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Dr. Sampath is a physician-scientist and Neonatology. He loves clinical care, mentoring/education and is big-time into research. Outside work he likes the outdoors, philosophy, and music.

Feeding strategies to prevent NEC - The current and future

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No financial or other conflicts.
Personal Biases are disclosed

Children's Mercy KANSAS CITY

UMKC School of Medicine

BEST CHILDREN'S HOSPITALS U.S.News 2017-18

MAGNET RECOGNIZED AMERICAN NURSES CREDENTIALING CENTER

Disclosure

No financial or other conflicts to disclose.

Recommendations that are not evidence-based are disclosed.

Will limit mouse data to minimum.

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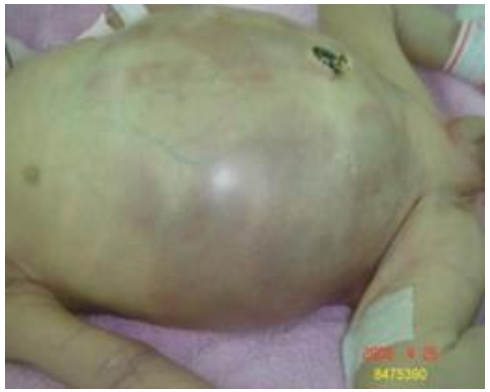
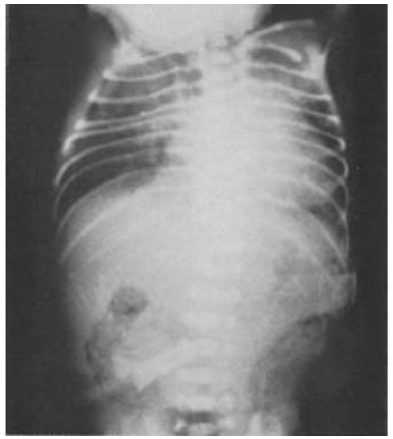
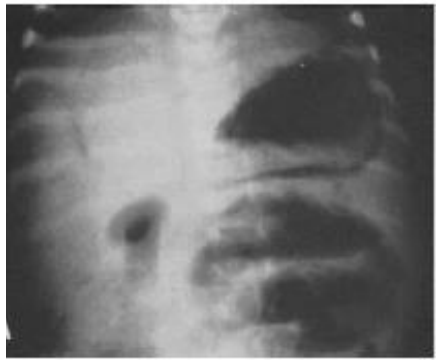
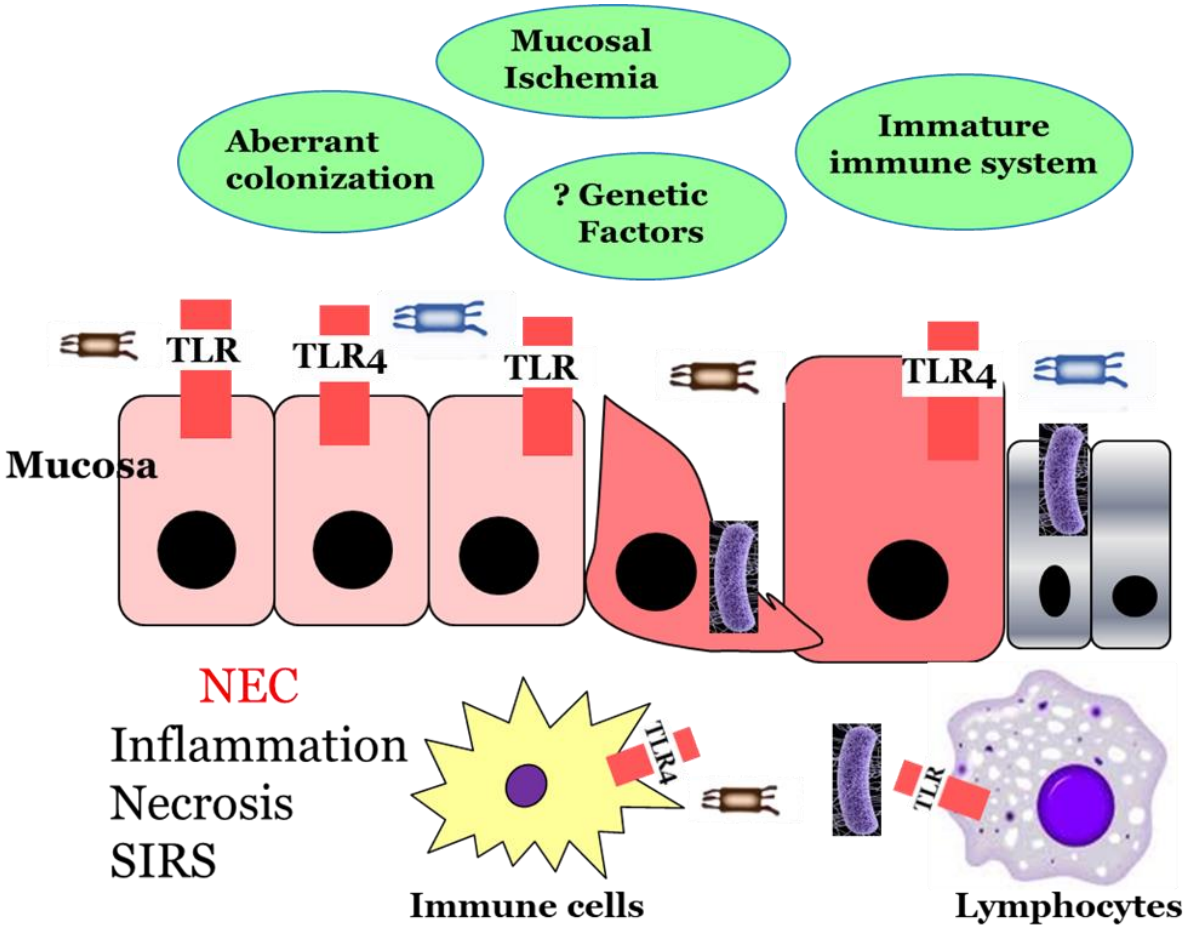
NEC pathogenesis - Current Understanding







Berdon et al. Radiology
1964 PMID: 14229131



NEC - Feeding practices (Breast milk protects)

Lucas Cole Lancet 1990; 336:1519-23.

TABLE III—NECROTISING ENTEROCOLITIS BY FEED GROUP

—	n	No (%) of cases	
		All cases	Confirmed cases
Formula only	236	24 (10·2%)	17 (7·2%)
Formula plus mother's milk	437	16 (3·7%)	11 (2·5%)
Human milk only	253	11 (4·3%)	3 (1·2%)

—	All cases		Confirmed cases	
	Formula only	Human milk*	Formula only	Human milk*
Gestation				
25–27 wk	7/35 (20%)	13/83 (16%)	5/35 (14%)	7/83 (8%)
28–30 wk	7/83 (8%)	11/231 (5%)	5/83 (6%)	6/231 (3%)
31–33 wk	6/75 (8%)	3/263 (1%)	3/75 (4%)	1/263 (0·4%)
34–36 wk	4/43 (9%)	0/113	4/43 (9%)	0/113

Is there a benefit of partial human milk intake ?

- Exclusive Human Milk vs. Exclusive preterm formula (Observational) - 3 trials; NEC - 6/555 infants vs. 24/438 infants. Risk ratio - 0.22 (0.09 - 0.54). **YAY !**
- Any human milk use vs. Exclusive preterm formula (Observational) - 9 studies; NEC – 102/2938 vs. 62/845 infants. Risk ratio - 0.51 (0.34 - 0.76). **YAY !**
- Higher vs. lower human milk intake (Randomized control trials) - 4 studies; NEC – 33/583 vs. 50/533. Risk ratio – 0.54 (0.28 – 1.02). **ALMOST YAY !**
- Higher vs. lower human milk intake (Observational) - >20 studies; NEC – 204/4242 vs. 363/4536. Risk ratio – 0.53 (0.42 – 0.67). **YAY !**
- *Take home: Breast milk better; Some (>40-50ml/kg/day) brings good benefit*

Miller et al. Nutrients 2018; PMID: 29857555

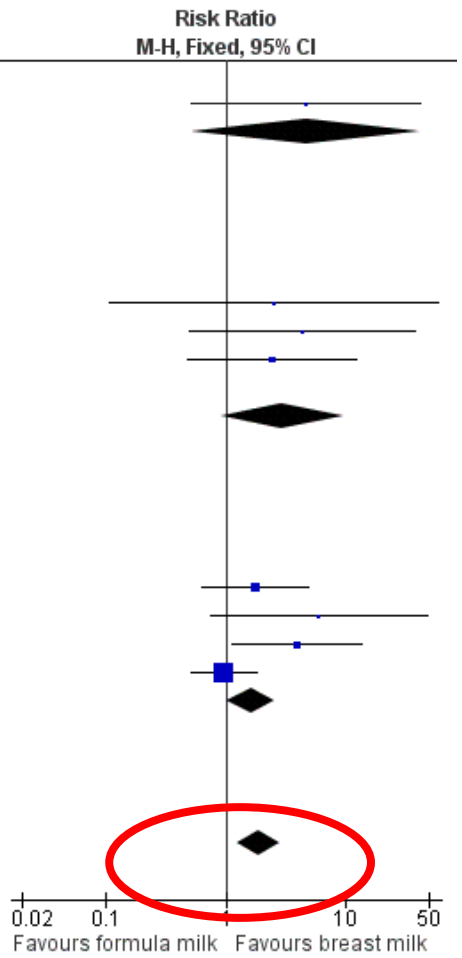
NEC: Donor EBM vs. Preterm formula

- 12 trials (1870 infants): 4 trials Term *vs*. Donor EBM; 8 Preterm Formula vs. Donor EBM. Only 5 trials fortified Donor EBM vs. preterm formula.
- Most studies sponsored by Preterm formula companies; Blinding infrequent; Allocation bias 6 trials.
- Formula-fed infants: Higher rate of weight gain (Mean difference 2.51, 95% CI 1.93 to 3.08 g/kg/day; 9 trials, N =1028; moderate-certainty evidence;)
- Significant subgroup differences existed with the largest effect size for the comparison of preterm formula with unfortified donor breast milk (MD 4.16, 95% CI 3.04 to 5.28 g/kg/day).

Cochrane Database Syst Rev. 2019 Jul 19;7:CD002971. Quigley M[1], Embleton ND, McGuire W.

