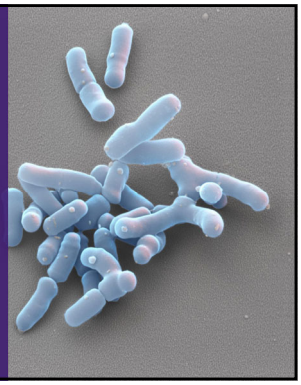
The intestinal microbiota in the first 1,000 days. Why should we care?



Kelly Tappenden, PhD, RD, FASPEN
Dean and Professor,
College of Health, University of Utah

September 10, 2024

1

Disclosures

Dr. Kelly Tappenden received an honorarium for this presentation.

Dr Tappenden is a Board Member/Advisory Panel/Speaker with:

- Abbott Nutrition Health Institute
- Dannon Institute North America
- Nutricia North America
- Takeda Pharmaceutical

The opinions reflected in this presentation are those of the speaker and independent of Nutricia.

2

Learning Objectives

- Demonstrate the importance of the intestinal microbiota in early life including it impact on the developing immune system;
- Explore data on the immune system and infections in those with food allergy;
- Illustrate the role of specific prebiotics and probiotics in the dietary management of children with food allergy;
- Discover tips for clinicians based on a growing body of evidence.

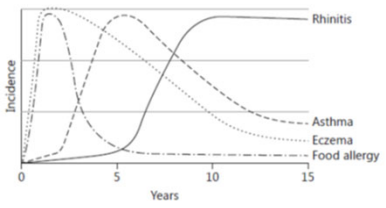
3

Audience Poll

1. Are you currently using prebiotics and/or probiotics in your clinical practice?
   - A. Yes - prebiotics
   - B. Yes - probiotics
   - C. Yes – both pre- and probiotics
   - D. No
   - E. I do not manage patients

4

Atopy comorbidities vary by age through adulthood



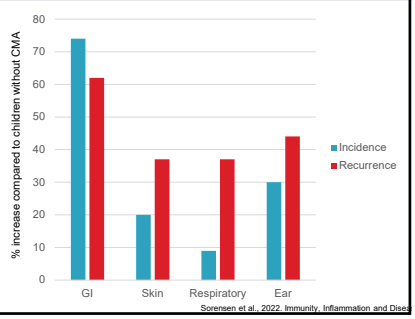
Nutten. Ann Nutr Metab 2015;66(suppl1):8-16.

https://nationaleczema.org/research/eczema-facts/

5

Cow milk allergy (CMA) is a significant burden in early childhood

MORE CHILDREN WITH CMA HAD SYMPTOMS AFFECTING MULTIPLE SYSTEMS.



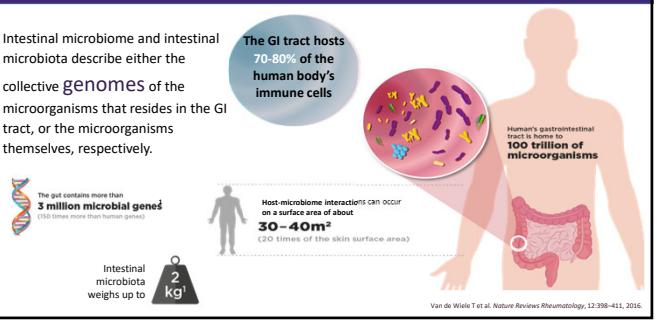
Fiocchi et al., 2021. Nutrients.

Sorensen et al., 2022. Immunity, Inflammation and Dise

6

The intestinal microbiota

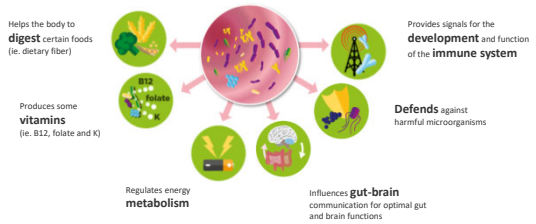
Intestinal microbiome and intestinal microbiota describe either the collective genomes of the microorganisms that resides in the GI tract, or the microorganisms themselves, respectively.



