Pediatric Nutrition
CONTINUING EDUCATION FOR CLINICIANS

COURSE TRANSCRIPT

Individualized Fortification of Breast Milk for Preterm Infants

Miami Neonatology 2018 – 42nd Annual International Conference

Overview

Mother's breast milk is notably considered the gold standard for infants' nutrition. However, **Christoph Fusch, MD, PhD**, discusses evidence that reveals the macronutrient composition from breast milk is variable and may not provide adequate nutrients for the essential growth and neurodevelopment needed in preterm infants. The challenge with enteral nutrition is determining how to achieve a healthy balance between appropriate nutritional intake while also reducing the risk of sepsis and necrotizing enterocolitis (NEC) in preterm infants in order to achieve good growth and developmental outcomes. Dr. Fusch reviews the benefits and draw backs of mother's milk, donor milk, and fortified infant formula in premature infants, as well as evaluating the optimum composition of infant milk fortification.

Content Areas

- Discern enteral nutrition needs of preterm infants
- Evaluate postnatal growth in preterm infants
- Recognize that neurodevelopmental outcome relies on nutrient intake of sufficient protein, calories, and minerals
- Understand carbohydrate-to-fat ratio that influences the rate and quality of growth in preterm infants
- Select the right infant formula for optimum composition of fortification

Target Audience

This activity was developed for pediatric physicians, nurses, nurse practitioners, dietitians, allergists and other health care providers who have an interest in newborns, infants and toddlers.

Obtain your CE/CME credit at:
https://pnce.org/IndividualizedFortification-CME

Learning Objectives

At the conclusion of this activity, participants should be better able to:

- Assess the benefits and optimum compositions of fortified human milk and formula in preterm infant enteral nutrition
- Develop individualized, evidence-based nutritional strategy for preterm infants to include the protective effect of breast milk, and provide appropriate growth and neurodevelopment rates

Faculty

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Christoph Fusch, MD, PhD

Research Support	Prolacta Bioscience – clinical area: nutrition and growth
Consultant	Baxter – clinical area: nutrition and growth Hamilton Medical – clinical area: ventilation and respiratory support
Speakers Bureau	Nestlé - clinical area: nutrition and growth Milupa (Danone) - clinical area: nutrition and growth Baxter – clinical area: nutrition and growth Prolacta Bioscience – clinical area: nutrition and growth

The faculty for this activity has disclosed that there will be no discussion about the use of products for non-FDA approved indications.

Additional content planners

The following have no significant relationship to disclose:
Erin Allen, MS, RD, LDN (RD reviewer)
Victoria Anderson (medical writer)
Heather Marie Jimenez, FNP (nurse reviewer)

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Individualized Fortification of Breast Milk for Preterm Infants

This activity is supported by an independent educational grant from **Mead Johnson Nutrition**.

This activity is an online enduring material. Successful completion is achieved by reading and/or viewing the materials, reflecting on its implications in your practice, and completing the assessment component.

The estimated time to complete the activity is 1.0 hour.

This activity was released on March 15, 2019 and is eligible for credit through March 15, 2021.

Contact Information

For help or questions about this activity please contact Continuing Education:

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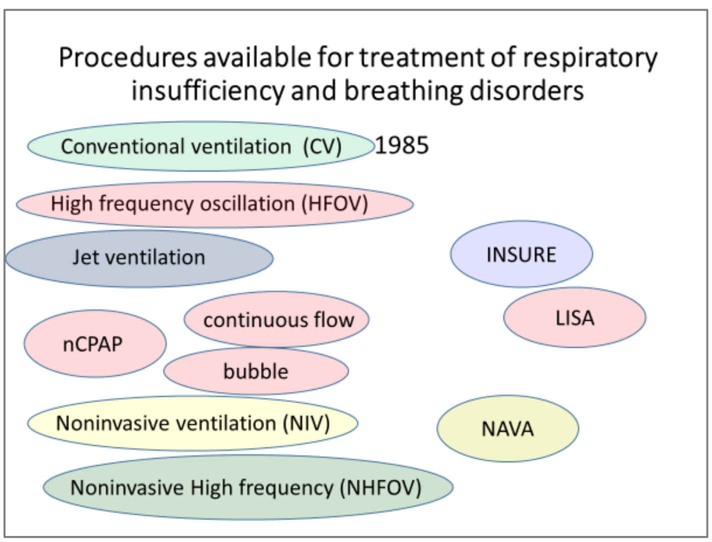
Editor's Note: This is a transcript of the live presentation from Miami Neonatology on November 14, 2018.

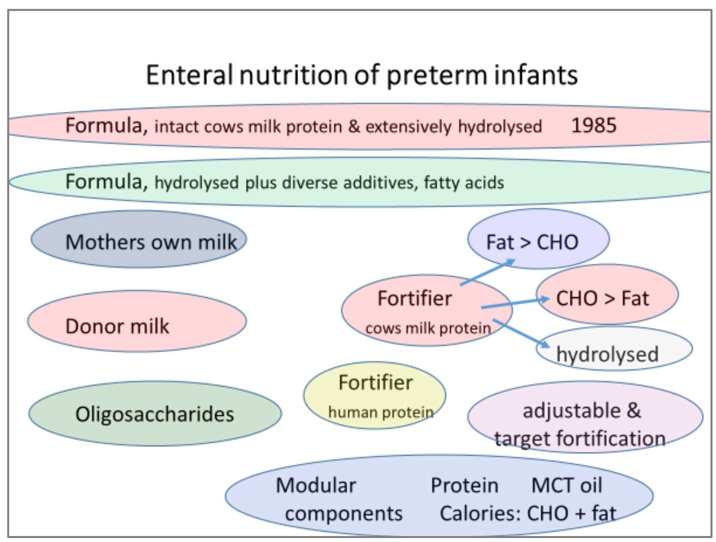
Dr. Cristoph Fusch: This [Slide 1] shows you procedures available for the treatment of respiratory insufficiency and breathing disorders. In 1985, we had conventional ventilation, which we were using in babies. Then we got an armada of new procedures— [some of which] you see here. There's lots of research on this. When should we use what? We have heard a few things about nasal CPAP, LISA, and so on. [Is this] the wrong talk? No, it is not.



Slide 1

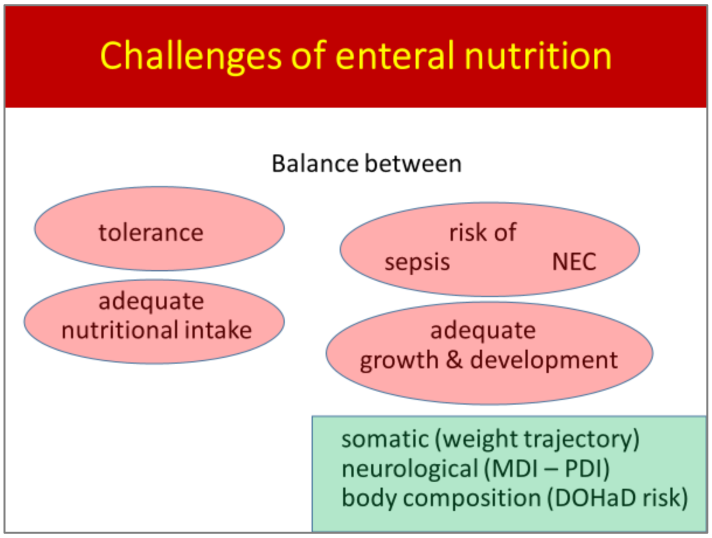
Enteral nutrition of preterm infants, [in] 1985, we had formula, [which included] intact cow's milk protein, and maybe extensively hydrolyzed [formula]. That was all we had. Rarely was someone feeding human milk to preterm babies.

Now, 30–33 years later, it looks like with ventilation, we have lots of new products, lots of new strategies. We need to work with these new products and do [more] research, to see which [will] grow babies best. There is also, maybe, a generational issue, moving away from ventilator lung towards growth. Both need to be done to get the right product: a good baby, at the end.



Slide 2

Challenges of enteral nutrition are to achieve a balance between—when we implement it—tolerance and adequate nutritional intake, thereby reducing the risk of sepsis and necrotizing enterocolitis (NEC), and still achieving, at the end, good growth and developmental outcomes.



Slide 3

I would define this as somatic, like weight trajectories; neurological, with [mental development index] MDI, [physical development index] PDI; but also body composition, because it's not only growth, but weight gain, that matters. We need to gain weight with[in] the right body compartments to reduce the risk for DOHaD, which is developmental origins of health and disease. This affects typically men my age, earlier or later, and to reduce this also in preemie babies, also for female preemie babies.

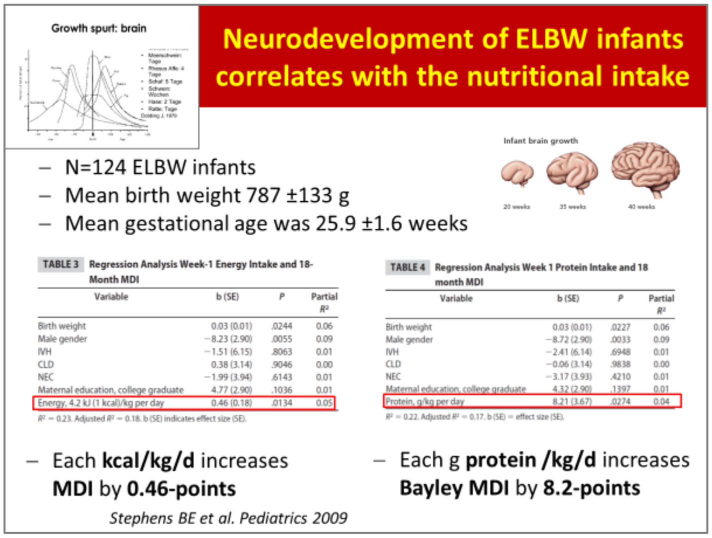
This is a comparison of different nutritional regimes that have different effects on outcome categories, and it's all summarized here [Slide 4]. So, I can stop the talk here, but I won't.

Comparison of different nutritional regimes: differential effects on outcome categories

	NEC/ Sepsis/Death	Growth	Neuro-development
Formula	-	+	+/-
HM			
MOM (native)	+	--	-
Donor (native)	+	--	-
HM + HMF (bovine)	+/-	+	+
Donor + HMF (bovine)	+/-	+/-	+/-
HM + HMF (human)	+	+	(+)
HM + TF	+/-	++	?
Formula + HMO	+	+	?

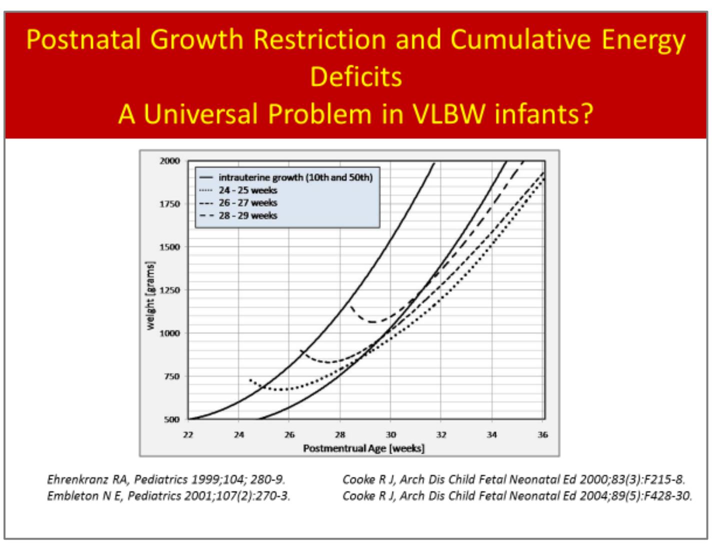
Slide 4

Apparently, neurodevelopmental outcome has to do with growth and with feeding, with nutrient intake. This study [Slide 5] from Bonne E. Stephens, MD, et al,[1] which is very frequently quoted, showed that early nutritional intake increases the MDI if you manage to bring the kcal up. Each kcal that you achieve during the first week of life increases your MDI by 0.46, and each g protein/kg/d will increase your MDI by 8.2. That is significant. Others have shown in studies, as well, that nutrition matters because it affects brain growth. And, with a bigger brain, at the end, you have better capacity, and fortunately it is currently as easy as that.