Donor EBM vs. Preterm formula – NEC rates

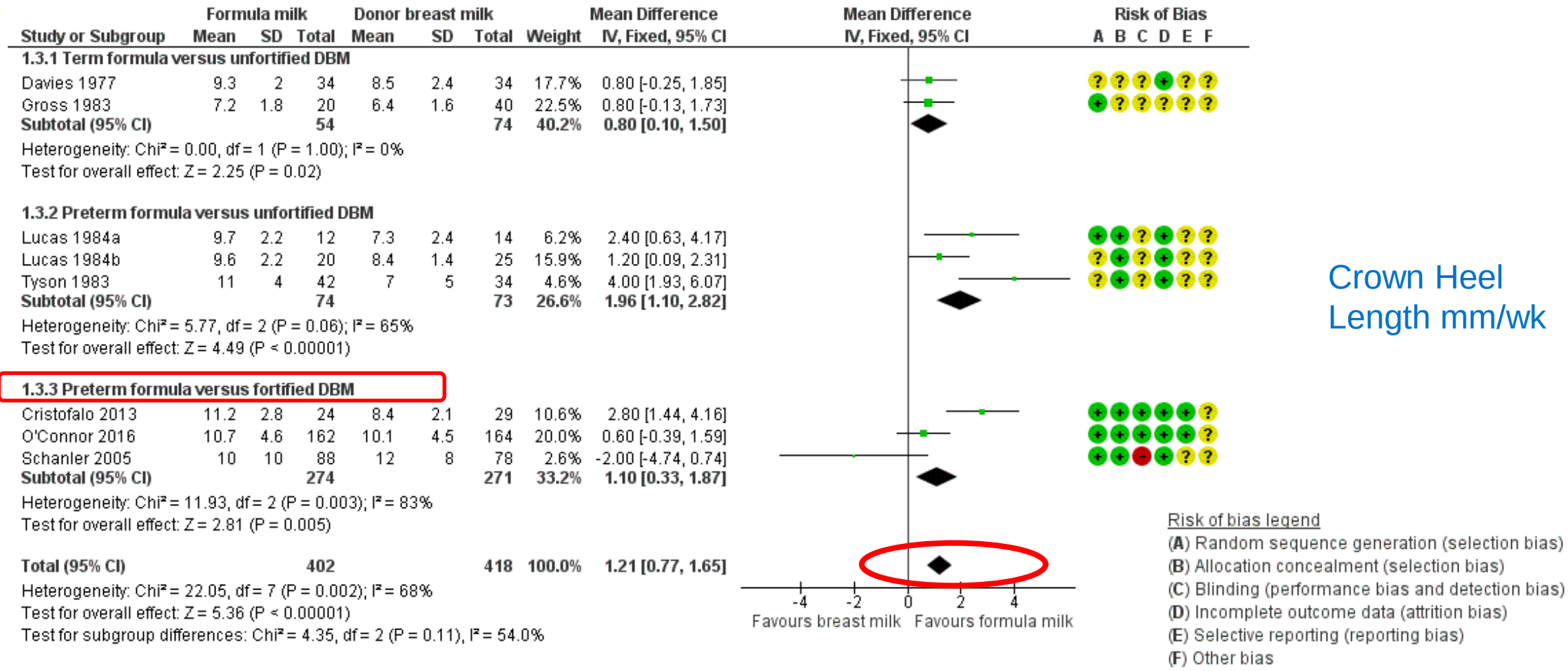
Study or Subgroup	Favours formula milk Events	Total	Donor breast milk Events	Total	Weight	Risk Ratio M-H, Fixed, 95% CI	Year
1.25.1 Term formula versus unfortified DBM							
Gross 1983	3	26	1	41	2.5%	4.73 [0.52, 43.09]	1983
Subtotal (95% CI)		**26**		**41**	**2.5%**	**4.73 [0.52, 43.09]**	
Total events	3		1				
Heterogeneity: Not applicable							
Test for overall effect: Z = 1.38 (P = 0.17)							
1.25.2 Preterm formula versus unfortified DBM							
Tyson 1983	1	44	0	37	1.8%	2.53 [0.11, 60.39]	1983
Lucas 1984a	4	76	1	83	3.1%	4.37 [0.50, 38.23]	1984
Lucas 1984b	5	173	2	170	6.5%	2.46 [0.48, 12.49]	1984
Costa 2018	0	35	0	35		Not estimable	2018
Subtotal (95% CI)		**328**		**325**	**11.4%**	**2.99 [0.90, 9.87]**	
Total events	10		3				
Heterogeneity: Chi² = 0.18, df = 2 (P = 0.91); I² = 0%							
Test for overall effect: Z = 1.80 (P = 0.07)							
1.25.3 Preterm formula versus fortified DBM							
Schanler 2005	10	88	5	78	17.2%	1.77 [0.63, 4.96]	2005
Cristofalo 2013	5	24	1	29	2.9%	6.04 [0.76, 48.25]	2013
O'Connor 2016	12	182	3	181	9.8%	3.98 [1.14, 13.86]	2016
Corpeleijn 2016	17	190	17	183	56.2%	0.96 [0.51, 1.83]	2016
Subtotal (95% CI)		**484**		**471**	**86.1%**	**1.64 [1.03, 2.61]**	
Total events	44		26				
Heterogeneity: Chi² = 6.12, df = 3 (P = 0.11); I² = 51%							
Test for overall effect: Z = 2.09 (P = 0.04)							
Total (95% CI)		**838**		**837**	**100.0%**	**1.87 [1.23, 2.85]**	
Total events	57		30				
Heterogeneity: Chi² = 8.17, df = 7 (P = 0.32); I² = 14%							
Test for overall effect: Z = 2.92 (P = 0.004)							
Test for subgroup differences: Chi² = 1.57, df = 2 (P = 0.46), I² = 0%							



All cause mortality was no different between groups

Cochrane Database Syst Rev. 2019 Jul 19;7:CD002971. Quigley M[1], Embleton ND, McGuire W.

Donor EBM vs. Preterm formula – Linear Growth



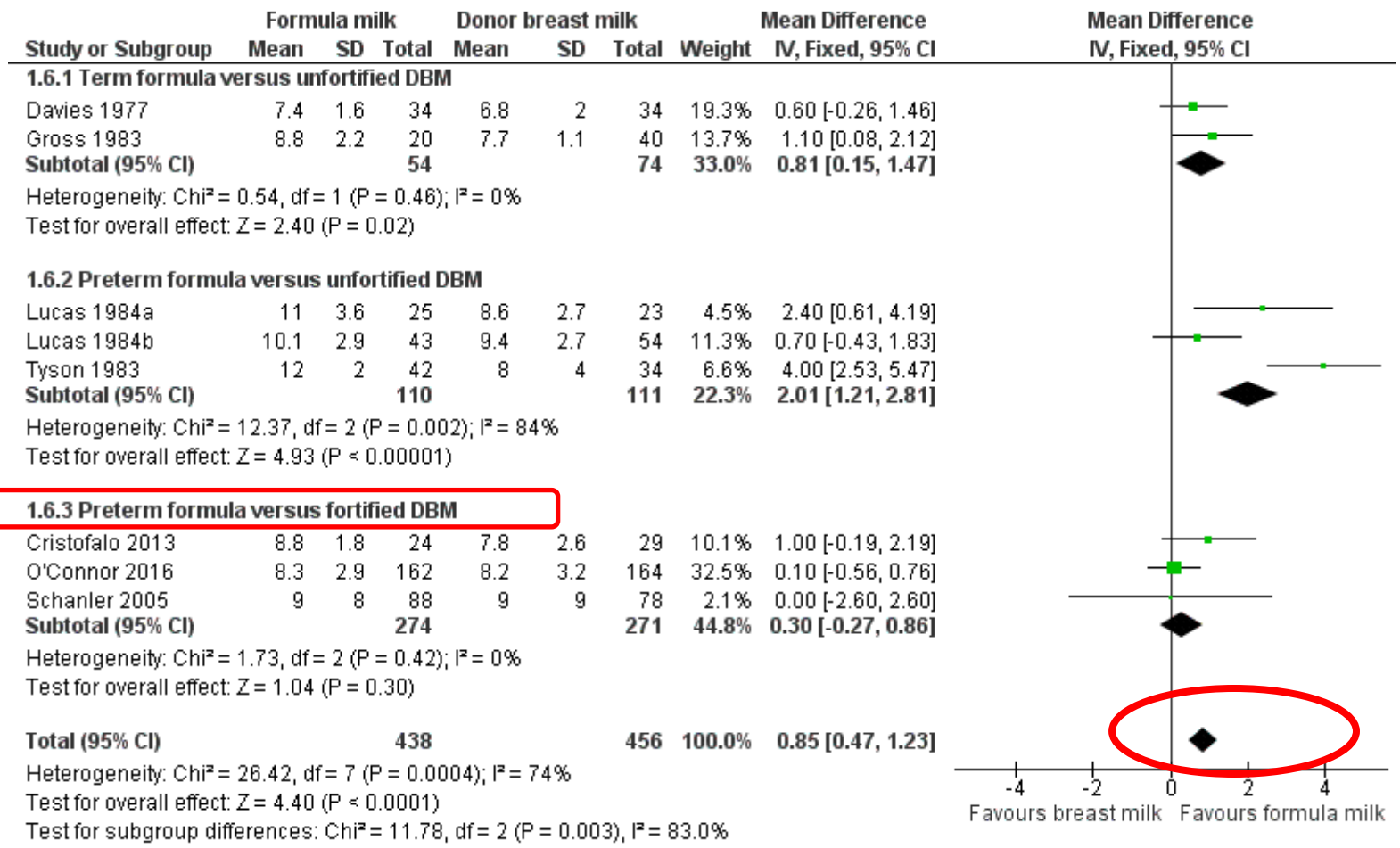
Study or Subgroup	Formula milk Mean	SD	Total	Donor breast milk Mean	SD	Total	Weight	Mean Difference IV, Fixed, 95% CI
1.3.1 Term formula versus unfortified DBM								
Davies 1977	9.3	2	34	8.5	2.4	34	17.7%	0.80 [-0.25, 1.85]
Gross 1983	7.2	1.8	20	6.4	1.6	40	22.5%	0.80 [-0.13, 1.73]
Subtotal (95% CI)			**54**			**74**	**40.2%**	**0.80 [0.10, 1.50]**
Heterogeneity: Chi² = 0.00, df = 1 (P = 1.00); I² = 0%								
Test for overall effect: Z = 2.25 (P = 0.02)								
1.3.2 Preterm formula versus unfortified DBM								
Lucas 1984a	9.7	2.2	12	7.3	2.4	14	6.2%	2.40 [0.63, 4.17]
Lucas 1984b	9.6	2.2	20	8.4	1.4	25	15.9%	1.20 [0.09, 2.31]
Tyson 1983	11	4	42	7	5	34	4.6%	4.00 [1.93, 6.07]
Subtotal (95% CI)			**74**			**73**	**26.6%**	**1.96 [1.10, 2.82]**
Heterogeneity: Chi² = 5.77, df = 2 (P = 0.06); I² = 65%								
Test for overall effect: Z = 4.49 (P < 0.00001)								
1.3.3 Preterm formula versus fortified DBM								
Cristofalo 2013	11.2	2.8	24	8.4	2.1	29	10.6%	2.80 [1.44, 4.16]
O'Connor 2016	10.7	4.6	162	10.1	4.5	164	20.0%	0.60 [-0.39, 1.59]
Schanler 2005	10	10	88	12	8	78	2.6%	-2.00 [-4.74, 0.74]
Subtotal (95% CI)			**274**			**271**	**33.2%**	**1.10 [0.33, 1.87]**
Heterogeneity: Chi² = 11.93, df = 2 (P = 0.003); I² = 83%								
Test for overall effect: Z = 2.81 (P = 0.005)								
Total (95% CI)			**402**			**418**	**100.0%**	**1.21 [0.77, 1.65]**
Heterogeneity: Chi² = 22.05, df = 7 (P = 0.002); I² = 68%								
Test for overall effect: Z = 5.36 (P < 0.00001)								
Test for subgroup differences: Chi² = 4.35, df = 2 (P = 0.11), I² = 54.0%								

Crown Heel Length mm/wk

Risk of bias legend
(A) Random sequence generation (selection bias)
(B) Allocation concealment (selection bias)
(C) Blinding (performance bias and detection bias)
(D) Incomplete outcome data (attrition bias)
(E) Selective reporting (reporting bias)
(F) Other bias

Cochrane Database Syst Rev. 2019 Jul 19;7:CD002971. Quigley M[1], Embleton ND, McGuire W.