Van de Wiele T et al. *Nature Reviews Rheumatology*, 12:398–411, 2016.

7

Why is intestinal microbiota important?

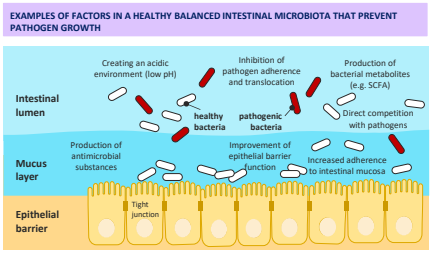


Shamir R, et al. *Essential Knowlede Briefing*, Wiley, Chichester (2015).
Van de Wiele T et al. *Nature Reviews Rheumatology*, 12:398–411, 2016.

8

The intestinal microbiota acts as a barrier against pathogens

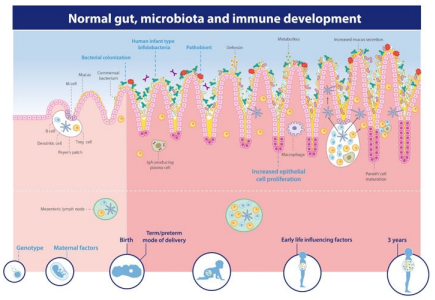
The healthy balanced intestinal microbiota acts a barrier against the infiltration and colonization and infiltration of pathogens, thereby protecting the infant against infections.



Zhang M, et al. *Front Immunol.* 2017;8:942.

9

Development of the immune system starts with the intestinal microbiota

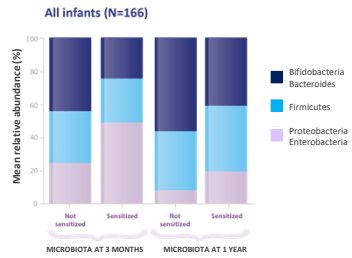


70-80% of all immune cells are organized in the gut-associated lymphoid tissue.

Immune maturation depends on intestinal microbiota signals.

10

Dysbiosis in infant intestinal microbiota precedes food sensitization



LOWER INTESTINAL MICROBIAL RICHNESS at age of 3 months is associated with INCREASED LIKELIHOOD OF FOOD SENSITIZATION by 1 year of age.

Azad et al., 2015. Clinical and Experimental Allergy

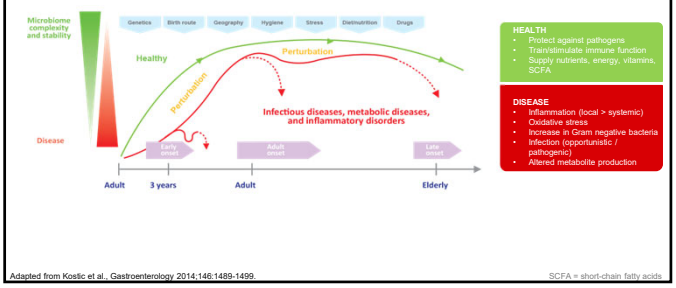
11

Dysbiosis precedes development of allergic phenotype

Age of dysbiosis	Phenotype	Age at diagnosis	Reference
1 month	Eczema	2 years	Abrahamsson et al., JACI 2012;129:434-440.
Day 7	Eczema	12 months	Ismail et al., PAI 2012:23:674-681.
1 week	Eczema	18 months	Wang et al., JACI 2008;121:129-134.
1 week/ 12 months	IgE, eos, rhinitis; NOT asthma, eczema	up to 6 years	Bisgaard et al., JACI 2011; 129:646-652.
3 weeks	Asthma		Vael et al., BMC Microbiol 2011;11:68.