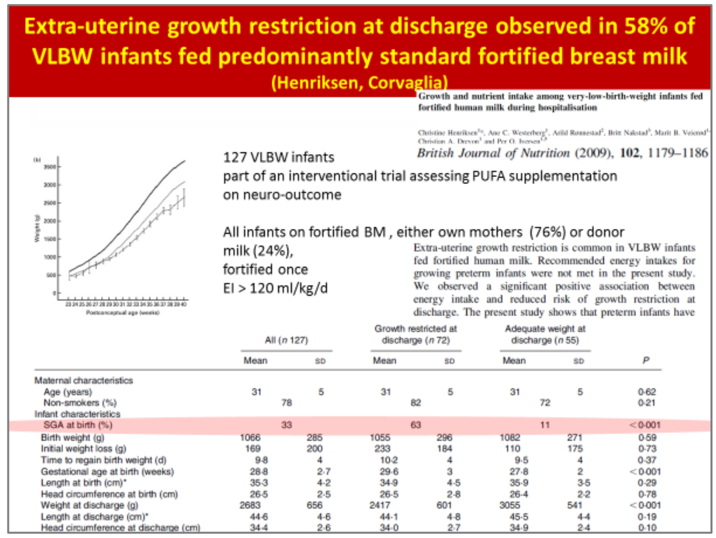
Slide 5

How do babies grow? This is a slide [Slide 6] from Richard Ehrenkranz, MD, to whom I would like to devote this lecture. It shows these are the reference curves (intrauterine), and we frequently achieve growth curves like this.[1] So, this is a severe risk of postnatal growth restriction due to cumulative energy deficits. This is maybe not the way babies should grow in the next 20 years.



Slide 6

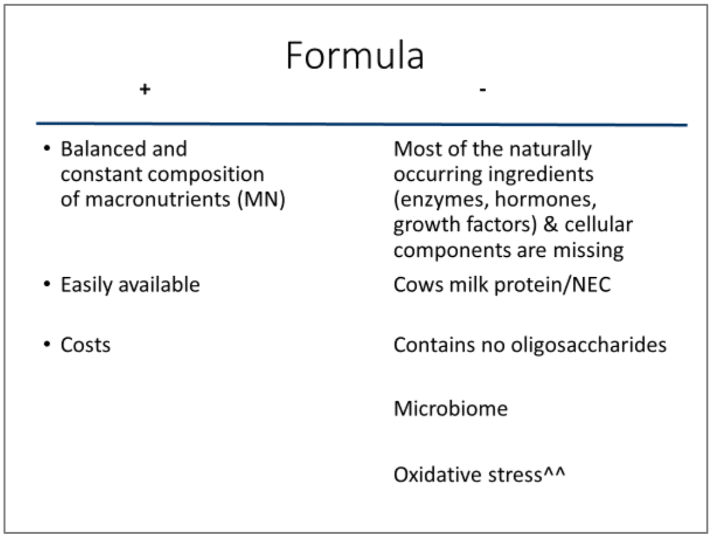
In this study, you see here [Slide 7] was done in [very low birth weight] VLBW babies due to another reason that they were SGA, small for gestation age, at admission in 33%, and at discharge, in 63%. So, you see here again [top left], growth curves like this.[2]



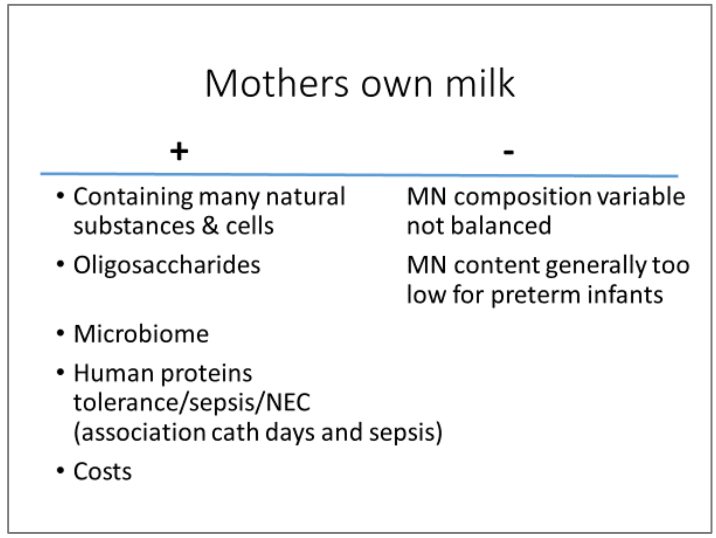
Slide 7

We are currently working on individualized trajectories, putting a few physiological observations together, creating individualized trajectories for babies to come out of this "no man's land" here, 'How to know to grow,' but I can't elucidate on that due to time constrictions. I will go now to the few different products we have to [help] achieve postnatal growth. On one hand, we can use formula in babies. The good thing on formula is it's balanced and a constant composition of macronutrient. It's easily available and costs are relatively low.

Individualized Fortification of Breast Milk for Preterm Infants



Slide 8

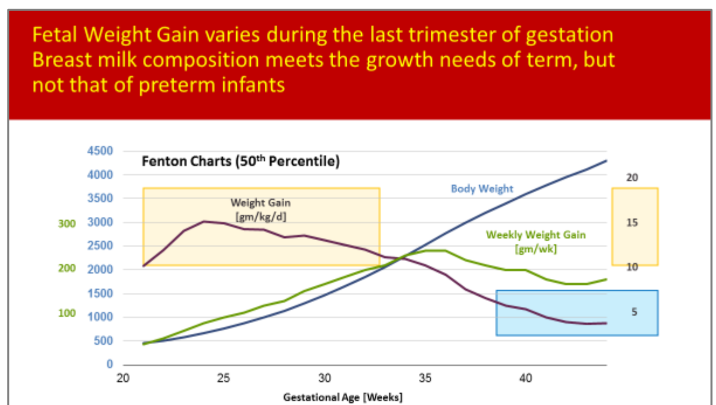


Slide 9

On the negative side [Slide 8, right], most of the natural occurring ingredients (ie, enzymes, hormones, growth factors and cellular components), are missing. And, it's based on cow's milk protein and has an association with the risk of NEC. It doesn't contain oligosaccharides. We have heard a little bit about microbiomes and how important oligosaccharides are to introduce, or to establish the right microbiome that might be protective also for NEC. Formula also causes more oxidative stress than human milk.

Mother's own milk contains many natural substances; [it] contains these oligosaccharides, which is good for the microbiome. The human proteins lead to a better tolerance, lower sepsis, and lower NEC rates; therefore, also to less catheter days and sepsis. Costs are still relatively low. The product itself is for free, but the handling and the storage might cost some money. However, **macronutrient composition is very variable in mother's own milk and is not balanced; and macronutrient content is generally too low for preterm infants.**

You see here [Slide 10], growth curves, gestational age here, weight, weight per week, and weight in g/kg/d. And you see, here, that term babies have a growth rate of about 5 g/kg/d [shaded blue], whereas preterm babies have 15–17 to 20 g/kg/d [shaded yellow]. So, much higher.



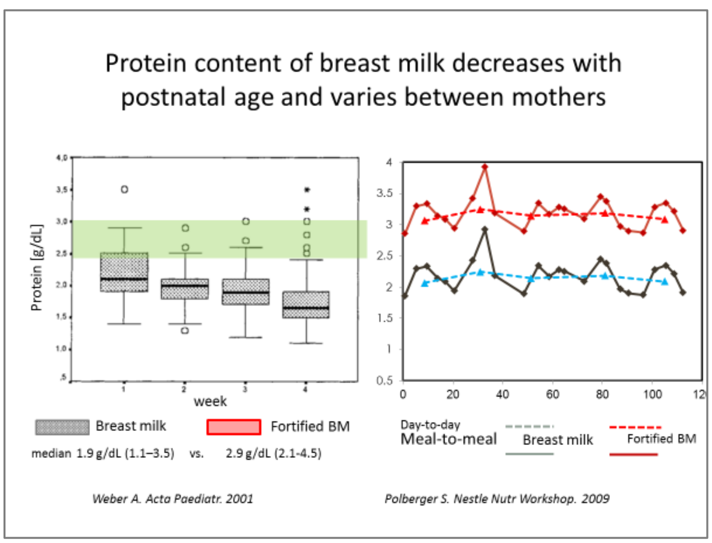
	Growth Velocity [g/kg/day]	Protein Intake [g/kg/day]	Protein Content of Milk [g/100 ml]
Preterm Infants 24-32 Weeks	(12 –) 18	3 – 4.5	2 – 2.9
Term Infants	(3 –) 8	1.5 – 2	1 – 1.3

Slide 10

Protein intake and growth velocity are very tightly correlated (I will come to that a little later). If you want to achieve 3–8 g/kg/d of growth velocity, then you need to have a protein intake of 1.5–2 g/kg/d. If you're assuming an intake of 150 ml/d, then the protein content of that milk that you are feeding to the baby would need to have 1–1.3 g/dL of protein, and that is exactly the composition of breast milk.

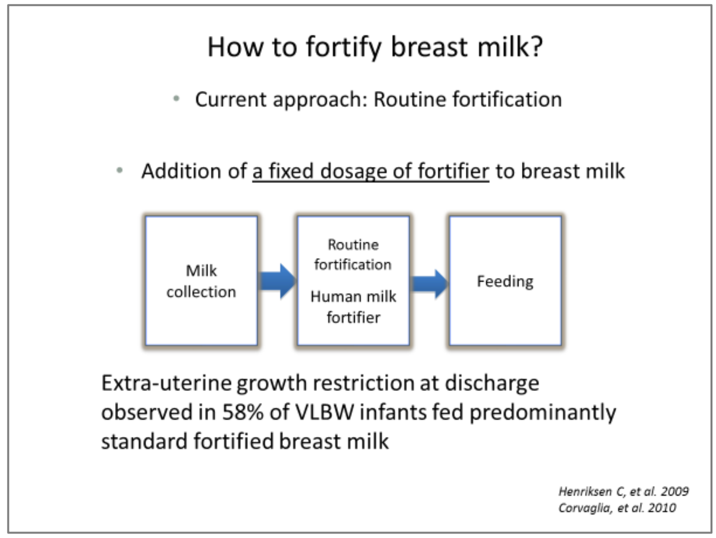
However, **preterm babies with a higher growth velocity need a much higher protein intake, up to 3 g/100 ml, which is basically not available in human milk.**

What you see here [Slide 11] is data on the composition of breast milk, protein content in weeks of lactation; so, the week 1, 2, 3, 4 after birth. You see on one hand that the protein content decreases during the first few weeks, and there's a huge inter-individual—but also intra-individual—variation, but most concerning is the inter-individual variation.[3] The green shaded areas are the ESPGHAN [European Society for Pediatric Gastroenterology Hepatology and Nutrition] recommendations to make preterm babies grow the right way.