Donor EBM vs. Preterm formula – Head Growth



Study or Subgroup	Formula milk Mean	SD	Total	Donor breast milk Mean	SD	Total	Weight	Mean Difference IV, Fixed, 95% CI
1.6.1 Term formula versus unfortified DBM								
Davies 1977	7.4	1.6	34	6.8	2	34	19.3%	0.60 [-0.26, 1.46]
Gross 1983	8.8	2.2	20	7.7	1.1	40	13.7%	1.10 [0.08, 2.12]
Subtotal (95% CI)			**54**			**74**	**33.0%**	**0.81 [0.15, 1.47]**
Heterogeneity: Chi² = 0.54, df = 1 (P = 0.46); I² = 0%								
Test for overall effect: Z = 2.40 (P = 0.02)								
1.6.2 Preterm formula versus unfortified DBM								
Lucas 1984a	11	3.6	25	8.6	2.7	23	4.5%	2.40 [0.61, 4.19]
Lucas 1984b	10.1	2.9	43	9.4	2.7	54	11.3%	0.70 [-0.43, 1.83]
Tyson 1983	12	2	42	8	4	34	6.6%	4.00 [2.53, 5.47]
Subtotal (95% CI)			**110**			**111**	**22.3%**	**2.01 [1.21, 2.81]**
Heterogeneity: Chi² = 12.37, df = 2 (P = 0.002); I² = 84%								
Test for overall effect: Z = 4.93 (P < 0.00001)								
1.6.3 Preterm formula versus fortified DBM								
Cristofalo 2013	8.8	1.8	24	7.8	2.6	29	10.1%	1.00 [-0.19, 2.19]
O'Connor 2016	8.3	2.9	162	8.2	3.2	164	32.5%	0.10 [-0.56, 0.76]
Schanler 2005	9	8	88	9	9	78	2.1%	0.00 [-2.60, 2.60]
Subtotal (95% CI)			**274**			**271**	**44.8%**	**0.30 [-0.27, 0.86]**
Heterogeneity: Chi² = 1.73, df = 2 (P = 0.42); I² = 0%								
Test for overall effect: Z = 1.04 (P = 0.30)								
Total (95% CI)			**438**			**456**	**100.0%**	**0.85 [0.47, 1.23]**
Heterogeneity: Chi² = 26.42, df = 7 (P = 0.0004); I² = 74%								
Test for overall effect: Z = 4.40 (P < 0.0001)								
Test for subgroup differences: Chi² = 11.78, df = 2 (P = 0.003), I² = 83.0%								

MD 0.85, 95% CI 0.47 to 1.23 mm/week; I^2 = 74%; 8 trials, 894 participants; moderate-certainty evidence

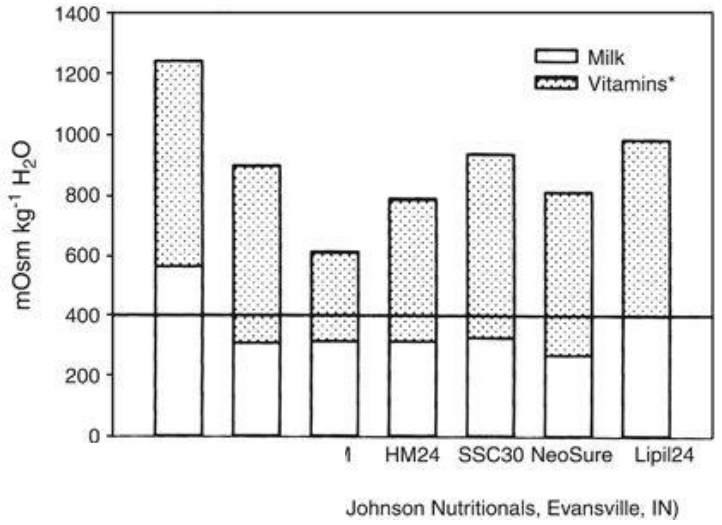
Cochrane Database Syst Rev. 2019 Jul 19;7:CD002971. Quigley M[1], Embleton ND, McGuire W.

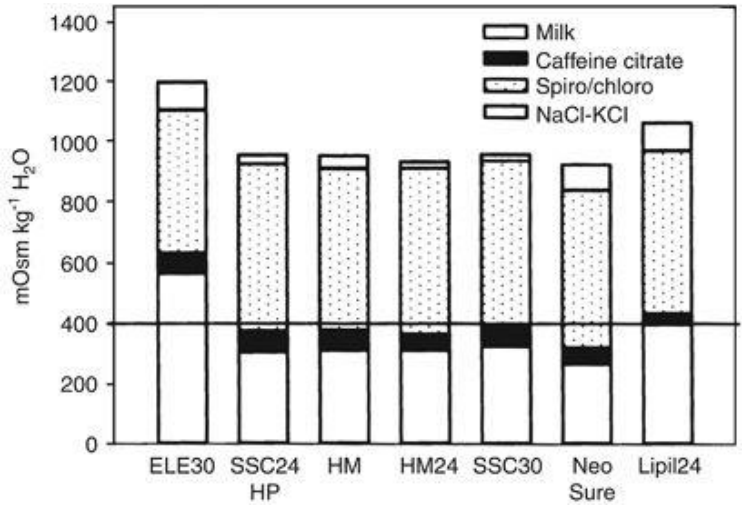
Donor Human Milk - "Yay" or "Nay"

- 18-month Developmental Outcomes in Donor vs. Formula – Less studied; No difference; One study; Trend towards worse outcomes with donor milk.
- No difference is all cause mortality between Donor vs. Formula milk.
- Decreased NEC with Donor EBM (40%) but worse short-term growth outcomes, CMV is also concern; 18-month neurological outcomes unclear.
- In premies receiving > 60mL/kg/day of MOM, Donor EBM does not confer additional NEC benefits (Schanler 2005; Others). If no access to DONOR milk, then even some MOM + formula milk might be ok.
- Three other clinical trials (US and international) ongoing.

Feed intolerance/potential NEC – Careful about Feed Osmolality

- *Journal of Perinatology* (2012) **32,** 227–229; Milk as a vehicle for oral medications: hidden osmoles. P G Radmacher et al.
  - Tested osmolality of fortified feeds with supplements/drugs.
- ~400mOsm/L is recommended; EBM is 300: most formulas are <450. Additives and drugs can markedly increase osmolality.



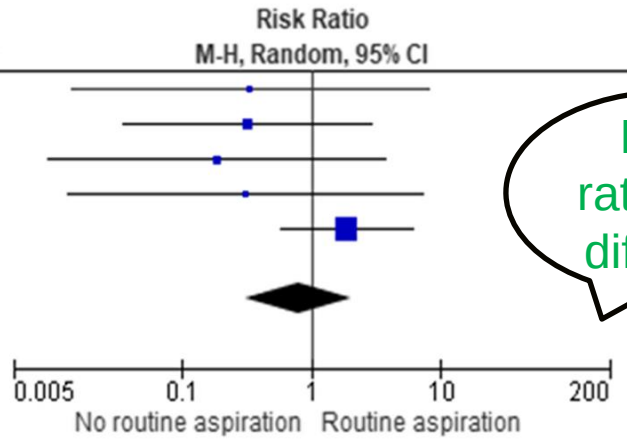


The Bane of a Fellow's existence: Pre-feed NG aspirates and NEC Kumar J et al. Eur J Peds (2021).

- 6 RCTs; 4 compared routine pre-feed aspirate checking to no checking.

Study or Subgroup	No routine aspiration Events	Total	Routine aspiration Events	Total	Weight	Risk Ratio M-H, Random, 95% CI	Year
Kaur A 2015	0	40	1	40	8.9%	0.33 [0.01, 7.95]	2015
Torrazza RM 2015	1	31	3	30	17.8%	0.32 [0.04, 2.93]	2015
Singh B 2018	0	45	2	42	9.8%	0.19 [0.01, 3.78]	2018
Thomas S 2018	0	26	1	24	9.0%	0.31 [0.01, 7.23]	2018
Parker LA 2019	7	69	4	74	54.5%	1.88 [0.57, 6.13]	2019
Total (95% CI)		211		210	100.0%	0.80 [0.31, 2.08]	
Total events	8		11				

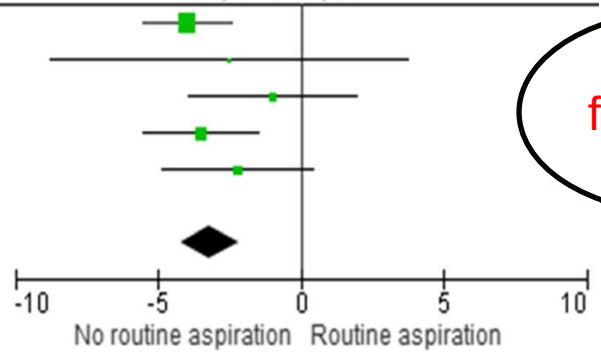
Heterogeneity: Tau² = 0.07; Chi² = 4.21, df = 4 (P = 0.38); I² = 5%
Test for overall effect: Z = 0.46 (P = 0.64)



NEC rates not different

Study or Subgroup	No routine aspiration Mean	SD	Total	Routine aspiration Mean	SD	Total	Weight	Mean Difference IV, Random, 95% CI	Year
Kaur A 2015	10	2.96	40	14	4.07	40	43.7%	-4.00 [-5.56, -2.44]	2015
Torrazza RM 2015	14.3	12.5	31	16.8	12.4	30	2.7%	-2.50 [-8.75, 3.75]	2015
Singh B 2018	11	3.99	45	12	8.99	42	12.1%	-1.00 [-3.96, 1.96]	2018
Thomas S 2018	6	2.04	26	9.5	4.63	24	26.2%	-3.50 [-5.51, -1.49]	2018
Parker LA 2019	15.9	7.9	69	18.1	8.2	74	15.2%	-2.20 [-4.84, 0.44]	2019
Total (95% CI)			211			210	100.0%	-3.19 [-4.22, -2.16]	

Heterogeneity: Tau² = 0.00; Chi² = 3.82, df = 4 (P = 0.43); I² = 0%
Test for overall effect: Z = 6.07 (P < 0.00001)



Time to full feeds slower

Bolus vs. continuous feeds in VLBW infants

- Bolus feeds more “physiological” or is it ? Baby vs. “fetus”

Continuous vs. Bolus feeds infants < 1500g. Premji SS, Chessell L (Cochran 2011)

- 7 trials, (n=511 infants), b.wt 500-1500g.
- No difference for time to full feeds or NEC rates.
- One study showed trend towards more apneas with bolus feeds.
- One study- sub-group analysis; Infants <1000g had better weight gain on continuous feeds, earlier discharge to home.

- Impact of Continuous vs Bolus Feeding on Splanchnic Perfusion in Very Low Birth Weight Infants: A Randomized Trial. Bolus feeds increases SMA Doppler flows, NIRS stable. Bozzetti V et al, J Pediatr.