12

Factors affecting stability and complexity of intestinal microbiome in health and disease

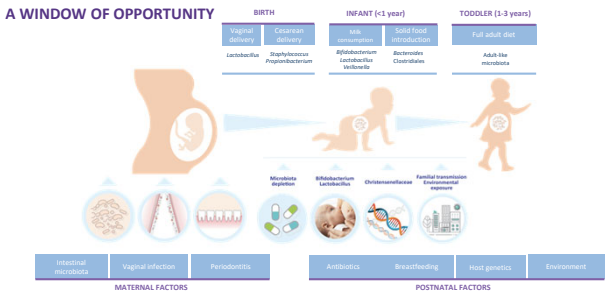


Adapted from Kostic et al., Gastroenterology 2014;146:1489-1499.

SCFA = short-chain fatty acids

13

Early intestinal microbiota development during the first 1,000 days



Adapted from Tamburini et al., *Nature Medicine*. 2016;22(7):713-722.

14

Factors disrupting microbial homeostasis during early life & development or protection against diseases

Disruptive factor	Study	Cohort characteristics	Outcomes
C-section	Sevelsted et al., 2015	1.9 million Danish term children, ages 0-15 years	Asthma, systemic connective tissues disorders, juvenile arthritis, IBDs, immune deficiencies and leukemia
	Huh et al., 2012	1,255 US children, age 3 years	Obesity, higher body-mass index and sum of skinfolds
	Eggesbø et al., 2003	2,803 Norwegian children, 0-2 years	Reactions to egg, fish or nuts, and a fourfold increase in egg allergy
Antibiotic treatment	Risnes et al., 2011 Hoskin-Parr et al., 2013	1,401 US children, ages 0-6 months 5,780 UK children, ages 0-2 years	Asthma and allergy Asthma and eczema
	Saari et al., 2015	12,062 Finnish children, ages 0-2 years	Overweight and obesity
	Schwartz et al. 2016	163,820 US children ages 2-18 years	Weight gain
	Kronman et al., 2012	9 million UK children	IBD development
Probiotics	Maldonado et al., 2012	215 Spanish children, ages 0-6 months	Reduction in gastrointestinal and upper respiratory tract infections
	Braegger et al., 2011	ESPGHAN Committee on Nutrition	Reduction in nonspecific gastrointestinal infections
Diet supplements	Zimmerman et al., 2010	Iron, 139 African children, ages 6-14 years	Intestinal inflammation, lower frequency of colic or irritability
Hygiene	Hesselmar et al., 2013	184 children, pacifier cleaning, ages 0-3 years	Lower risk of developing asthma, allergy and sensitization
Pets	Virtanen et al., 2014	3,143 Finnish children, ages 0-1 year	Reduction in risk of preclinical type I diabetes

Adapted from Tamburini et al., *Nature Medicine*. 2016;22(7):713-722.

IBD = inflammatory bowel disease

15

HUMAN milk is best for infant health

16

Human milk is best for infant health
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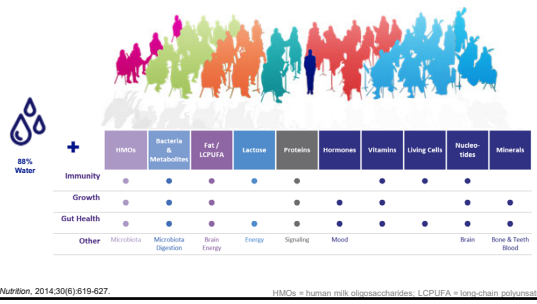
17

Human milk: a complex system with an orchestra of functions



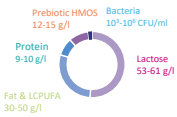
Pereira et al., *Nutrition*, 2014;30(6):619-627.

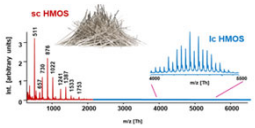
HMOs = human milk oligosaccharides; LCPUFA = long-chain polyunsaturated fatty acids

18

PREBIOTIC HMOs: a key component of human milk

PREBIOTIC HMOs consist of different structures with >162 identified





1000s of structures
Stahl et al., *Analyt Biochem* 1994;223:218-226.

162 structures identified
Urashima et al., *Trends Glycosci Glycotechnol* 2018;30(172):51-65.