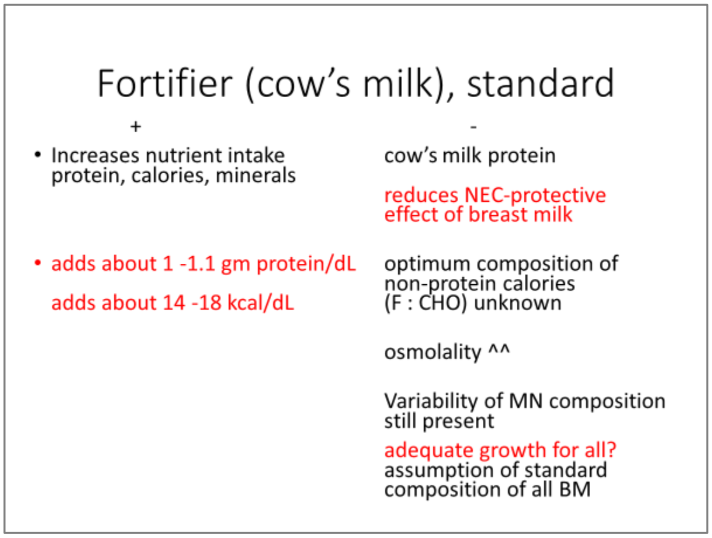
Slide 11

So, how do we get out of this? Everybody does it. We are doing routine fortification using human milk fortifiers. Basically, this work has been done in the '80s and '90s. The products have developed since then but have not improved a lot. They are basically based on a standard—assuming a standard composition of breast milk. If you have this standard composition of breast milk, then everything is fine. But if you don't, then things are not so good any longer for the preterm babies.



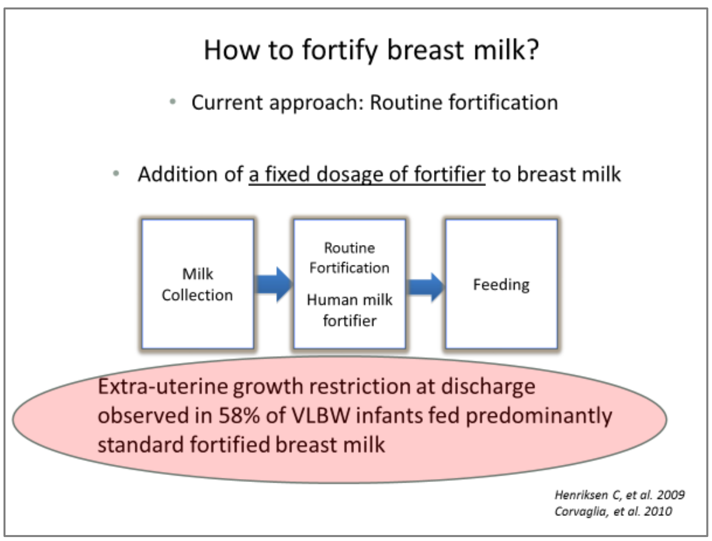
Slide 12

How are these fortifiers [composed]? What are their properties? They increase nutrient intake, because they add extra protein, extra calories and minerals. They add about 1–1.1 gm protein/dL, and they add about 14–18 kcal/dL. But, they are based on cow's milk protein; therefore, they reduce the NEC-protective effect of breast milk. The optimum composition of non-protein calories, fat vs carbohydrates, is unknown (I'll come to that later). There are concerns about osmolality, which I think are very weak because the basis of osmolality recommendation is extremely weak. The variability of the macronutrient composition is still present. The big question is, is it adequate for all? Because we are assuming the standard composition of breast milk.



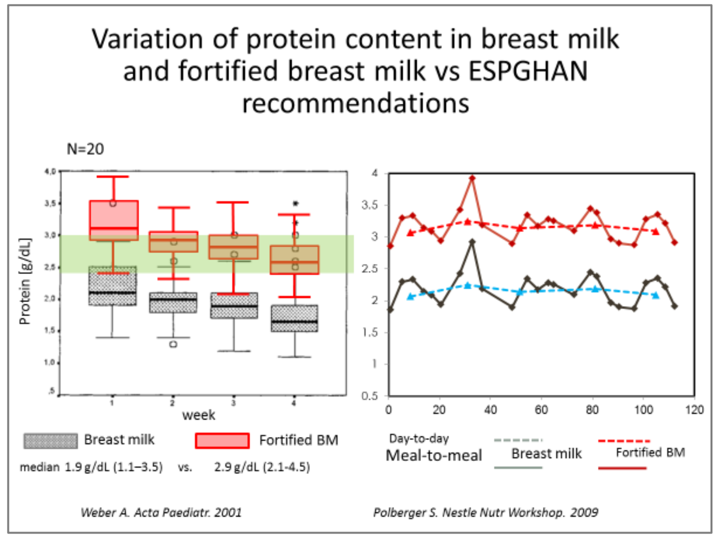
Slide 13

If we look at growth under human milk fortifiers, we still see that 58% of the VLBW infants fed predominantly standard fortified breast milk do not grow well.[4,5]



Slide 14

What does the standard fortification do? Basically, it shifts up these variations by this 1–1.1 gm/dL, and you see that for the first week, it's fine. Second week, third week is fine, but here [into week 4] already you see a significant amount of milk samples and mothers who have a protein concentration, despite standard fortification, that is not sufficient.

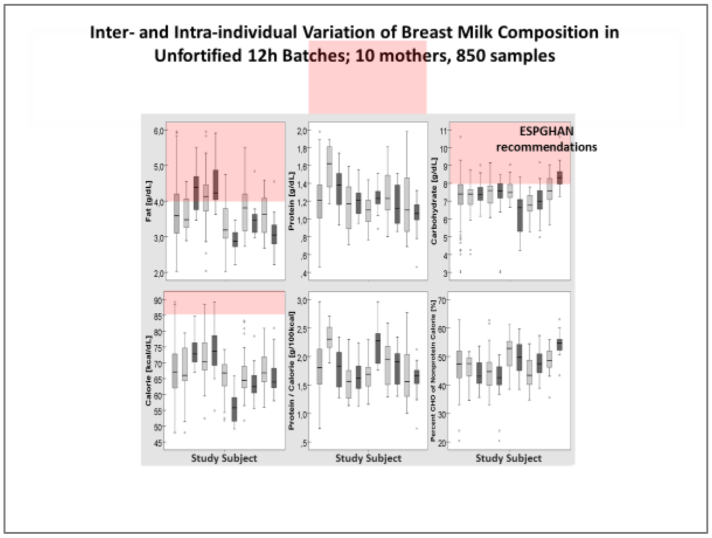


Slide 15

And we are here only on week 3 and 4, and for a 24-weeker, week 4 would be 28 weeks, and that's still a long way to go until term.[4] So, basically the babies here would be depleted with protein intake.

These are data [Slide 16] from our own study where we had 10 mothers from an overall sample of 850 using 12-hour batches, and you see for the different macronutrients here,[6] fat, protein and carbohydrate, the huge variation. The shaded areas are the ESPGHAN recommendations. You see here calories; you see here protein-to-calorie ratio (I will come to that later). That is very important. This is the carbohydrate-to-fat ratio in the non-protein energy, which is also extremely important. It might explain some of the observations that Paul Rozance, MD, did about carbohydrate depletion[7]—what you do if you put insulin into an organism.

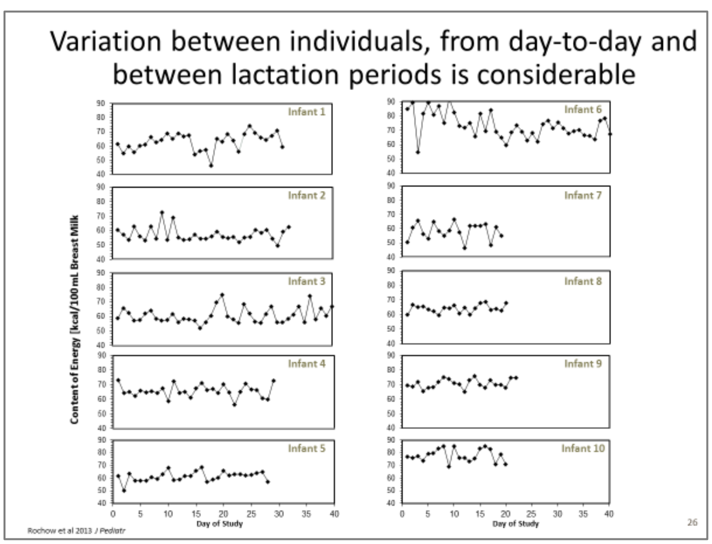
Individualized Fortification of Breast Milk for Preterm Infants



Slide 16

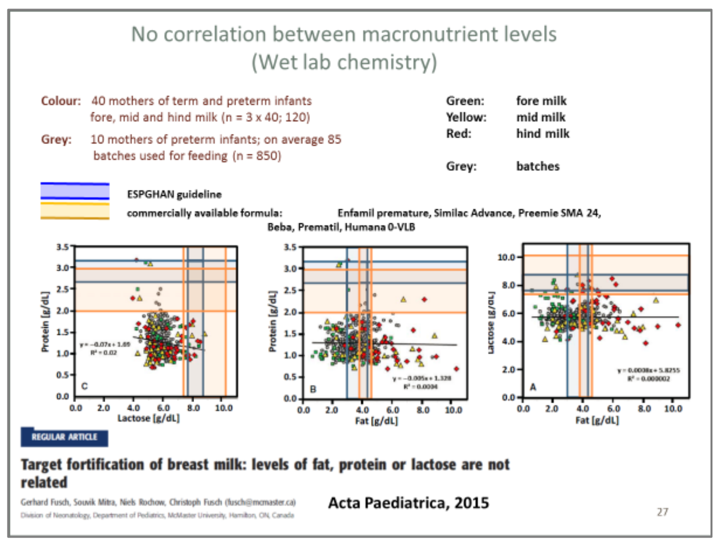
Here are data [Slide 17] on calories. And you see here that this mom produces a breast milk with 65 kcal/100 ml [Infant 8].[8] That is the assumption. This one is close to it, as well [Infant 4]. This mom behaves according to protocol.

This mom, here [Infant 6], does not behave according to protocol. She produces a rich breast milk, 90 kcal, and stays [at day 40] still at 70 kcal/100 ml. Here [With Infant 5] and here [Infant 3] we are seeing that we have only 55 kcal/100 ml, so that is very depleted breast milk which is fed to the baby.



Slide 17

I first thought having all this data together that Mother Nature produces either a rich breast milk or a poor diluted breast milk—it's a matter of dilution. When we did the x-, y-plots of the different macronutrients against each other, we were blown away, and I think that has never been shown before about the huge variability. That is shown in this paper, which we published [in] 2015, *Acta Paediatrica*.[9] What you see here [Slide 18] in these 850 samples plus 120 samples, 3 times from 40 moms, fore, mid, and hind milk, [plot 1] lactose vs protein, which is all over the place. There is not a diluted or undiluted milk, the same for fat and protein [plot 2] and for fat and lactose [plot 3]. There are moms who produce milk that is rich in protein but poor in energy. There are moms who are producing breast milk that is poor in protein and rich in energy. All these babies cannot grow. I will show you, a little later, why. The same is true for fore, hind, and mid milk.