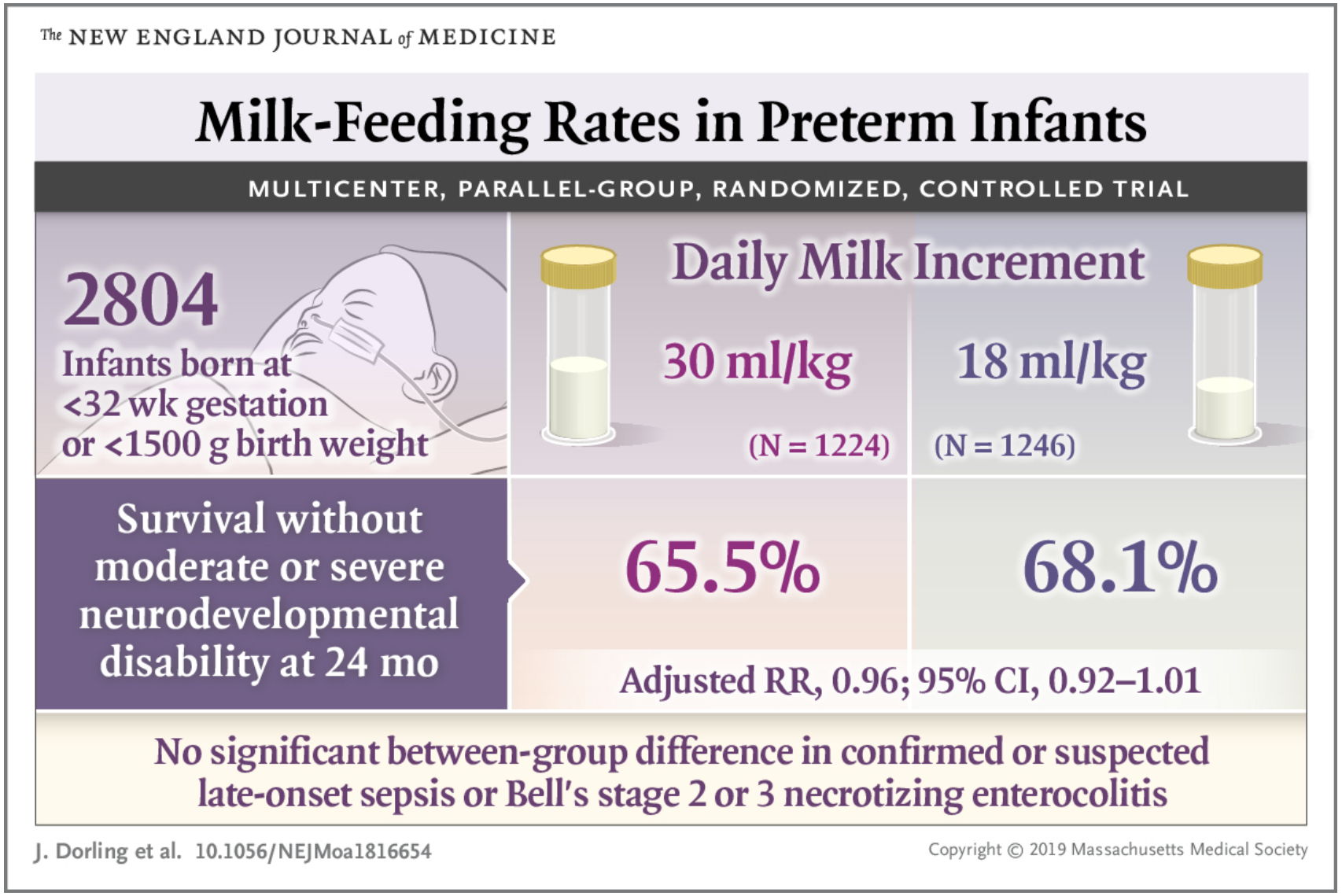
Conclusion: It’s a wash, bias towards continuous feeds in ELBW babies.

Does advancing feeds slowly prevent NEC ?

14 RCTs. N=4033 infants (2804 - one large trial). 33% were ELBW infants.

15-24 ml/kg/day vs. 30-40ml/kg/day. Oddie et al. Cochrane Database Sys Rev 2021 Aug 24;8(8):CD001241.

Outcomes	Anticipated absolute effects* (95% CI)		Relative effect (95% CI)	No. of participants (studies)	Certainty of the evidence (GRADE)
	Risk with faster rates of enteral feed advancement	Risk with slow rates of enteral feed enhancement			
Necrotising enterocolitis before hospital discharge	54 per 1000	57 per 1000 (45 to 77)	RR 1.06 (0.83 to 1.37)	4026 (14 trials)	⊕⊕⊕⊖ MODERATE[a]
Mortality before hospital discharge	71 per 1000	80 per 1000 (64 to 98)	RR 1.13 (0.91 to 1.39)	3860 (13 trials)	⊕⊕⊕⊖ MODERATE[a]
Feed intolerance before hospital discharge	282 per 1000	333 per 1000 (268 to 412)	RR 1.18 (0.95 to 1.46	719 (9 trials)	⊕⊕⊖⊖ LOW[a, b]
Invasive infection before hospital discharge	170 per 1000	194 per 1000 (168 to 223)	RR 1.14 (0.99 to 1.31)	3583 (11 trials)	⊕⊕⊖⊖ LOW[a, b]



SIFT trial
Death+mod-severe developmental impairment at 24 month CGA – No difference

TANEC - Grading of Recommendations Assessment, Development and Evaluation system. Hay et al. Seminar Perinatol Feb 2017

- Transfusion associated NEC happens in chronically anemic preterm infants 48 hr after transfusion. Usually ELBW infants, can be quite severe.
- 45 studies, narrowed it down to 26 studies (23 observational, 3 RCT).
- Overall quality of studies "low" to "very low". RCTs- NEC not primary outcome.
- TANEC (<48hr) - 1.13 (0.99-1.29), NEC anytime after PRBCs (1.95 (1.6 -2.4)], From RCT (n=3), NEC was lower with liberal transfusion [0.6 (0.3 – 1.21)].
- A) Major bias – unadjusted for co-variates; potential for confounding for indication. *Example – apnea – transfusion – NEC.* B) Significant inconsistency among studies in trend of results; C) No specific definition of TRALI process.

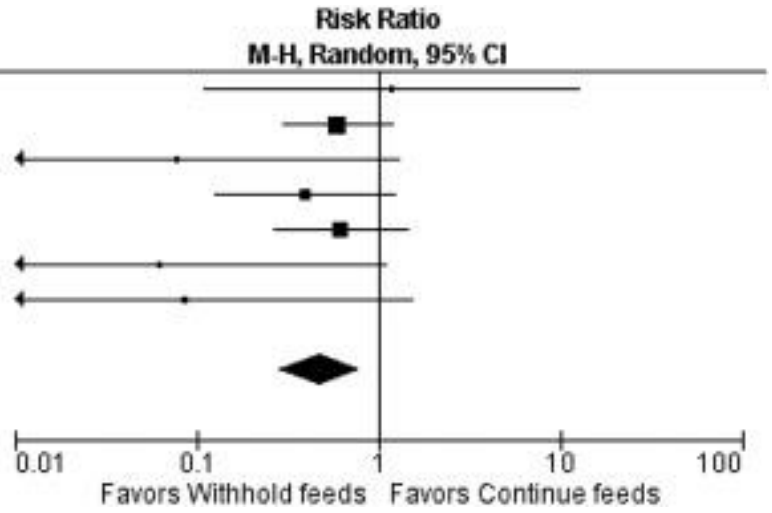
TANEC - Impact of NPO before and after transfusion

- Holding feeds 4-6 hr before, during, and 4-6 hr after PRBC transfusion common.
- One very small RCT (N=22); no differences in Splanchnic blood flow or NEC.
- 7 studies which compared pre- and post policy change. (N=7492 infants)

Study or Subgroup	Withhold feeds Events	Total	Continue feeds Events	Total	Weight	Risk Ratio M-H, Random, 95% CI
Bajaj et al. (12)	2	86	1	51	4.7%	1.19 [0.11, 12.76]
Derienzo et al. (9)	9	315	51	1065	38.8%	0.60 [0.30, 1.20]
Doty et al. (14)	0	64	11	116	3.3%	0.08 [0.00, 1.31]
Meneses et al. (10)	4	573	11	628	18.0%	0.40 [0.13, 1.24]
Mohamed et al. (11)	7	872	20	1550	28.7%	0.62 [0.26, 1.47]
Perciaccante and Young (5)	0	306	7	289	3.3%	0.06 [0.00, 1.10]
Rindone et al. (13)	0	742	6	835	3.2%	0.09 [0.00, 1.53]
Total (95% CI)		**2958**		**4534**	**100.0%**	**0.47 [0.28, 0.80]**
Total events	22		107			

Heterogeneity: $Tau^2 = 0.06$; $Chi^2 = 6.71$, $df = 6$ ($P = 0.35$); $I^2 = 11\%$

Test for overall effect: $Z = 2.81$ ($P = 0.005$)



Take home : Possibly safer to hold feeds for PRBC Rx (low quality of evidence)

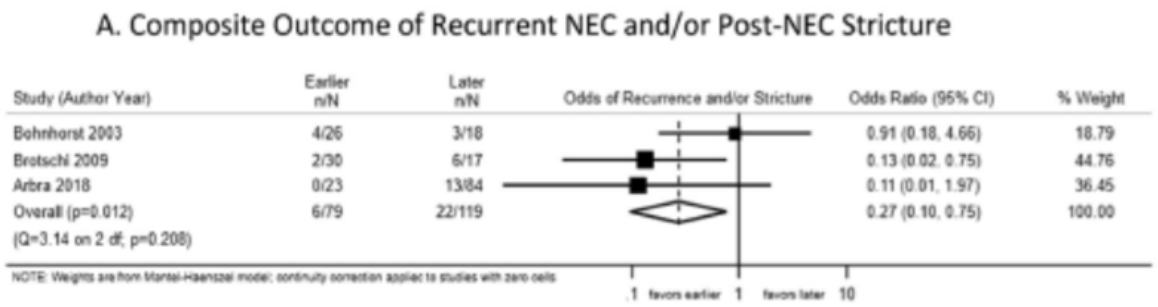
Quick Takes

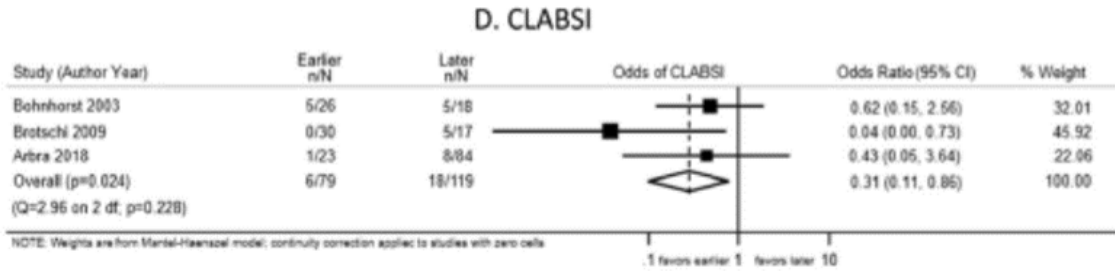
Can we feed infants during medical treatment of PDA ? (Stage I or II)

YAY: Observational study Louis D et al. J Perinatol. 2016 Jul;36(7):544-8.

YAY: RCT (Clyman R, J Pediatr 2013). Trophic vs. NP). Safe, time to FEF faster.