Key Milestones:
- Egge 1991 FABMS of HMOS
- Stahl 1991 MALDI-Mass Spec of OS
- Thurl 1991 LC of scHMOS
- Stahl 1994 Mass Spec of sc- & lcHMOS
- Thurl 1996 4 types of HMOS based on genes
- Thurl 2017 Systematic Rev on HMOS quantities
- Mank 2019 New LC Mass Spec of Key HMOS

HMOs = human milk oligosaccharides; scHMOs = short-chain human milk oligosaccharides; lcHMOs = long-chain human milk oligosaccharides; LCPUFA = Long-chain polyunsaturated fatty acids

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PREBIOTIC HMOs: a key component of human milk

HMOs have a multitude of functions

Prebiotic Effect: Growth and Activity of Beneficial Bacteria

- Brand-Miller et al., *J Pediatr* 1998;133:95-98.
- György et al., *Eur J Biochem*, 1974;43:29.
- Roberfroid et al., *Br J Nutr* 2010; 104:S1-63.

Brain-Building Blocks

- Hart et al., *J Pediatr Psych* 2003;28:
- Wang et al., *Am J Clin Nut* 2003;78: 1024-1029.
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- Murrey et al *PNAS* 2006;103

Direct Effect on Immune Cells

- Eiwegger et al., *Pediatr Res* 2004;56:1–5.
- Bode et al., *Thromb Haem* 2004, 92:1402-10.
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Anti-Infective Effect as Receptor Analogues

- Boehm & Stahl, in: Mattila-Sandholm T(ed): *Funct. Dairy Prod.*, 2002
- Newburg et al., *Glycobiology*, 2004, 14
- Coppa GV et al *Lancet* 1990

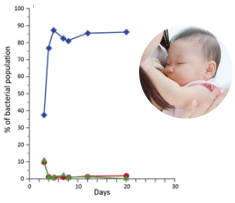
HMOs = human milk oligosaccharides

20

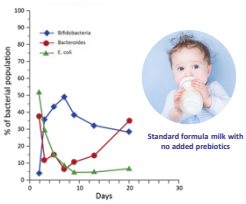
Microbiota: breast vs. bottle

HIGHER BIFIDOBACTERIA COUNT IN BABIES WHO BREASTFEED

Breastfeeding



Formula Feeding



Standard formula milk with no added prebiotics

Harmsen et al., *J Pediatr Gastroenterol Nutr.* 2000;30:61-67.

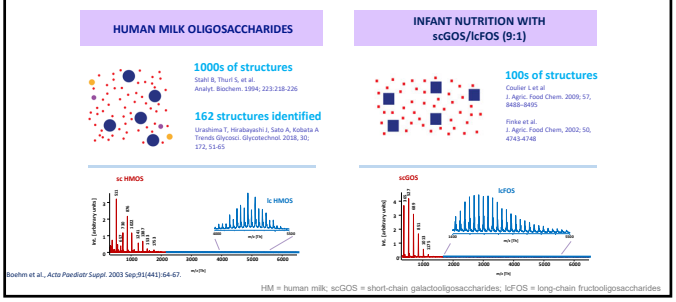
21

Synbiotics = prebiotic + probiotic

Can nutritional formulas be modified
- using a **SYNbiotic** approach -
to alter the intestinal microbiota and
improve clinical outcomes in children?

22

Prebiotics in HM and infant formula with scGOS/lcFOS



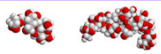
Boehm et al., *Acta Paediatr Suppl.* 2003 Sep;91(441):64-67.

HM = human milk; scGOS = short-chain galactooligosaccharides; lcFOS = long-chain fructooligosaccharides

23

Prebiotics and bifidogenic intestinal colonization

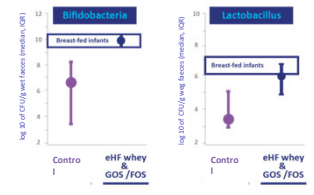
scGOS & lcFOS



- 6 clinical trials
- With a specific mixture of short-chain galacto-oligosaccharides (scGOS) and long chain fructo-oligosaccharides (lcFOS)
- In a ratio 9:1 in infant milk formulas
- Showed consistent positive effects on stool consistency and stool frequency