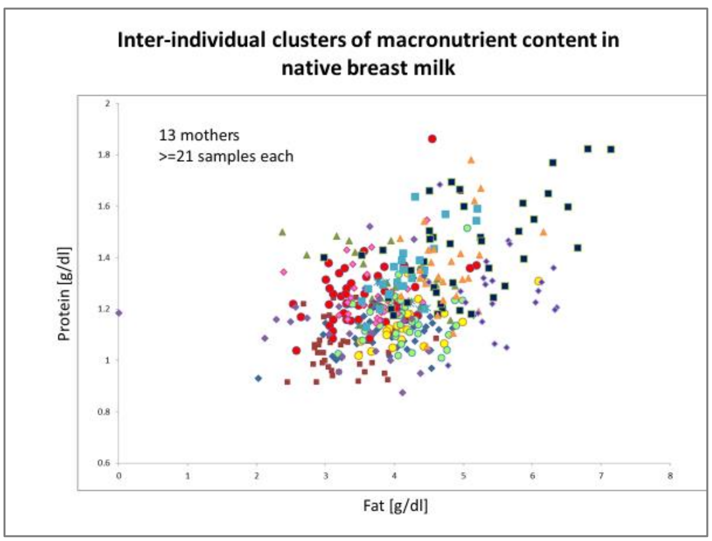


Slide 18

Individualized Fortification of Breast Milk for Preterm Infants

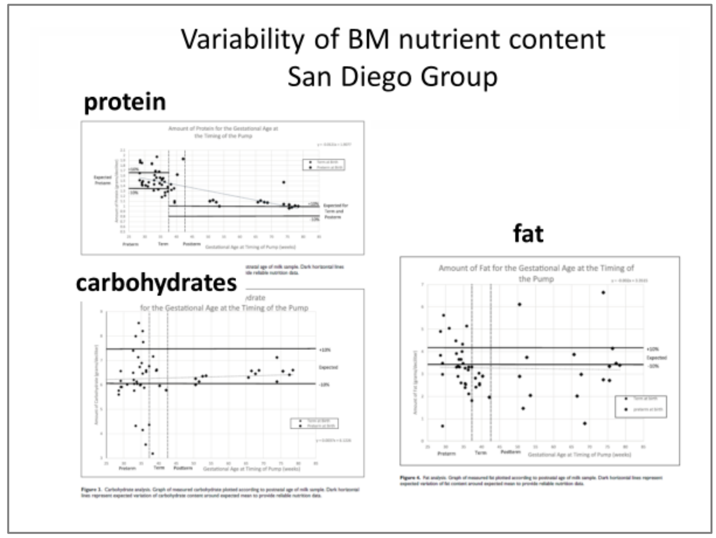
"There are moms who produce milk that is rich in protein but poor in energy. There are moms who are producing breast milk that is poor in protein and rich in energy. All these babies cannot grow."

What you see here [Slide 19] is 13 mothers who had at least 21 samples. [Fat is plotted on the x-axis; protein along the y-axis] These mothers produce milk that is significantly different from [each other]. I'll come to this graph a little later. Basically, this is energy [x-axis]—fat is a major determinant for energy—and this [y-axis] is protein intake.



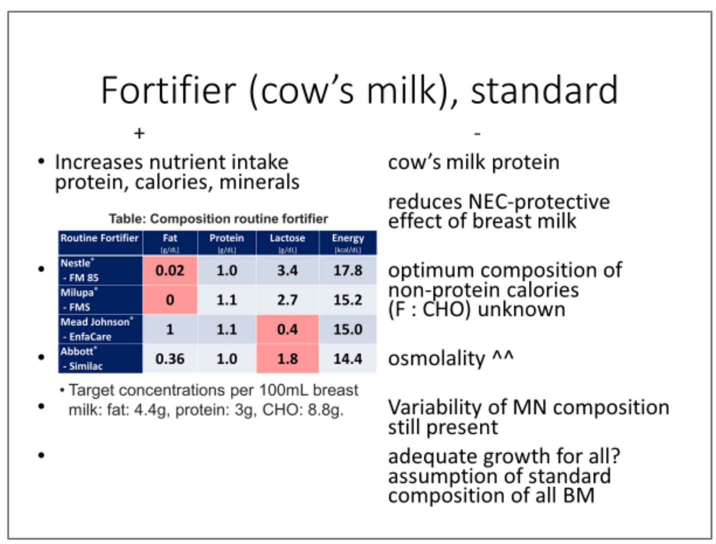
Slide 19

We're not the only ones, fortunately, who have looked into this. Recently, Jae Kim, MD, PhD, from San Diego, published a paper last year[10] that basically confirms what we have published in *Acta Paediatrica*.[11]



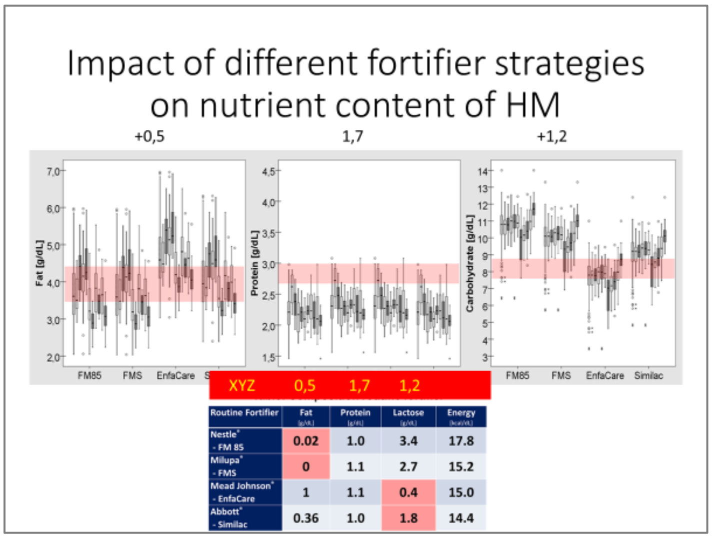
Slide 20

Fortifiers now are composed in different ways. Actually, we have 4 products, and there are 2 mainly used in Europe, 2 mainly used in North America. You see that the composition for protein is the same, but to gain the extra fat, the North American fortifiers are heavily fat-based, nearly free from lactose (only a little bit), whereas the European ones are not using fat, but achieve everything with lactose. The question is, where does it lead to?



Slide 21

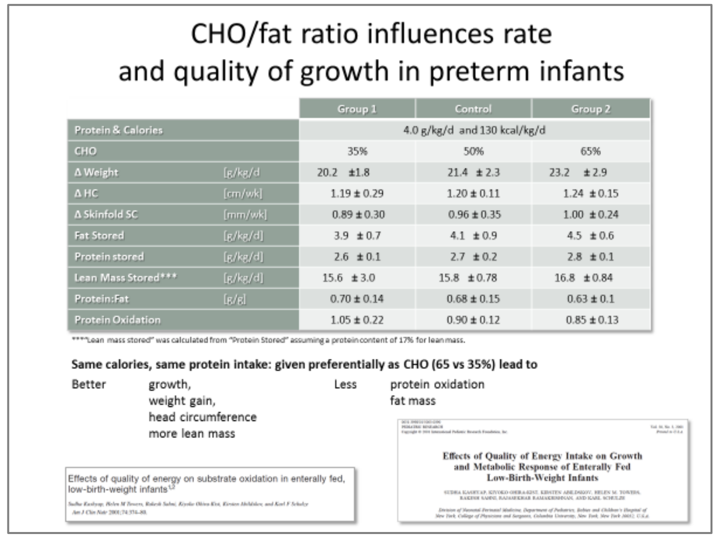
We used our data here [Slide 22] and did the calculations.[13] When we fortify milk with these 4 different fortifiers, where do we come up to with fat, with protein, and with carbohydrates? And you'll see here that for fat, the European fortifiers [are] a little bit under fortified, and the North American fortifiers, over fortified. So, North American babies get a good amount of fat when they are on breast milk. For protein, they all add the same amount, and it is not sufficient. And for carbohydrates, the European ones over fortify with carbohydrates, and the North Americans under fortify, at least this one is a little higher.



Slide 22

We said, what should the optimum composition be for a fortifier to meet the needs of most [infants]? And then we found that we should have 0.5 g/dL of fat, 1.7 g/dL of protein, and 1.0 g/dL of lactose. That would be the ideal fortifier that at least serves the purpose of most of these individuals. Interestingly, there are 2 fortifiers now in clinical studies that have a composition that is very close to what we thought should be in there. I wonder what results will come in terms of growth.

The carbohydrate-to-fat ratio influences the rate and quality of growth in preterm infants. Sudha Kashyap, MD, from New York, from [Children's Hospital of New York] did a very nice study that was published around 2000 with a few papers.[12,13] She had 3 groups of stable growing preemie babies. They all got the same amount of protein, 4.0 g/kg/d, and the same amount of calories, 130 kcal/kg/d. She provided the non-protein calories, here [Slide 23, Group 1] 35% as fat, 65% as carbohydrates, here [Group 2] it was 50%:50%, and here [Group 3] it was the other way around.



Slide 23

And what you find interestingly is that the weight gain is significantly different. Three g/kg/d is a significant difference. Head circumference was different, and babies accumulated more lean mass here, but they also accumulated a little bit more fat mass when they were receiving a little bit more carbohydrates. I think that nicely explains what you see when we add insulin, because basically you make the carbohydrates disappear, and babies don't grow so well.

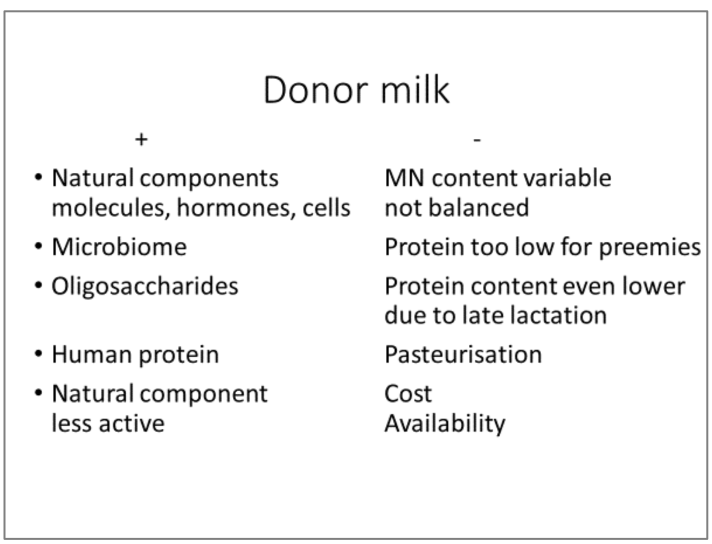
Why did that happen? She measured protein, better said, amino acid oxidation, and she found that the amino acid oxidation here [Group 1, final row] is much higher, when you give the same amount of calories as fat, compared to carbohydrates, than when you do it the other way around [Group 2, final row].

What does amino acid oxidation mean? It means that the proteins, the amino acids, are not being put together because that's an energy consuming process that might need quick energy, which means carbohydrates and not fat, but a bigger portion now goes into gluconeogenesis. To do gluconeogenesis with protein means you have to strip off the nitrogen from the skeleton, and you will have the carbon skeleton. Carbon skeleton goes into gluconeogenesis, and the nitrogen is being used to form urea. That is a very costly process. Urea is also a strong osmolyte and binds water.