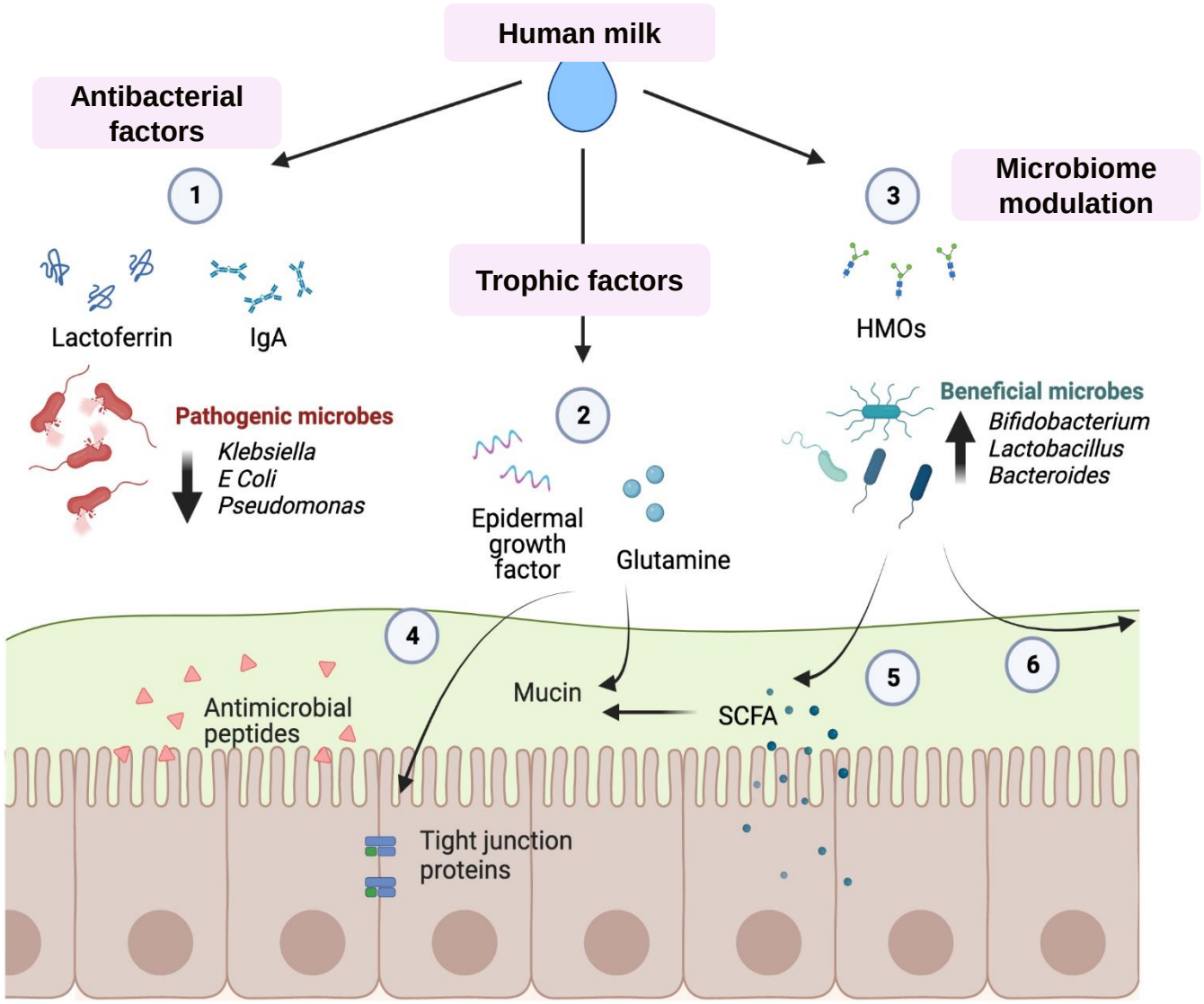
How soon after medical NEC can we feed ? 5d, 7d or forever? (Patel/Ryan et al.)

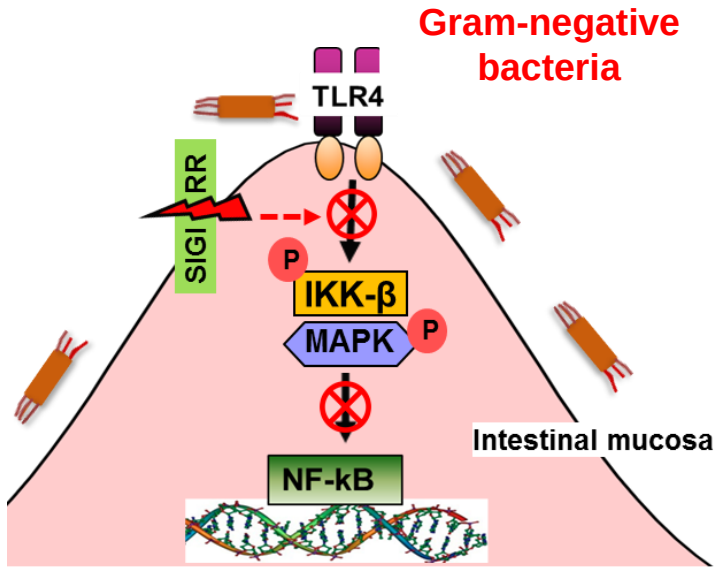




Take home: Early limited evidence - Can re-feed after medical NEC around 4-5 days.

Human Milk - Beyond Nutrition





Intestinal inflammation, injury and NEC

Adapted from Sampath et al. Mucosal Immunology March 2013

Probiotics and NEC: mechanisms

1. Improved barrier function (preventing bacteria from invading gut)
    1. Apoptosis
    2. Tight junctions
    3. Mucin production
2. Decreased inflammation by gene regulation
    1. Less TLR4, More SIGIRR, A20
    2. IL1-beta, IL6, TNF-alpha
    3. Tryptophan metabolites (Indole-3-lactic acid)
3. Alteration of the microbiota
    1. Bacteriocins
    2. Competition for nutrients (HMOs, Fe)

Yu Y, PLoSOne 2020
Meng D, Pediatr Res 2020
Halloran K, Early Hum Dev 2019
Cuna/Wei/Sampath 2020

Probiotics and NEC: Observational

Systematic review of non-RCTs using Cochrane methodology
Good-quality studies from 18 countries

Outcome	Studies	Preterm infants	OR (95% CI)	P value	Quality of evidence
NEC stage 2 or 3	30	77,018	0.60 (0.50, 0.73)	<0.00001	Moderate
LOS	21	65,858	0.85 (0.74, 0.97)	0.02	Low
Death	27	70,977	0.77 (0.68, 0.88)	0.0001	Low

ELBWs: NEC stage >2: 4.5% (probiotic) vs 7.9% (no probiotic)

Deshmukh H
Adv Nutr 2021

Courtesy of Dr. Mark Underwood MD

Probiotics and NEC: RCTs

Systematic review of RCTs using Cochrane methodology

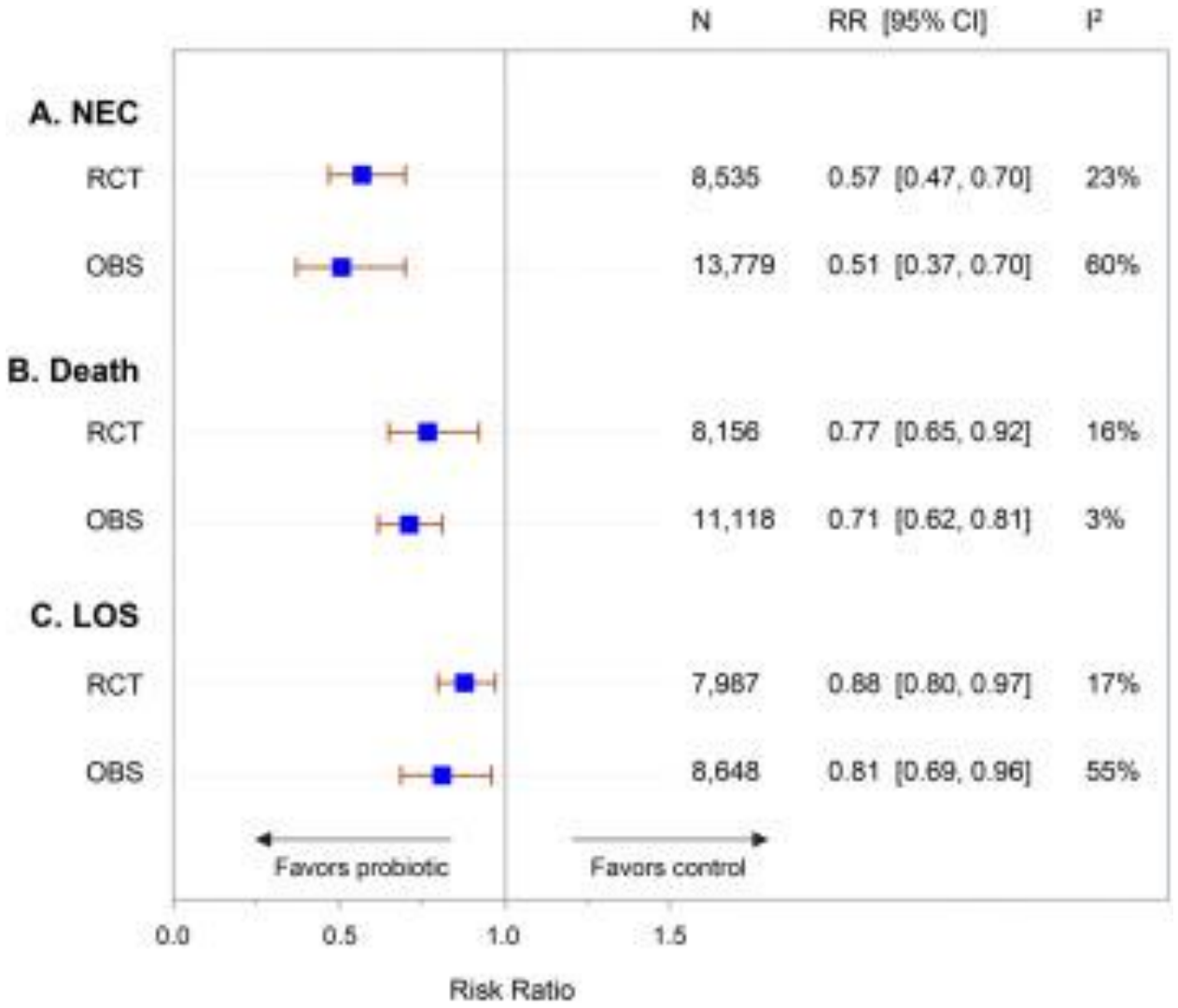
Outcome	Trials	Preterm infants	RR (95% CI)	NNTB (95% CI)	Certainty of evidence
NEC stage 2 or 3	54	10,604	0.54 (0.45, 0.65)	33 (25, 50)	Low
NEC stage 2 or 3 (low risk of bias)	16	4597	0.70 (0.55, 0.89)	50 (33, 100)	Moderate
LOS	47	9762	0.89 (0.82, 0.97)	50 (33, 100)	Moderate
Death	51	10,170	0.76 (0.65, 0.89)	50 (50, 100)	Moderate

Meta-analysis did not show effects on NEC, death, or infection for ELBW infants (low-certainty evidence)

Courtesy of Dr. Mark Underwood MD

Sharif S, Cochrane Database Sys Rev 2020

Probiotics in preterm infants – Striking evidence



R.M. Patel, M.A. Underwood / Seminars in Pediatric Surgery 27 (2018) 39–46

Probiotics - Drawbacks

- Sepsis from probiotics - Rare, Saccharomyces, Lactobacillus rhamnosis in preterm infants. Under reported ?; In RCTs, sepsis rate lower with probiotics. Contamination with pathogenic bacteria reported.
- Main issue; Which preparation; how many bacteria? IND vs.dietary supplement.
- Cross-contamination (49%) of placebo infants in trials get colonized.
- Which probiotics is best ?
  - Combination generally better (Infloran – Bifidobacteria + Lactobacillus).
  - Bifidobacteria> Lactobacillus> Saccharomcyes.
  - Underwood MA: parental consent trial appropriate ?? (best in US Florababy, L. rhamnsosus + Bifidobacteria 4 strains); Infloran (L.acidophilus B.infantis)

.