Reviewed in Scholtens et al., *World Gastroenterol* 2014

EFFECT ON INTESTINAL MICROFLORA (TERM INFANTS AFTER 28 DAYS FORMULA FEEDING)



Moro et al., *J Ped Gastroenterology & Nutrition* 2002

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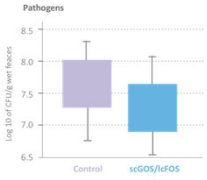
scGOS/lcFOS supports intestinal microbiota by discouraging the growth of potential pathogens

Target population
Preterm

Conclusions
Supplementation of a preterm formula with scGOS/lcFOS sign decreases the sum of pathogens. Also, the sum of pathogens as % of total bacterial count was lower than control.

Study design
25 preterm infants
- 0.0g scGOS/lcFOS [Control] (n=15)
- 1.0 g/100ml scGOS/lcFOS (n=12)



scGOS/lcFOS sign reduces the number of clinically relevant pathogens in stools of preterm infants

Knol, J. et al. (2005). Acta Paediatrica;94 (Suppl 449):31–33.

scGOS = short-chain galactooligosaccharides; lcFOS = long-chain fructooligosaccharides

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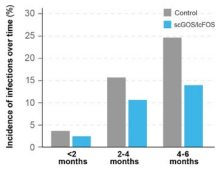
scGOS/lcFOS reduces infections during the first 6 months of life

Target population
Healthy infants

Conclusions
0.8 g/100ml scGOS/lcFOS reduced the number of infectious episodes during the first 6 months of life.

Study design
Randomized, double blind, controlled study; Healthy, term infants with parental history of atopic eczema, allergic rhinitis, or asthma:
- 0.0 g scGOS/lcFOS [Control] (n=104)
- 0.8 g/100ml scGOS/lcFOS (n=102)



scGOS/lcFOS results in lower incidence of infections over time

Arslanoglu, et al. J Nutr 2007;137:2420-2424.

scGOS = short-chain galactooligosaccharides; lcFOS = long-chain fructooligosaccharides

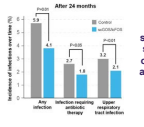
26

scGOS/lcFOS reduces infections and incidence of allergic manifestations during the first 2 years of life

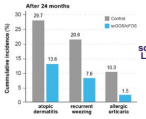
Target population
Healthy infants

Conclusions
Early nutritional intervention with 0.8 g/100ml scGOS/lcFOS is effective in priming the infant's immune system, providing substantial protection against both allergy and infection.

Study design
Randomized, double blind, controlled study; Healthy, term infants with parental history of atopic eczema, allergic rhinitis, or asthma; for 2 years:
- 0.0 g scGOS/lcFOS [Control] (n=68)
- 0.8 g/100ml scGOS/lcFOS (n=66)



scGOS/lcFOS results in sign. Lower number of overall infections, URTI and infections requiring antibiotics.



scGOS/lcFOS results in sign. Lower cumulative incidence of atopic dermatitis, wheezing and allergic urticaria.

Arslanoglu, et al. J Nutr 2008;138:1091-1095.

scGOS = short-chain galactooligosaccharides; lcFOS = long-chain fructooligosaccharides; URTI = upper respiratory tract infections

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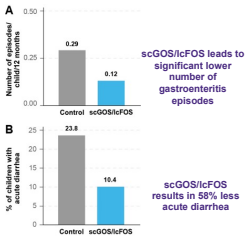
scGOS/lcFOS reduces intestinal infections

Target population
Healthy infants

Conclusions
These data show that scGOS/lcFOS 0.4 g/100 ml reduced intestinal infections in healthy infants during the first year of age.