My hypothesis is that some of the cosmetic edema that we see in healthy growing babies is just an overproduction of urea, and because we give imbalanced nutrition to these kids.

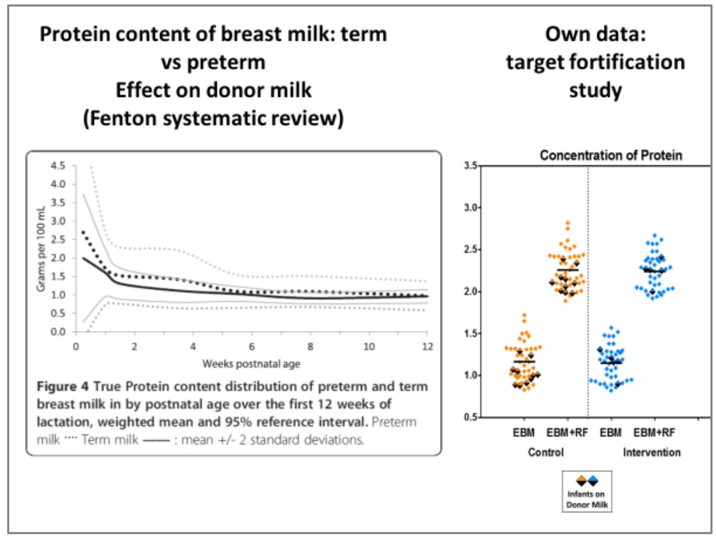
Donor Milk and Its Components

Now, let's go to donor milk. Donor milk contains natural components, molecules, hormones, and cells. It does the same as mother's milk; however the natural component is less active. Still, the macronutrient content is variable and not balanced. The protein content is even lower because donor milk is usually obtained late in lactation. We do certain procedures with donor milk: we pasteurize [it], we freeze it, which might contribute to—that the components are less active. And donor milk is relatively costly, and you don't get it everywhere.



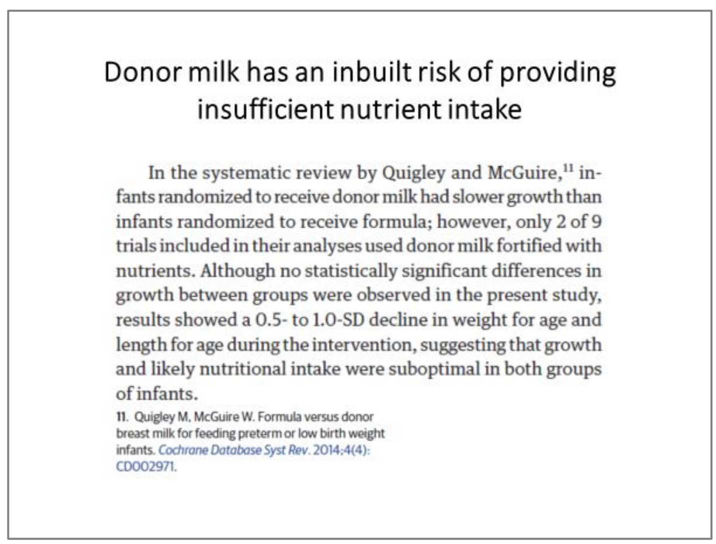
Slide 24

These are data [Slide 25, left plot] [showing] how the protein concentration goes down after birth. These are data [right plot] from our own study where you see according to protein content, these diamonds are donor milk babies, so they really receive protein at the lower end.



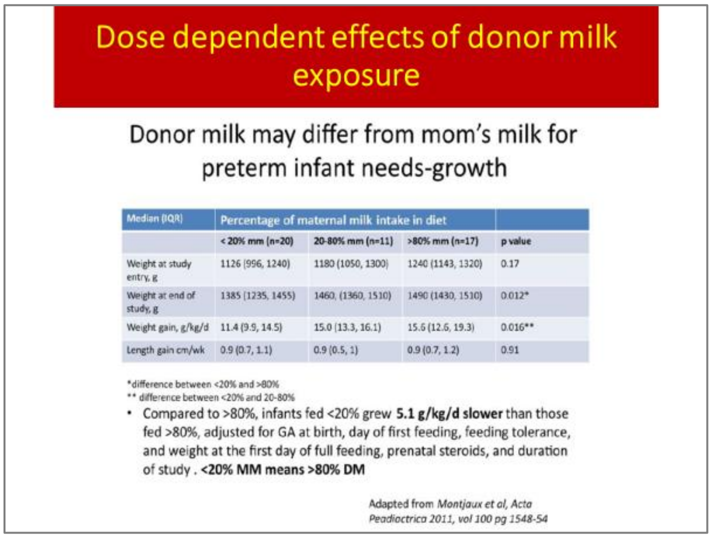
Slide 25

Donor milk has an inbuilt risk of providing insufficient nutrient intake if you don't take care.[14]



Slide 26

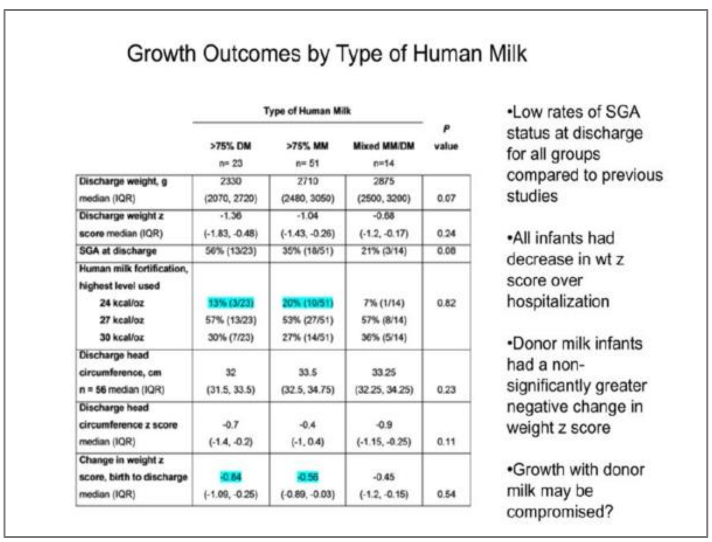
Interestingly, it has been shown in different studies here [Slide 27], that the amount of donor milk compared to mother's milk influences growth rates, as in this study.[15]



Median (IQR)	Percentage of maternal milk intake in diet			
	< 20% mm (n=20)	20-80% mm (n=11)	>80% mm (n=17)	p value
Weight at study entry, g	1126 (996, 1240)	1180 (1050, 1300)	1240 (1143, 1320)	0.17
Weight at end of study, g	1385 (1235, 1455)	1460, (1360, 1510)	1490 (1430, 1510)	0.012*
Weight gain, g/kg/d	11.4 (9.9, 14.5)	15.0 (13.3, 16.1)	15.6 (12.6, 19.3)	0.016**
Length gain cm/wk	0.9 (0.7, 1.1)	0.9 (0.5, 1)	0.9 (0.7, 1.2)	0.91

Slide 27

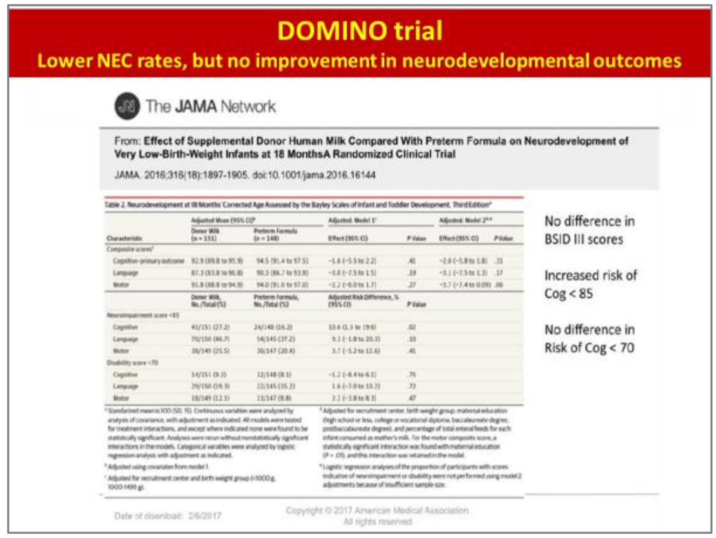
Also, in this study, the more donor milk you give, the worse the babies grew.



	Type of Human Milk			
	>75% DM n= 23	>75% MM n= 51	Mixed MM/DM n=14	P value
Discharge weight, g median (IQR)	2330 (2070, 2720)	2710 (2480, 3050)	2875 (2500, 3200)	0.07
Discharge weight z score median (IQR)	-1.36 (-1.83, -0.48)	-1.04 (-1.43, -0.26)	-0.68 (-1.2, -0.17)	0.24
SGA at discharge	56% (13/23)	35% (18/51)	21% (3/14)	0.06
Human milk fortification, highest level used				
24 kcal/oz	13% (3/23)	20% (10/51)	7% (1/14)	0.82
27 kcal/oz	57% (13/23)	53% (27/51)	57% (8/14)	
30 kcal/oz	30% (7/23)	27% (14/51)	36% (5/14)	
Discharge head circumference, cm n = 56 median (IQR)	32 (31.5, 33.5)	33.5 (32.5, 34.75)	33.25 (32.25, 34.25)	0.23
Discharge head circumference z score median (IQR)	-0.7 (-1.4, -0.2)	-0.4 (-1, 0.4)	-0.9 (-1.15, -0.25)	0.11
Change in weight z score, birth to discharge median (IQR)	-0.84 (-1.09, -0.25)	-0.56 (-0.89, -0.03)	-0.45 (-1.2, -0.15)	0.54

Slide 28

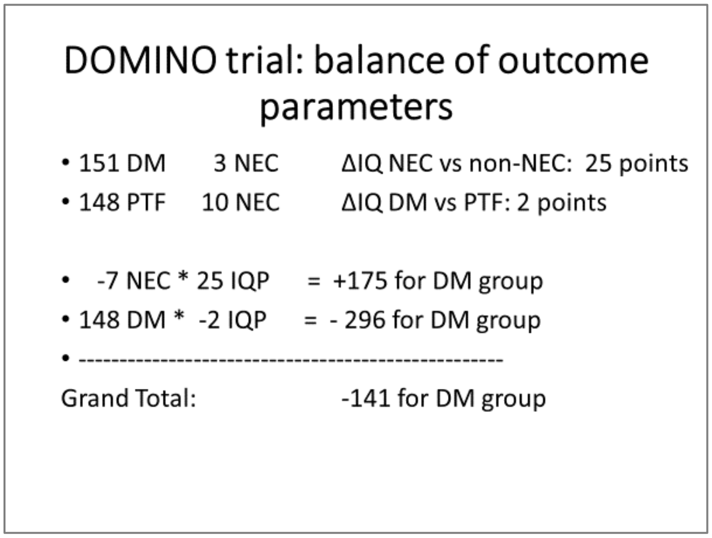
Interestingly, the DOMINO trial[16] from Deborah O'Connor, PhD, RD, which we also were part of at McMaster University, showed that babies on donor milk have a reduced NEC rate and should have a better outcome, but the neurodevelopmental outcome was not better. So, this is a little bit surprising that you have less NEC rate, but your neurodevelopmental outcome from the group is not better.