Lactoferrin

- Innate Immune protein; 20x concentration in human milk > bovine milk. Anti-bacterial, anti-viral effect. Deprives Iron and direct Iron membranes.
- Both human recombinant and bovine lactoferin commercially available.
- No differences in NEC stage II or III (typical RR 1.10, 95% CI, 0.86 to 1.41; 7 studies, 4874 participants; low-certainty evidence).
- Confirmed late-onset sepsis (typical RR 0.83, 95% CI 0.73 to 0.94; typical RD -0.03, 95% CI, -0.04 to -0.01; NNTB 33, 12 studies, 5425 participants, low-certainty evidence).
- Combined with probiotics - NEC stage II or III (RR 0.04, 95% CI 0.00 to 0.62; NNTB 20, 95% CI 12.5 to 33.3; 1 study, 496 participants; very low-certainty evidence),

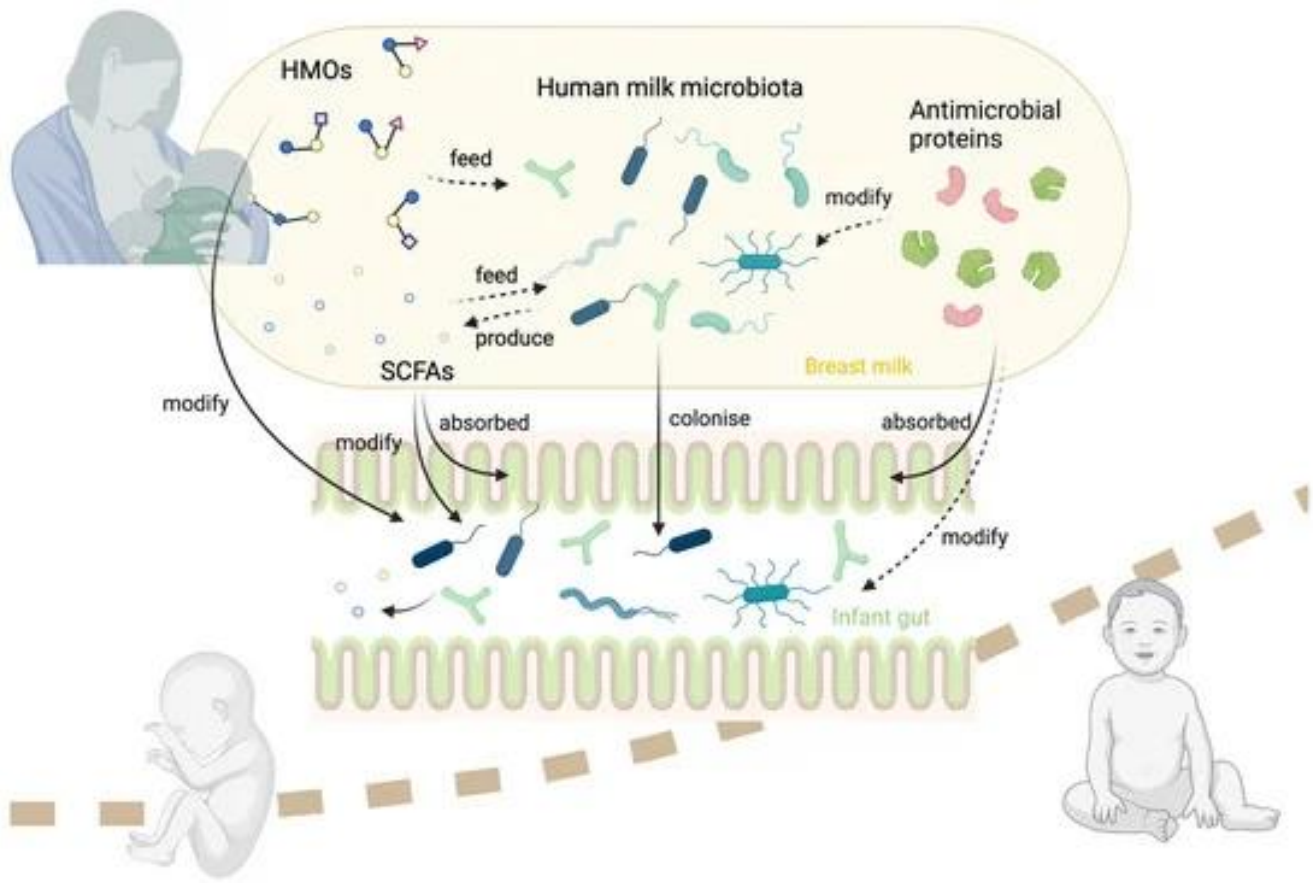
•Pammi et al. 2020 Cochrane Database PMID: 32232984, PMCID: PMC7106972

NEC prevention – Bacteria-free Prebiotics ?

- 3rd most abundant component of human milk is ?

- Human milk oligosaccharides - 10-15g/L of mother's milk. Non absorbable carbohydrates not of direct nutritional value to babies.

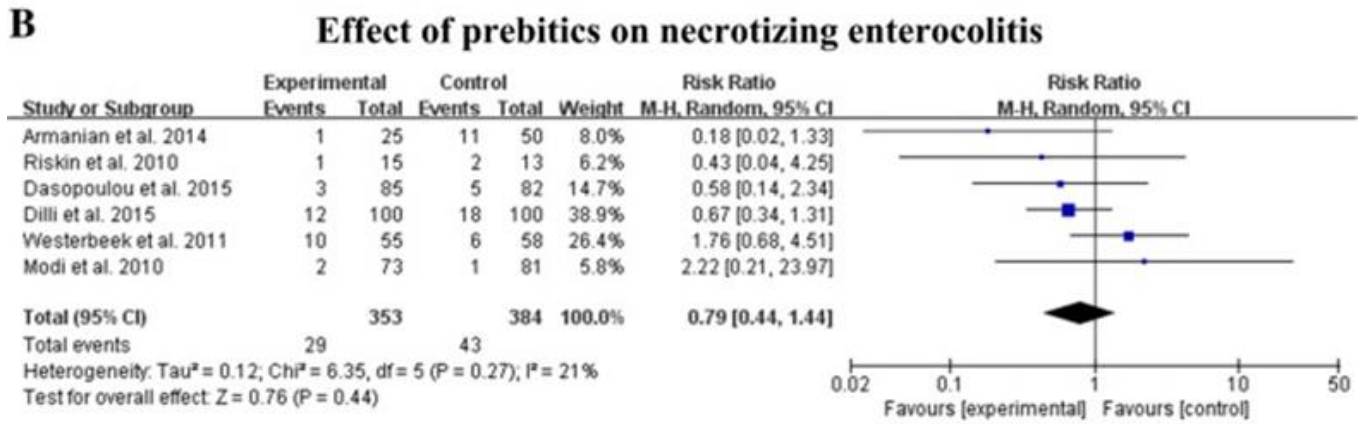
- Favor growth of Good bacteria Bifidobacteria spp.
  Underwood et al. *Pediatric Research* **volume 86**, pages749–757 (2019)

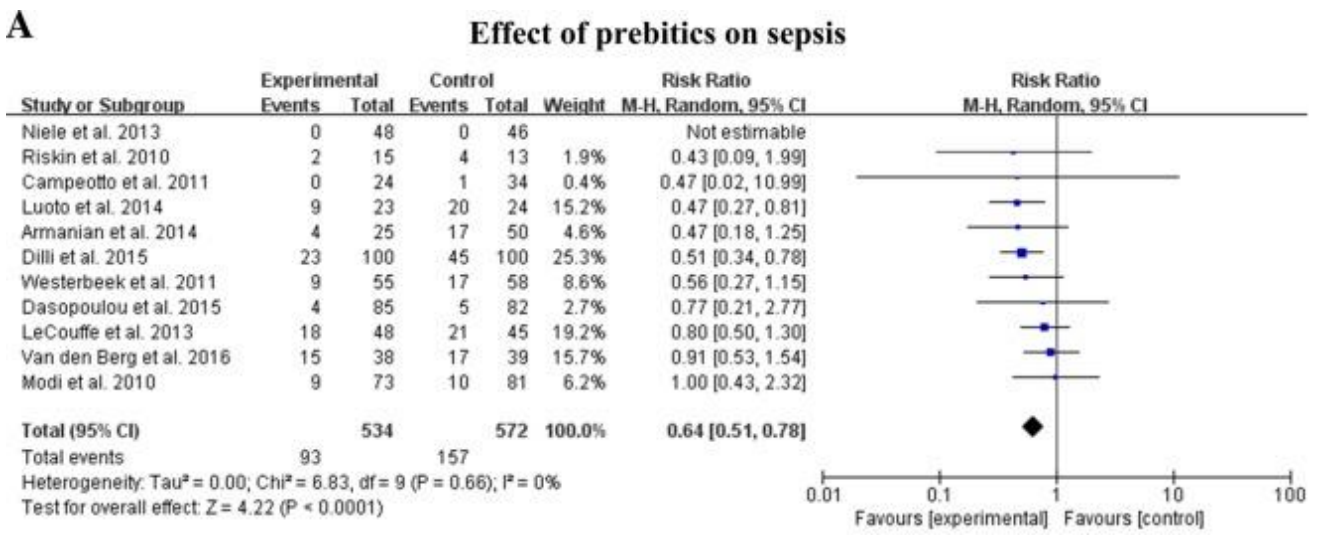


Ma et al. *Nutrients* **2022**, *14*(23), 5148; https://doi.org/10.3390/nu14235148

NEC prevention – Bacteria-free Prebiotics ?