Study design
A prospective, randomized, controlled, open trial.
Healthy infants aged between 15 and 120 days were enrolled in two intervention groups; formula feeding for 12 months
- 0.0 g scGOS/lcFOS [Control] (n=105)
- 0.4 g/ 100mL scGOS/lcFOS (n=96)



scGOS/lcFOS leads to significant lower number of gastroenteritis episodes

scGOS/lcFOS results in 58% less acute diarrhea

Bruzzese, et al. *Clinical Nutrition* 2009;28:156-161

scGOS = short-chain galactooligosaccharides; lcFOS = long-chain fructooligosaccharides

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AAF with specific synbiotic blend aims to eliminate allergens for active management of CMA while helping to target intestinal dysbiosis

maximal allergen elimination + **helps to address underlying intestinal dysbiosis**

- Hypoallergenic formula
- Protein source: 100% amino acids
- 0-12 months of age

- **scFOS / lcFOS** (9:1 ratio)
- 0.60 g / 100 ml
- no GOS, to avoid cow milk protein contamination

- ***Bifidobacterium breve* M-16V**
- 10^8 *CFU*/g powder
- processed in a milk protein-free environment

AAF = amino acid-based formula; CMA = cow milk allergy; scFOS = short-chain fructooligosaccharides; lcFOS = long-chain fructooligosaccharides

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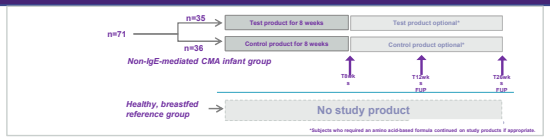
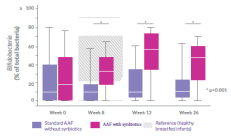
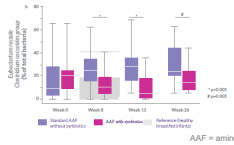
AAF + specific synbiotic blend promotes sustained bifidobacteria growth and reduces *Eubacterium / Clostridia*, similar to breastfed infants



***Bifidobacterium* species in fecal microbiota**



***E. rectale / C. coccoides* cluster in fecal microbiota**



Fox, et al. Clin Transl Allergy. 2019;9:5

AAF = amino acid-based formula

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Meta-analysis of AAF + specific synbiotic blend

Lead Author (Date)	Population and Type of Study	Male	Mean Age (Months)	Amount of Formula Consumed/Day (mL) Mean ± SD	n AAF-Syn	n AAF	Intervention Duration	Timepoint Outcomes Measured
Harvey (2014) [39] Full paper	Infants with IgE mediated CMPA aged 0-36 months. One arm DBPCCFC and 7 day feeding period	61%	17.3, range 3.5–46.9	Not reported	30	30	7 days	7 days
	Full-term healthy infants aged 3-16 months, RCT [¶]	67%	10.6, range 3–16	AAF-Syn: 349 ± 127 [§], AAF: 332 ± 124 [§]	59	56	16 units	2, 4, 8, 12 & 16 units
Burks (2015) [29] Full paper	Infants with IgE or non-IgE mediated CMPA aged 0–8 months, RCT	62%	4.5, range 0.6–8.9	Not reported. Intake was reported as comparable in both groups	54	56	16 weeks	4 & 16 weeks
Candy (2018) [32] ASSIGN study, full paper	Infants with non-IgE mediated CMPA aged 0–13 months, RCT Included breast-fed healthy reference group (not randomised)	73%	6, range 1.2–12.8	Week 8 AAF-Syn 652 ± 176; AAF 639 ± 212	35	36	8 weeks	4 & 8 weeks
Fox (2019) [33] [¶] ASSIGN study, full paper	Infants with non-IgE mediated CMPA aged 0–13 months 26-week follow-up of Candy (2018)	73%	6, range 1.2–12.8	Week 8 AAF-Syn 652 ± 176; AAF 639 ± 212	35	36	8 weeks	8, 12 & 26 weeks
Wopereis (2019) [40] [¶] ASSIGN study, full paper	Infants with non-IgE mediated CMPA aged 0–13 months Gene-sequencing analysis from Candy (2018) and Fox (2019)	73%	6, range 1.2–12.8	Week 8 AAF-Syn 652 ± 176; AAF 639 ± 212	35	36	8 weeks	8, 12 & 26 weeks
Chatchatee (2019) [31] PRESTO study [‡], conference abstract	Infants with confirmed IgE mediated CMPA aged 0–13 months, RCT	72%	9.36, SD 2.53	At 12 months: AAF-Syn: 347 ± 302; AAF: 530 ± 309	80	89	12 months	12 months
Wopereis (2020) [30] PRESTO study [‡], conference abstract	Infants with confirmed IgE mediated CMPA aged 0–13 months, RCT							

