI 10.7759/cureus.2691
- Dermyshe E; et al. The “Golden Age” of Probiotics: A Systematic Review and Meta-Analysis of Randomized and Observational Studies in Preterm Infants. *Neonatology* 2017;112:9–23
- Garland et al. The ProPrems trial: investigating the effects of probiotics on late onset sepsis in very preterm infants. *BMC Infectious Diseases* 2011, 11:210
- Patel R, et al. Causes and Timing of Death in Extremely Premature Infants from 2000 through 2011. *N Engl J Med* 2015;372:331-40.