- 18 RCTs; N=1322 infants
- A variety of Non-human manufactured oligosaccharides tested in preterm infants.
- Short chain galacto-Oligos (scGOS); Long chain fructo-Oligos (lcFOS); Pectin-derived Oligos (pAOS)





Lange et al. Nutrients. 2021 May; 13(5): 1726. PMC8161173; Chi et al. Eur J Clin Nutr. 2019; 73(5): 657–670. PMC6760619

Short Chain Fatty acids

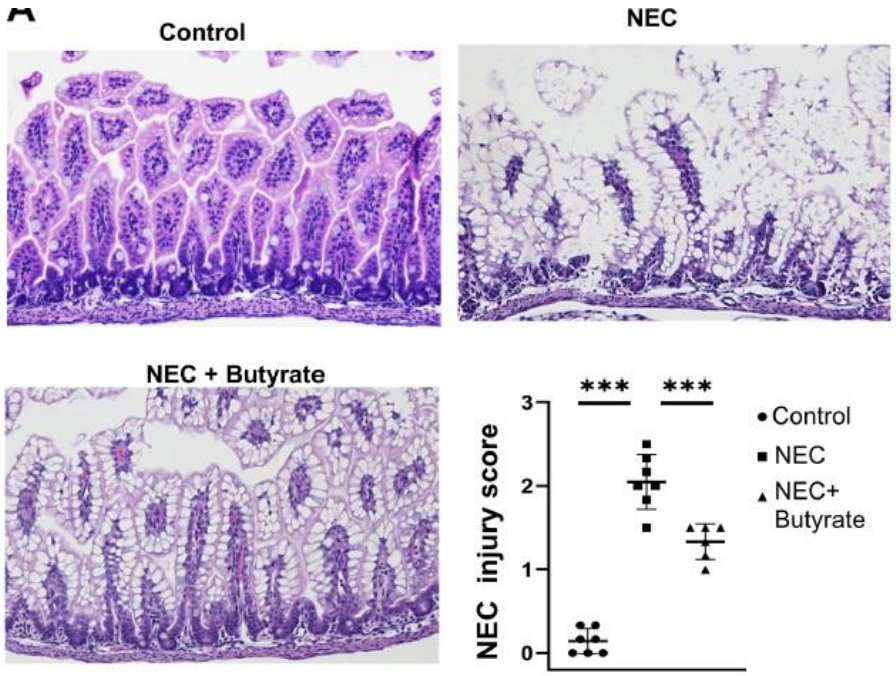
Therapeutic Potential of Gut Microbiota and Its Metabolite Short-Chain Fatty Acids in Neonatal Necrotizing Enterocolitis *Life* **2023**, *13*(2, **Alsharairi**

- Produced by "Good bacteria" .
- Bifidobacteria/Clostridia use HMO's for growth and secrete SCFA.
- SCFA sustain mucosal barrier, regulate immunity and suppress inflammation.

Take home: Interesting!

Short-chain fatty acids ameliorate necrotizing enterocolitis-like intestinal injury through enhancing Notch1-mediated single immunoglobulin interleukin-1-related receptor, toll-interacting protein, and A20 induction

Wei Yu,[1,2] Aparna Venkatraman,[1,2] Heather L. Menden,[1,2] Maribel Martinez,[1,2] Shahid Umar,[3] Venkatesh Sampath[1,2]



Quick Takes

- Polyunsaturated fatty acids
- n-3 long chain PUFA: 11 trials, N>1700 infants, No reduction in NEC rates.
- Enteral glutamine supplementation - No reduction in NEC
- Enteral arginine in small trials reduced NEC (moderate certainty)
- Oral administration of IgA and IgG – di not reduce NEC.
- **Future:** Direct Toll Like Receptor Antagonism – used novel peptides/nanoparticles ??

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Megan Tucker, MD
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Dr. Ravi Patel, MD (Emory)
Dr. Misty Good, MD (WashU)
Dr. Matt Rogers, PhD & Mike Morowitz, MD (UPMC)
Dr. Mark Underwood, MD

Neonatal Diseases Research Program

Interested in joining – vsampath@cmh.edu

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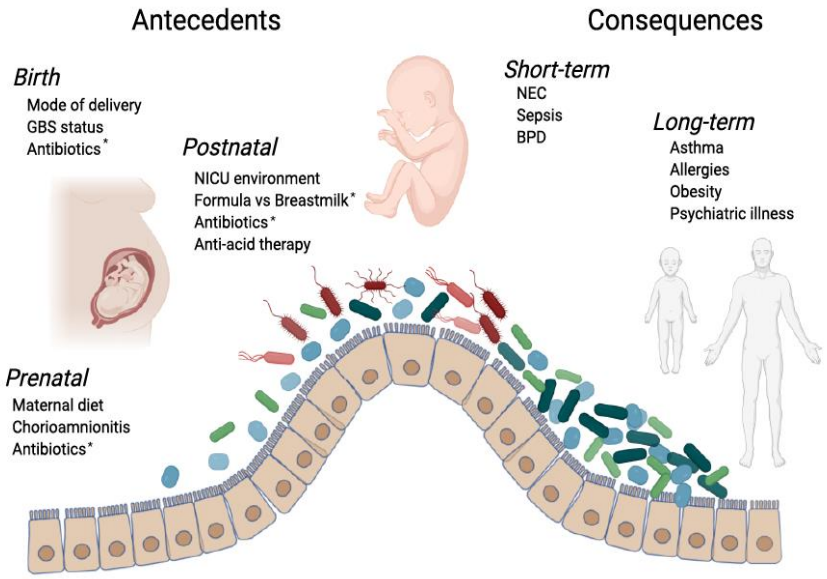
Microbiome in NEC



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Summary

Prenatal influences – Long term consequences

NEC associated microbiota patterns include:

- Gammaproteobacterial excess (Gram-ve)
- Decreased diversity & Bifidobacteria
- Effect of formula feeding/delivery mode

Evidence for dysbiosis and late-onset sepsis

(Cuna/Sampath et al. AJP Gastro and Hepatol April 2021)

Breast Milk - Nutrition, Anti-pathogen Immunity, Symbiosis

NEC-protective factors in human milk

Nitrate and/or nitrite and antioxidant factors[66,163]

L-arginine[164,165]

Human milk oligosaccharides and prebiotics[138,139,166–168]

Lactoferrin[121,169–172]

Secretory IgA[173]

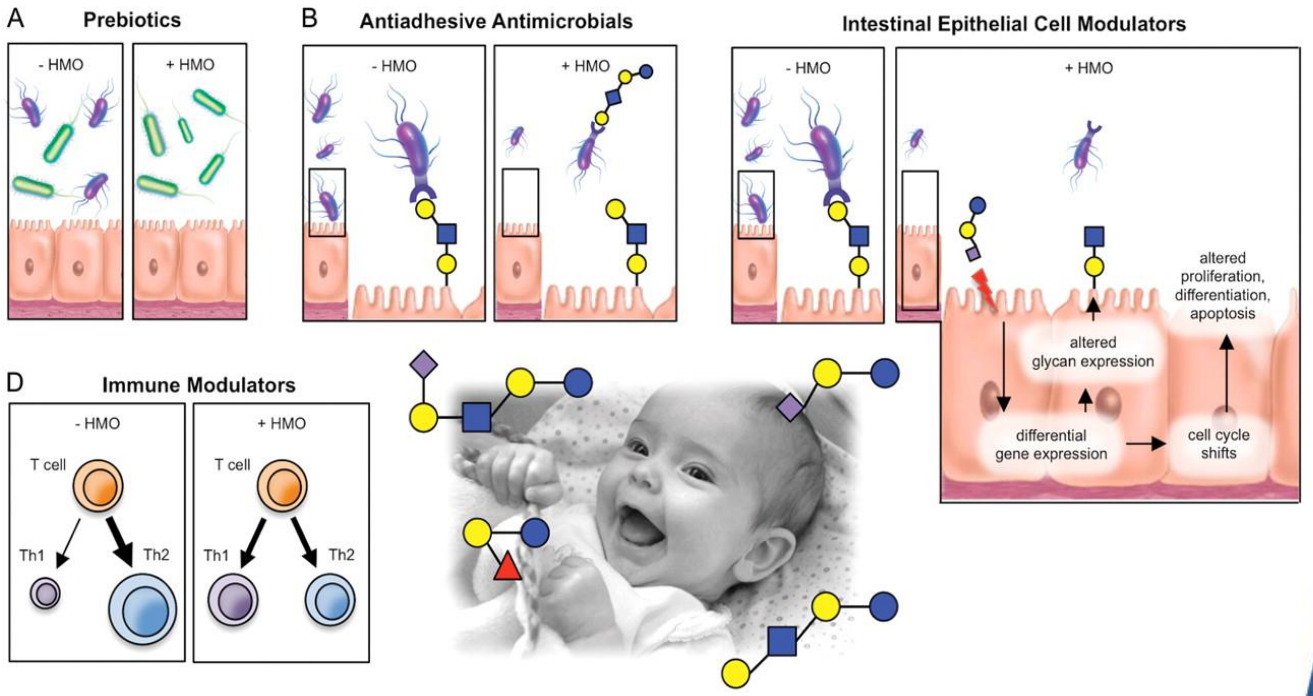
Platelet-activating factor acetylhydrolase[90,95]

Growth factors:

- Epidermal growth factor[174–176]
- Heparin-binding EGF-like growth factor[177–179]
- Transforming growth factor β2[96]
- Erythropoietin[180]

Nino et al. Necrotizing enterocolitis: new insights into pathogenesis and mechanisms. Nat Rev Gastroenterol Hepatol. 2016 October ; 13(10): 590–600.

Human Milk Oligosaccharides: Glycosylated non-digestibe sugars that are used by ONLY good bacteria (Bifidobacteria)



Glycobiology, Volume 22, Issue 9, September 2012, Pages 1147–1162, https://doi.org/10.1093/glycob/cws074

Preterm formula vs. Donor EBM for premies – Neurodevelopmental outcomes

Centres	Cambridge, Ipswich, King's Lynn		
No randomised	502		
Neonatal diets assigned randomly			
Trial A: Diets used as sole enteral feed (mother chose not to provide her EBM)	BBM	*v*	PTF
Trial B: Diets used as supplements to mother's EBM	BBM (+EBM)	*v*	PTF (+EBM)

Developmental test	*Diet*		*Advantage for PTF (95% CI)*
	BBM	*PTF*	
Trial A*	*n=62*	*n=52*	
MDI	94·8 (2·1)	95·3 (2·7)	0·5 (−6·2 to 7·1)
PDI	93·0 (1·8)	94·2 (2·2)	1·2 (−4·4 to 6·8)
Trial B†	*n=134*	*n=139*	
MDI	102·2 (1·7)	103·8 (1·7)	1·6 (−3·1 to 6·2)
PDI	95·5 (1·3)	94·5 (1·4)	−1·0 (−4·8 to 2·7)
Trials A plus B*	*n=196*	*n=191*	
MDI	99·9 (1·3)	101·5 (1·4)	1·6 (−2·3 to 5·5)
PDI	94·7 (1·1)	94·4 (1·2)	−0·26 (−3·4 to 2·8)

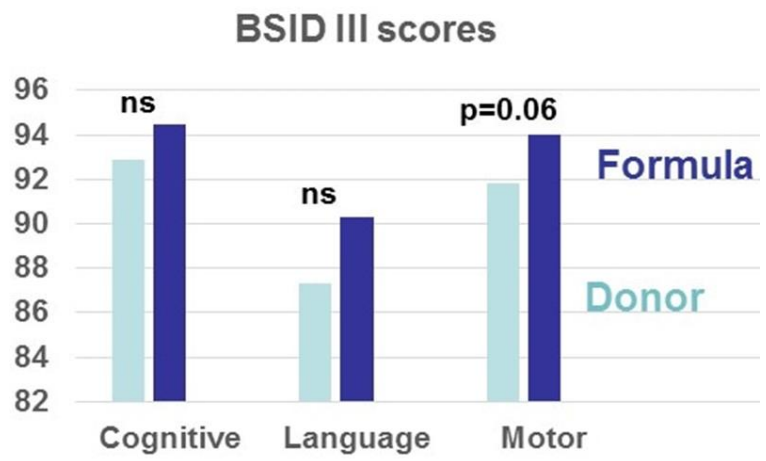
A randomised multicentre study of human milk versus formula and later development in preterm infants A Lucas, R Morley, T J Cole, S M Gore