Slide 29

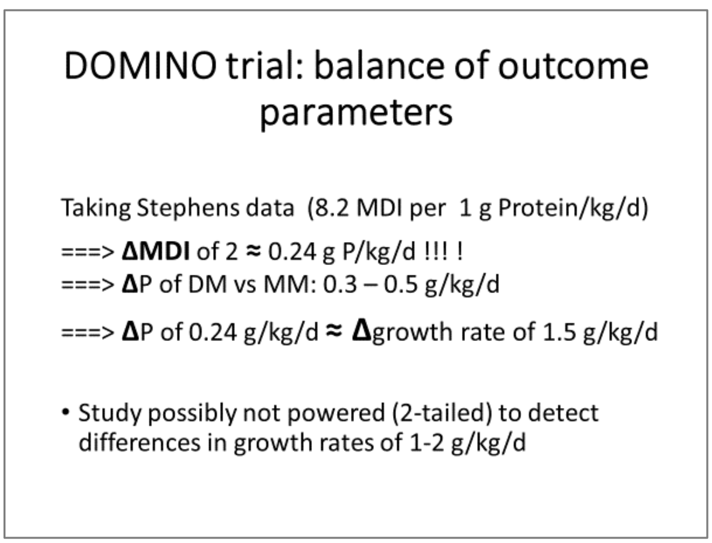
So, I thought about it and said, no, these are the number of infants on donor milk and on the standard preterm formula [n=151 DM, 148 PTF]. This was the reduction in NEC rate [-7]. And if we assume that one baby who has a NEC and a short gut, has an IQ that might be 25 points lower, then the donor milk group would gain 175 IQ points.

If the babies on donor milk grow a little less, and let's say they lose only 2 IQ points, then we are already in a negative balance, and babies on donor milk, all over, have a worse performance for neurodevelopmental outcome because each baby is affected by only a few IQ points. Whereas, here [Slide 30, row 3], few babies are affected with a big number of IQ points.



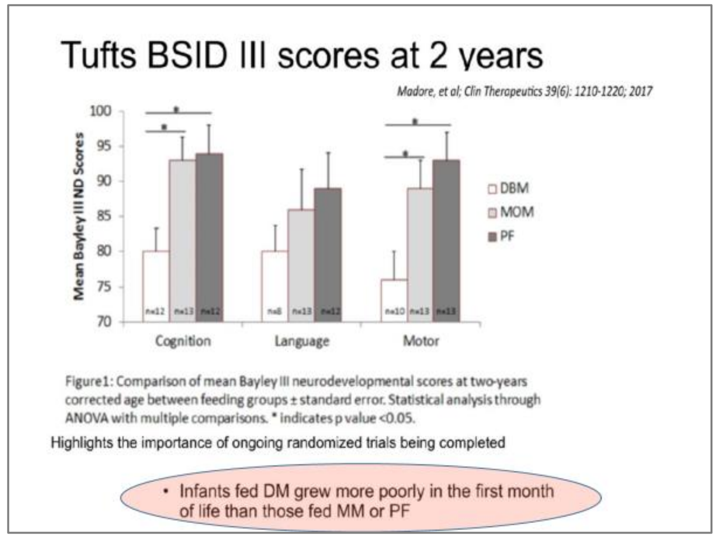
Slide 30

Now what does 2 IQ points mean? If we take Dr. Stephen's data that I've shown you in the beginning with 8.2 MDI/g protein/kg/d[17] and *delta* MDI of 2 would mean there is a difference of 0.24 g protein/kg/d. The difference between donor milk and mother's milk is between 0.3 and 0.5 g protein/kg/d. So, the *delta* of 0.24 g/kg/d here would add a growth rate difference of 1.5 g/kg/d.



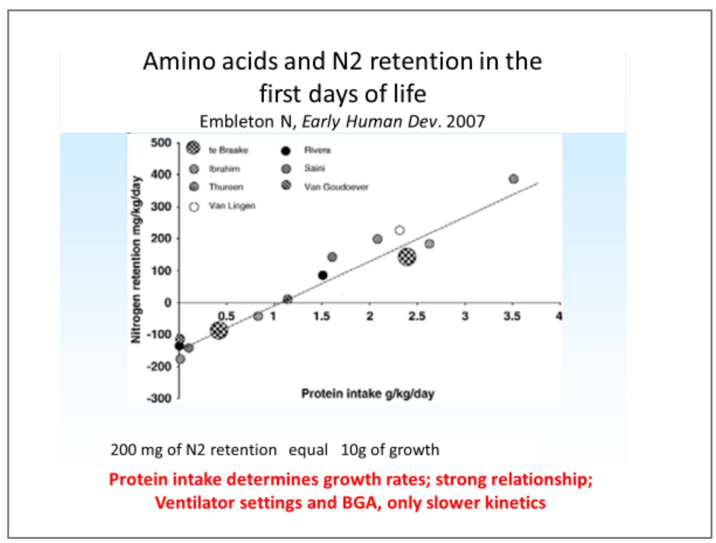
Slide 31

The study was not powered to see this difference in growth rate. So, it is still an explanation why babies on mother's milk do not do better if we don't take care of that problem. Same thing is here [Slide 32] in the data from Tufts.[18]



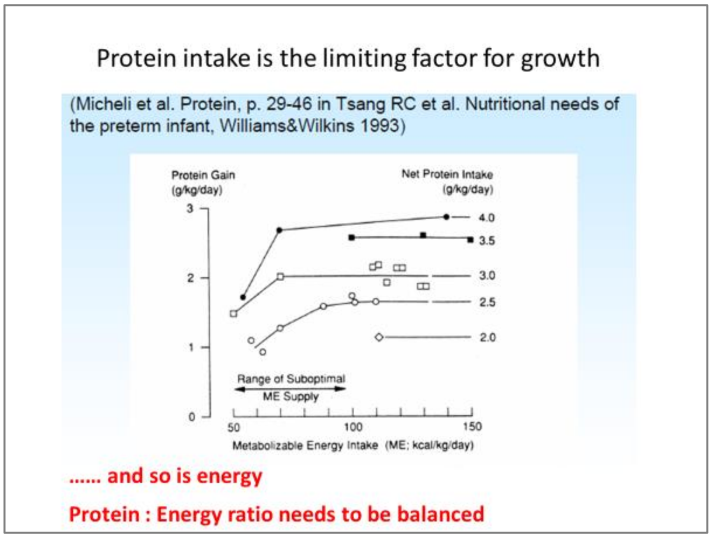
Slide 32

Okay, now how do we grow? We have protein intake [that] determines our nitrogen retention, which is the buildup of lean mass, and you need about 1g not to grow. And then, it's a very linear correlation.[19]



Slide 33

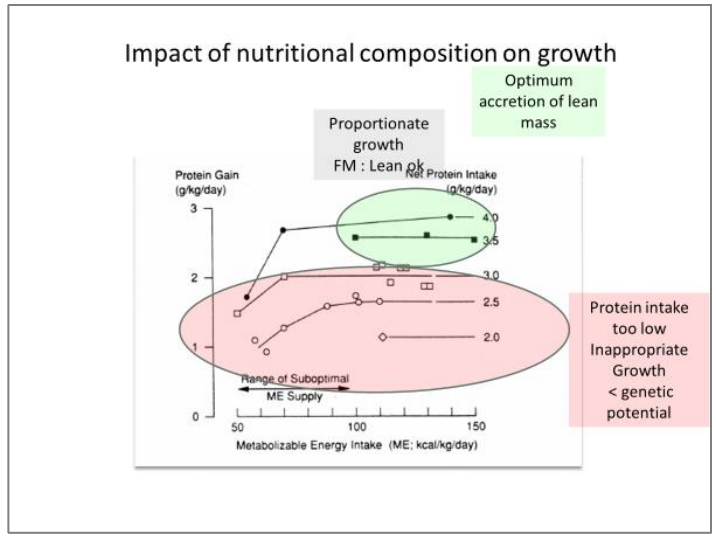
But you also need energy. So, here, basically [Slide 34], is the intake of protein.[20] This is the net gain of protein. Lean mass and here's metabolizable energy. So, if you don't give enough metabolizable energy, then you won't grow though you get an appropriate amount of protein. You need both.



Slide 34

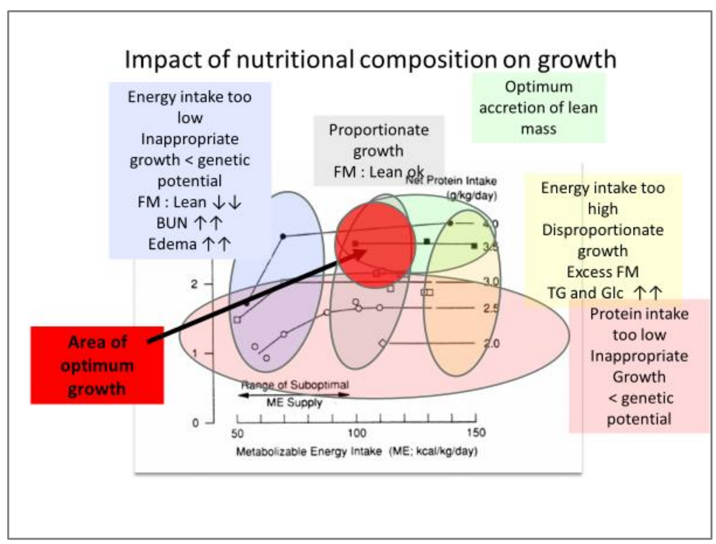
Why does it happen? To build up lean mass is an energy consuming process. To build up 1g costs about 10 kcal. If you don't provide these kcals, you won't grow. Plus, your protein will go down the urea wave because you do protein oxidation. We have seen that in Sudha Kashyap's data.[13,14]

You need both protein and energy, in the right way. If we now look into this graph and see in this area [Slide 35, shaded green], babies might grow appropriately. We get 4 g/kg/d. Three now, 4 g/kg/d. That is fine, and enough energy, so we gain a good amount of protein. Here [shaded pink], we don't grow enough because we don't get enough protein.



Slide 35

You can do it for all other circumstances. You achieve different kinds of growth. The only way where you achieve the best growth is here [Slide 36, shaded red]. This is the area of optimum growth. Where you have so much energy and so much protein. If you give more energy, you become fat [shaded yellow], you will weigh more, but that is maybe not desirable.

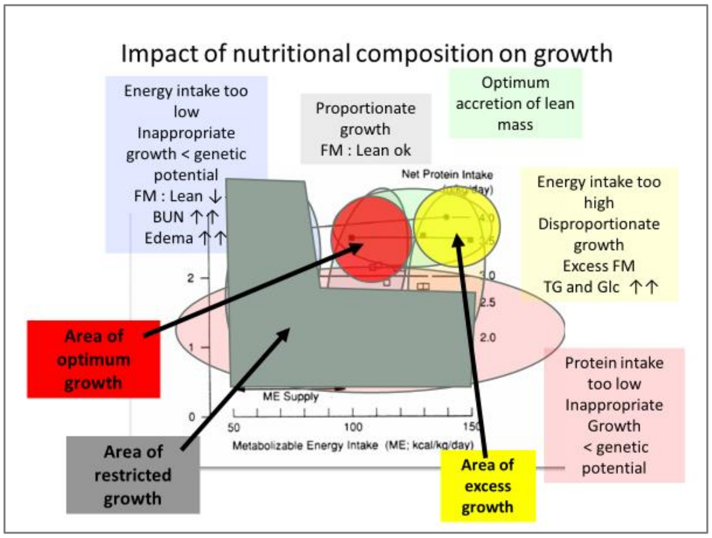


Slide 36

These are areas here of overgrowth [Slide 37, shaded yellow] and these are areas of undergrowth

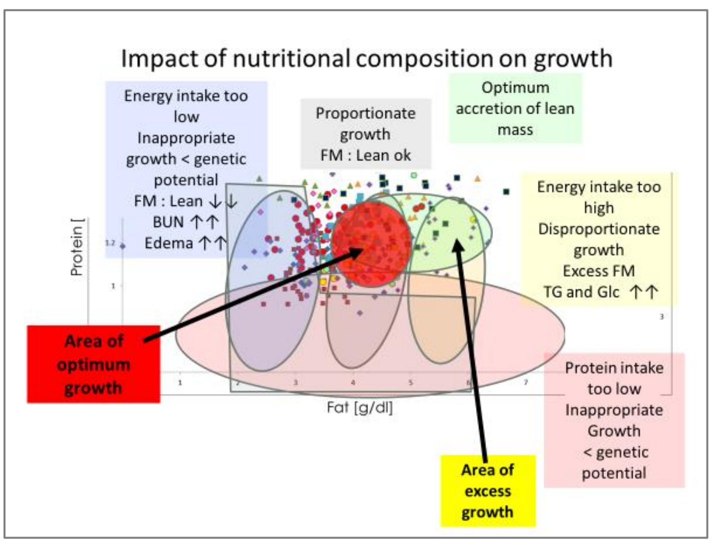
Individualized Fortification of Breast Milk for Preterm Infants

when you are here [Slide 37, shaded grey]. So, why am I showing that to you?