CMPA: cow's milk protein allergy; RCT: randomised controlled trial; DBPCCFC: double-blind placebo-controlled crossover food challenge; AAF-Syn: amino acid formula with synbiotics (Nocuate Syneo®, Nutricia); AAF (control): amino acid formula (Nocuate LCP®, Nutricia); [‡] original RCT (Candy 2018 [32]) was for 8 weeks, Fox (2019) [33] and Wopereis (2019) [40] were published after original RCT. † Chatchatee (2019) [31] and Wopereis (2020) [30] report different outcomes from same study; [§] Converted from reported oz/d (13.8 ± 4.3 oz/d AAF-Syn; 13.2 ± 4.2 oz/d AAF); [¶] Included in table for completeness, but outside scope of review as subjects were not required to have CMPA.

Sorensen et al., Nutrients, 2021;13:935-954. AAF = amino acid-based formula

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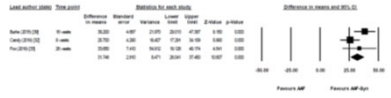
Meta-analyses reveal consumption of AAF + specific synbiotic blend enhance microbiota community

- Increased percentages of fecal infant-like bifidobacterial species with AAF-Syn



- Lower percentages of adult-like *Eubacterium rectale* and *Clostridium coccoides* species with AAF-Syn

Sorensen et al., Nutrients, 2021;13:935-954. AAF = amino acid-based formula; AAF-Syn = amino acid-based formula with synbiotics

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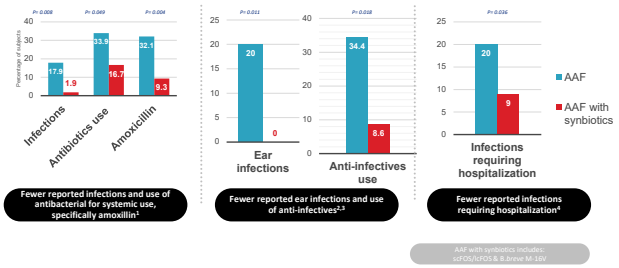
Evaluation of adverse events: 3 RCTs with AAF with synbiotics show a reduction in reports of infections & antibiotic usage



1. Burks, et al. Pediatr Allergy Immunol. 2015;26:316-22. 2. Candy, et al. Pediatr Res. 2018;83:677-86. 3. Fox, et al. Clin Transl Allergy. 2019;9:5. 4. Chatchatee, et al. J Allergy Clin Immunol. 2022;149:650-8.e5.

RCTs = randomized controlled trial; AAF = amino acid-based formula

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Multiple studies demonstrate consumption of AAF with synbiotics improve clinical outcomes

Compared to AAF, significantly fewer infants fed AAF-Syn had infections*

Overall medication use,* including antibacterials and anti-infectives, was lower among infants fed AAF-Syn.

Significantly fewer infants had hospital admissions (arising from infections) with AAF-Syn compared to AAF (8.8% vs. 20.2%, p = 0.036; 56% reduction), leading to potential cost savings† per infant of £164.05–£338.77.

*Exploratory findings, from component studies (not powered to test these outcomes), were the results of safety evaluations.
†Cost savings based on UK hospital admission costs. Costs may vary in the US.

Sorensen et al., Nutrients, 2021;13:935-954.

AAF = amino acid-based formula; AAF-Syn = amino acid-based formula with synbiotics

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Conclusion

Nutritional strategies employing
PRObiotics + PREbiotic fiber

– hence SYNbiotics –

are important for addressing dysbiosis of the developing intestinal microbiota and stimulating critical development of the immune system in early life.

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We need to consider (and feed) the complex ecosystem

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