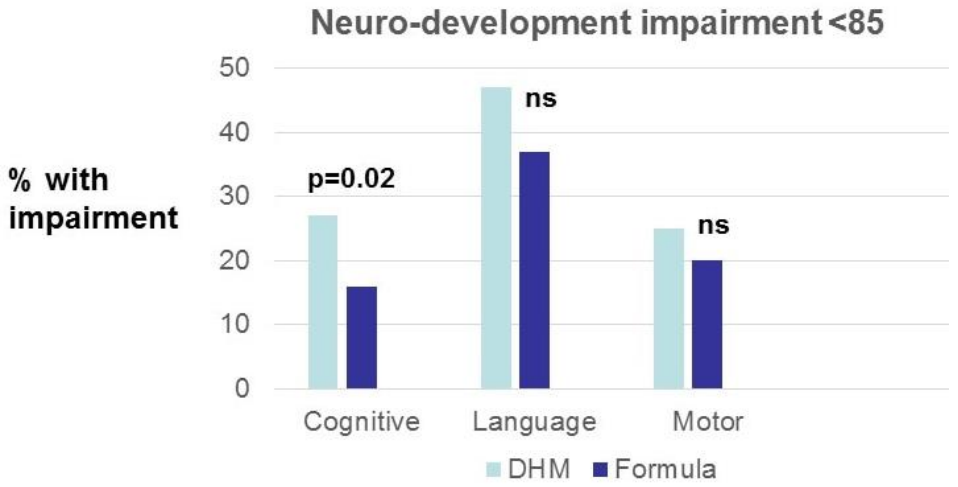
Archives of Disease in Childhood 1994; **70:** F141–F146

Preterm formula vs. breast milk (donor) no adverse 18 month neurological outcomes

Donor or Formula for supplementation of MOM and 24 month outcomes; O'Connor et al. 2016 JAMA Pediatrics

- Effect of Donor human milk vs. Formula for MOM shortfall. 18 month neurodevelopmental outcome was primary outcome.





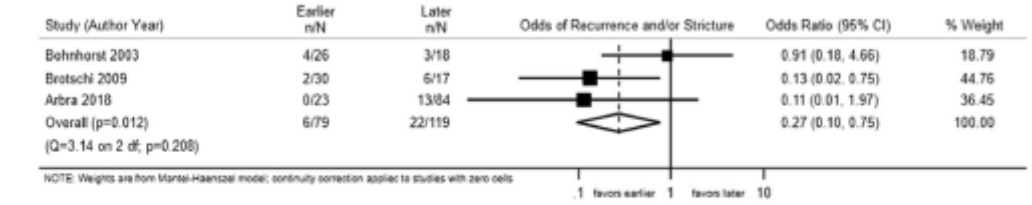
- No differences in growth and head circumference at 24 months.

Take home: DHM does not confer improved 24month outcomes, trend towards worse neurological outcomes with DHM (need further studies).

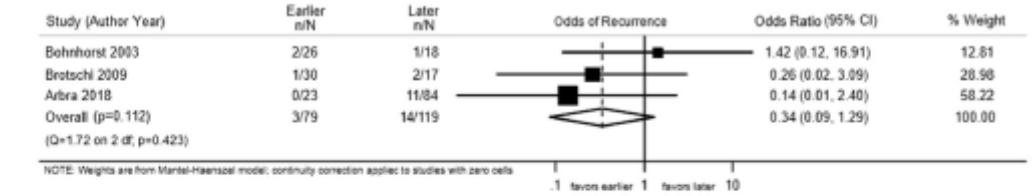
Courtesy of Nick Embleton, MD Neonatologist (Newcastle upon Tyne, UK)

Patel…Ryan et al

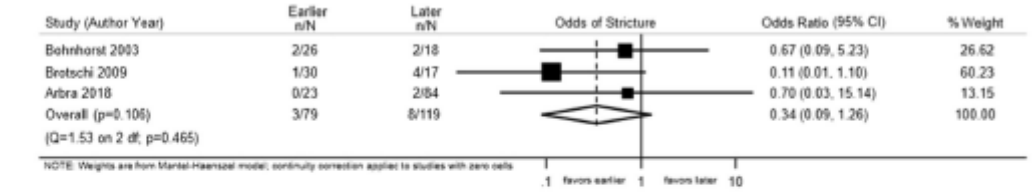
A. Composite Outcome of Recurrent NEC and/or Post-NEC Stricture



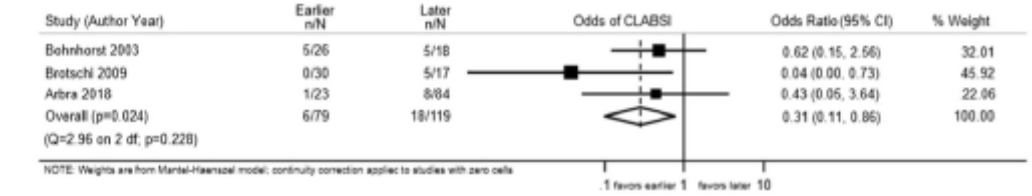
B. Recurrent NEC



C. Post-NEC Stricture



D. CLABSI



No increase in any negative outcomes with earlier refeeding. NEC and post-NEC stricture improved with early re-feeding

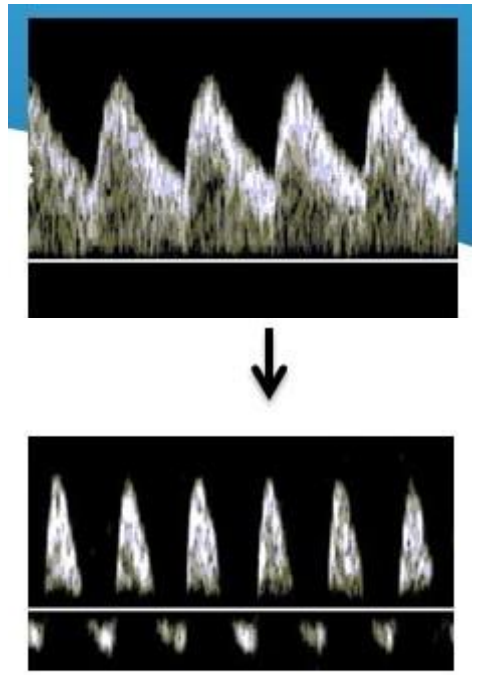
- Early re-feeding (<5 days or < 7 days) after NEC onset not associated with worse medical NEC outcomes.
- Trend towards less strictures, early discharge with early feeds; No change in mortality. Bias: More severe NEC and later re-feeding.
- *Early refeeding safe in infants who develop stage I/IIA NEC; possibly safe in stage IIB NEC if pneumatosis has resolved.*

NEC ischemia perfusion and TANEC

- An outbreak of necrotizing enterocolitis. Association with transfusions of packed red blood cells. McGrady GA et al; Am J Epidemiol. 1987 Dec;126(6):1165-72.
- **Transfusion associated necrotizing enterocolitis: Meta-analysis of observational data.** Mohamed A[1], Shah PS. Pediatrics. 2012 Mar;129(3):529-40. Feb 20.
  - No RCT. 11 case-control studies; NEC associated with PRBC transfusion <48hr. TANEC infants less mature, 500gm heavier, PDA, etc. More severe disease.
- **Transfusion-associated necrotising enterocolitis in neonates.** Stritzke et al. Arch Dis Child Fetal Neonatal Ed. 2013 Jan;98(1):F10-4.
  - Canadian neonatal research network; 2003-08, All NEC (n=927), controls- 2700. PRBC <48 hrs higher in cases vs. controls (15% vs. 7.5%). TANEC cases were smaller and less mature, higher SNAPII-scores.
  - Outcomes for TANEC *vs*. other NEC no different for mortality/CNS injury/ROP.

Feeding in high-risk premature infants - ADEPT trial

- Leaf et al , Pediatrics April 2012; N=404; UK; 52 centers.
  - Infants < 35 week with Absent or reversed diastolic flows, <10% centile for weight randomized.
  - Feeds early **Day 2** vs. Late **Day 6**; once started similar rate of increase.
  - MOM 77% at start on Day 2 vs. 89% at start on Day 6.
  - Full feeds reach 18 days vs. 21 days. (p=0.008).
  - No effect on NEC (18% vs. 15%; stage III NEC 3 vs 5%). Less cholestasis with early feeding.



Courtesy of Nick Embleton, MD (Newcastle upon Tyne)

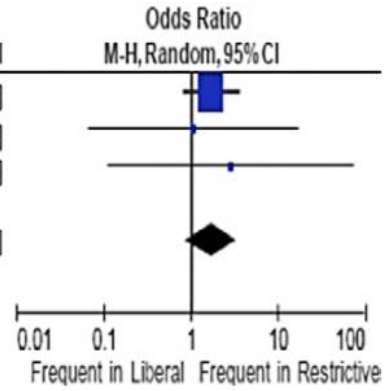
TANEC - Transfusion? Anemia ? Or Both ?

1.19 Necrotising enterocolitis

Study or Subgroup	Restrictive Events	Total	Liberal Events	Total	Weight	Odds Ratio M-H, Random, 95% CI
PINT 2006	19	223	12	228	89.0%	1.68 [0.79, 3.54]
Bell 2005	1	50	1	53	6.3%	1.06 [0.06, 17.44]
Chen 2009	1	19	0	17	4.7%	2.84 [0.11, 74.42]
Total (95% CI)		292		298	100.0%	1.67 [0.82, 3.38]
Total events	21		13			

Heterogeneity: Tau² = 0.00; Chi² = 0.20, df = 2 (P = 0.90); I² = 0%

Test for overall effect: Z = 1.42 (P = 0.15)



- More PRBC TRx for maintaining high Hgb did not result in more NEC.
- Trend towards more NEC in restrictive PRBC transfusion group.

Haresh Kirpalani, BM, and John A.F. Zupancic, MD, Seminars Perinatol 2012 Aug;36(4):269-76.

? Is there an interaction between PRBC transfusions and anemia in causing NEC

- Prospective multi-center, observational study of anemia (Hgb≤8.0g/dl), PRBC transfusions, and NEC stage II-+ (Patel RM, JAMA 2016; 315 (9)).
- 600 VLBW infants, NEC=44 (7.4%), 319 infants transfused (1430 TRx).

Feeding during PDA medical treatment

- Enteral feeding during indomethacin treatment for patent ductus arteriosus: association with gastrointestinal outcomes. Louis D et al. J Perinatol. 2016 Jul;36(7):544-8.
  - Retrospective chart review: (Group A: NPO, n=229); Group B<60ml/kg/d (n=142); Group C:>60 ml kg/d (n=44). Birth weight (A: 864±239; B: 847±202; C: 932±234 g). Postnatal age at Indomethacin (A: 5.3±2.9; B: 7.2±4.9; C: 15.4±6.6 days).
    - Primary outcome NEC (A: 6.1%, B: 7.8% and C: 4.6%, respectively)
- Enteral feeding during indomethacin and ibuprofen Rx of PDA J Pediatr. 2013 Aug;163(2):406-11. Clyman R et al.
  - Infants (N = 177, 26.3 ± 1.9 wk) were randomized at 6.5 ± 3.9 days to receive "trophic" feeds ("feeding" group, n = 81: indomethacin 80%, ibuprofen 20%) or no feeds ("fasting [nil per os]" group, n = 96: indomethacin 75%, ibuprofen 25%).
  - NEC/perforation 13% (NPO) vs. 10% (feed). Time to 120ml/kg/day 3 days earlier.