Slide 37

Going back to this slide here [referencing Slide 19], where we have energy and protein, and I do the overlay of what I've shown to you [Slide 38], then you see that there are, indeed, combinations of breast milk where babies cannot grow. And unfortunately, there are more combinations where babies cannot grow than that they grow too much.



Slide 38

To overcome this, what I would like to say is that breast milk is a highly unstandardized diet that sometimes can make babies fail to grow. Even if we give more of everything, we cannot fix certain deficits. So, in this study here [Slide 39],[21] adjustable fortification was done. It was a stepwise procedure having 6 strengths of protein, and that was added according to the BUN [blood urea nitrogen levels]. As long as BUN was low, [HMF and protein] was increased. As long as the BUN was high, [HMF and protein] was decreased.

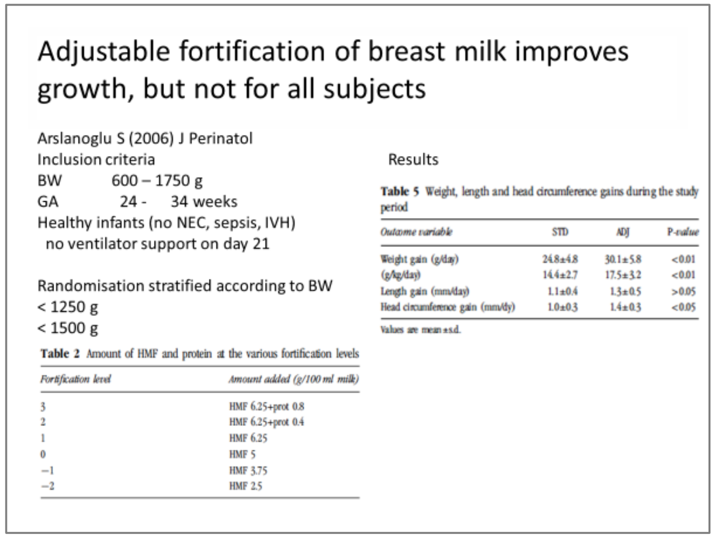


Table 2 Amount of HMF and protein at the various fortification levels

Fortification level	*Amount added (g/100 ml milk)*
3	HMF 6.25+prot 0.8
2	HMF 6.25+prot 0.4
1	HMF 6.25
0	HMF 5
−1	HMF 3.75
−2	HMF 2.5

Table 5 Weight, length and head circumference gains during the study period

Outcome variable	STD	ADJ	*P-value*
Weight gain (g/day)	24.8±4.8	30.1±5.8	<0.01
(g/kg/day)	14.4±2.7	17.5±3.2	<0.01
Length gain (mm/day)	1.1±0.4	1.3±0.5	>0.05
Head circumference gain (mm/dy)	1.0±0.3	1.4±0.3	<0.05

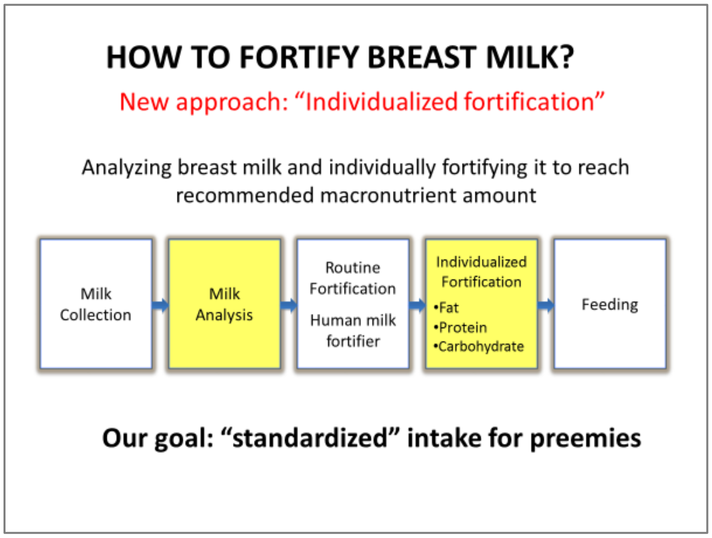
Values are mean ±s.d.

Slide 39

Here's the difference in weight gain [right table]. So, the adjusted fortification gets a better weight gain. You'll see here, 14.4 g/kg/d vs 17.5 g/kg/d. Still, with this variation, you see that there are babies that will grow with 14 g/kg/d and also with 11 g/kg/d, which is not appropriate.

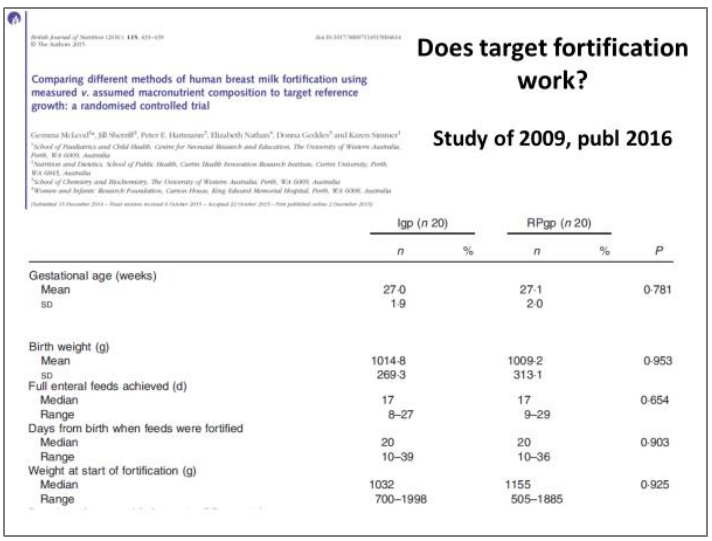
That's why we thought we need to do individualized fortification, which basically means we do milk analysis here, then add, additionally, what is missing.

Individualized Fortification of Breast Milk for Preterm Infants



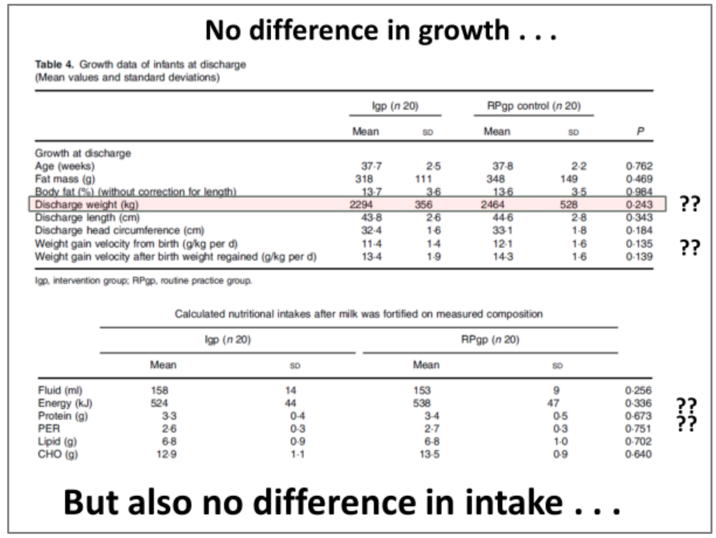
Slide 40

There's one study from Karen Simmer, MD, PhD, from [University of Western] Australia that did it already in 2009,[22] and didn't find differences doing it. Why?



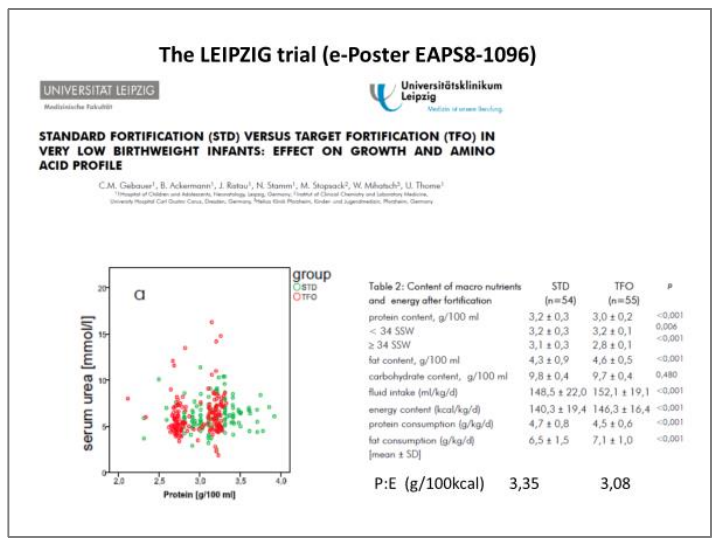
Slide 41

If you look into the data, then you'll see here [Slide 42] protein-to-energy ratio, which she achieved in both groups, the same intake, so there was no difference, and 2.6 g is too low for proper growth. So, that's why she didn't find an intake, a difference in growth.



Slide 42

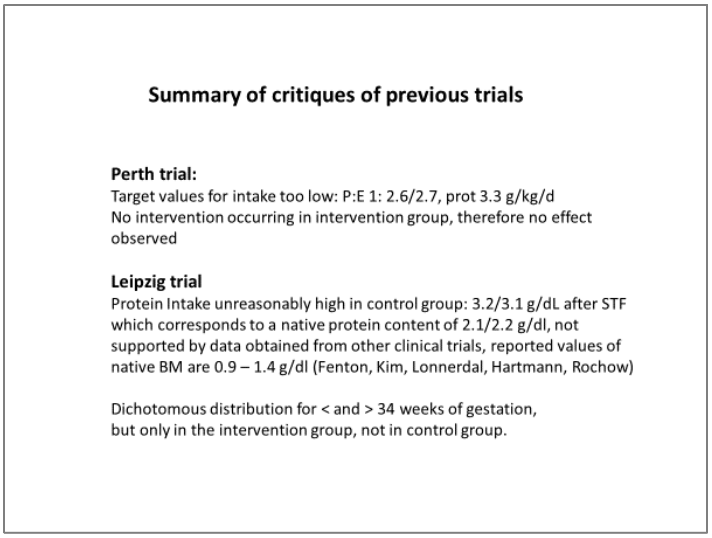
There is a trial from Leipzig, where they did a similar thing, but there must be some methodological issues because you cannot, in a blinded trial, get a dichotomous distribution in the same group. That would not work. They didn't find a difference either.



Slide 43

There are some critiques on these previous trials. I don't want to go into too much detail. They are methodological and also the way how they measure.

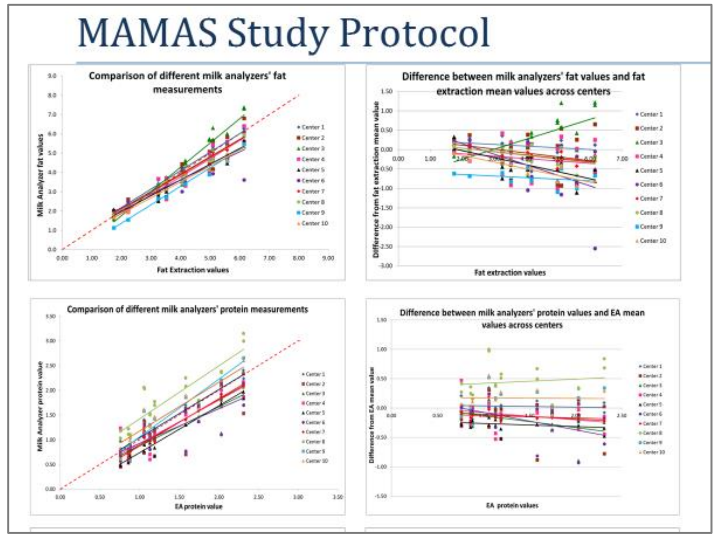
Individualized Fortification of Breast Milk for Preterm Infants



Slide 44

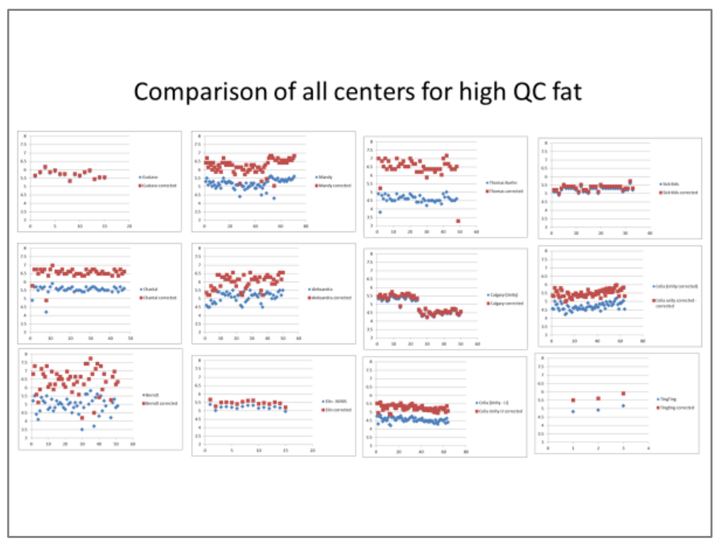
We spend a good amount of time in finding out how can we measure macronutrient content at the bedside? We use these dairy industry milk analyzers, which is a big business, and use them to measure human milk. Unfortunately, you need to do a lot of work to basically tame the shrew. Because human milk and cow's milk are different.

We did that, I can't show you all the data because it would take too much time, but these are recent data from a multicenter study where we sent out the same samples to 13 labs in North America and in Europe, and you see that the data here [Slide 45, lower left plot] were all over the place. So, you can't do target fortification if you have this bad precision.



Slide 45

So, we were training and modifying these devices, and here you see how the different units measured differently at quality controls. Obviously, this unit [Slide 46, 1st column, last plot] will get different results from target fortification compared to this one [3rd column, last plot].

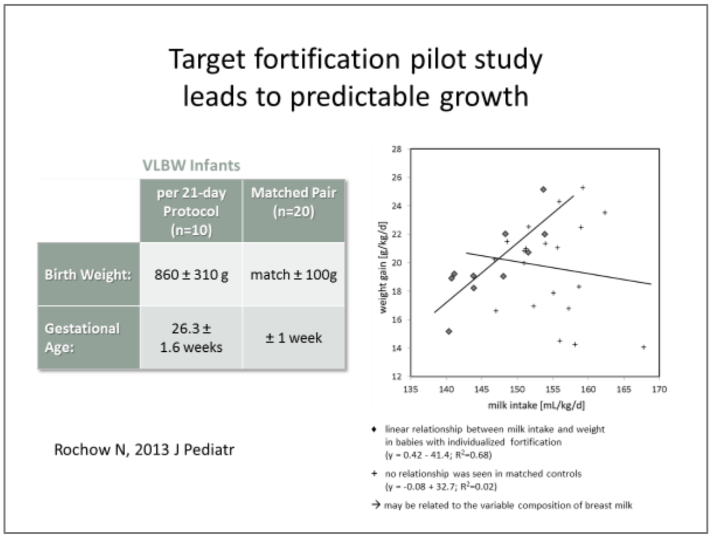


Slide 46

The good thing is we could train them with corrected values. We measure the right thing with a lot of training. That can be overcome, but it's not just buying a milk analyzer and then you have the right values.

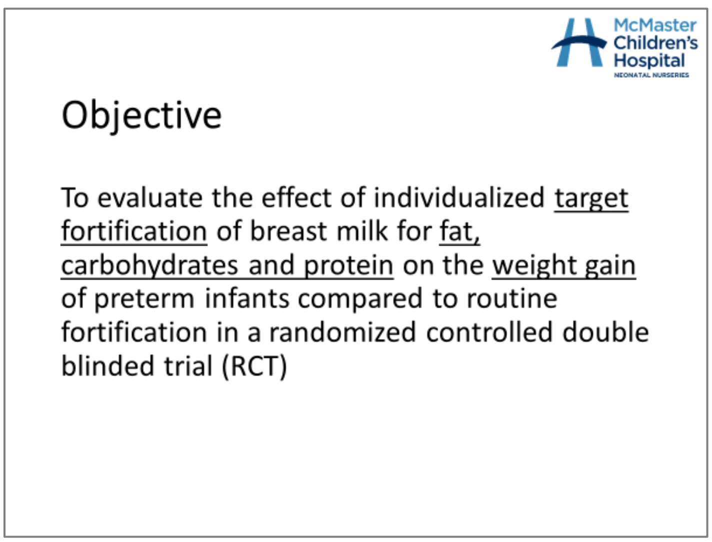
Individualized Fortification of Breast Milk for Preterm Infants

This is a study [Slide 47] that we did as a pilot study about target fortification.[23] And we saw a predictable weight here [plot] in babies that got target fortification and was dependent upon milk intake, which was not the case for babies that were on standard fortification. These babies did not grow, though they got a huge amount of fluid here.



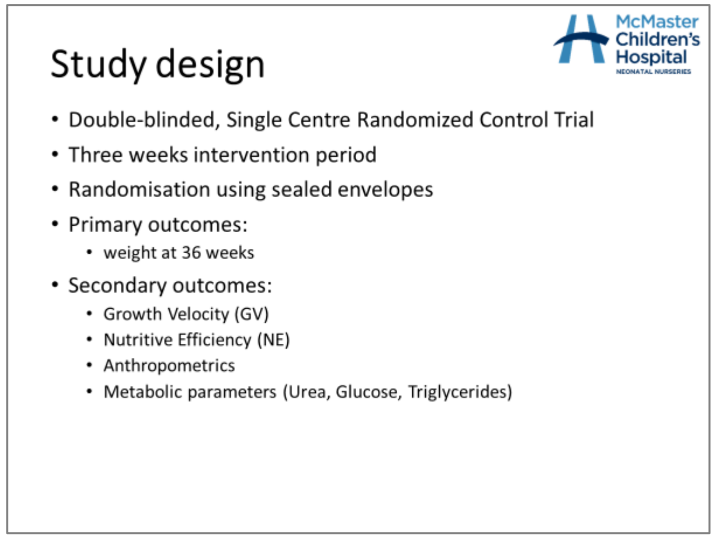
Slide 47

Then we did a randomized controlled trial, and I will show you the data in this last part of my talk.



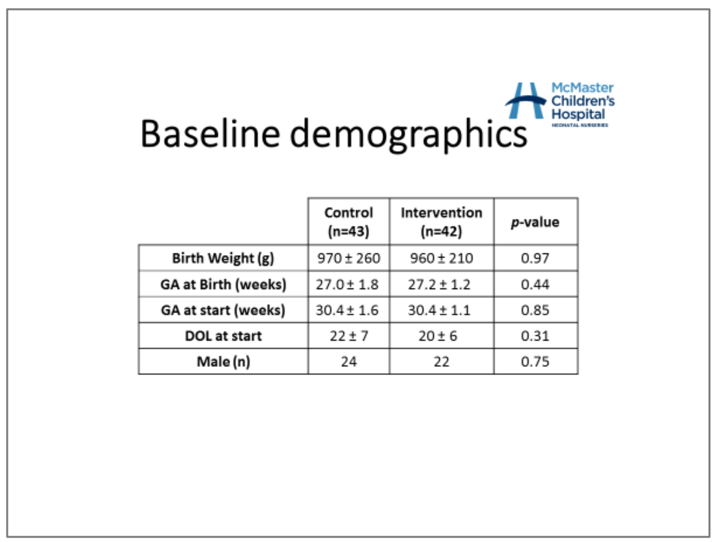
Slide 48

The randomized trial in 100 babies. It was double-blinded single-center randomized control, but with 3 weeks of intervention. Randomization was done with sealed envelopes, and the primary outcome was weight at 36 weeks. And we had lots of secondary outcomes.



Slide 49

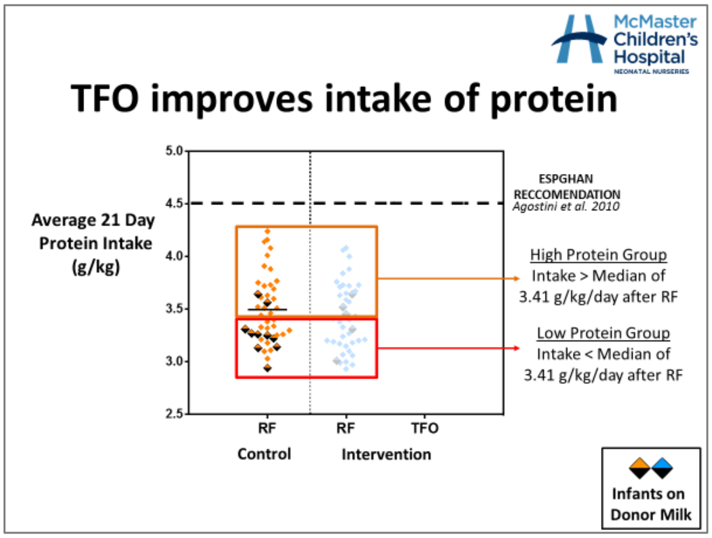
These are the baseline demographics [Slide 50]: no difference, 27 weeks, 970 g. We started with staggered fortification around 3 weeks of life.



	Control (n=43)	Intervention (n=42)	*p*-value
Birth Weight (g)	970 ± 260	960 ± 210	0.97
GA at Birth (weeks)	27.0 ± 1.8	27.2 ± 1.2	0.44
GA at start (weeks)	30.4 ± 1.6	30.4 ± 1.1	0.85
DOL at start	22 ± 7	20 ± 6	0.31
Male (n)	24	22	0.75

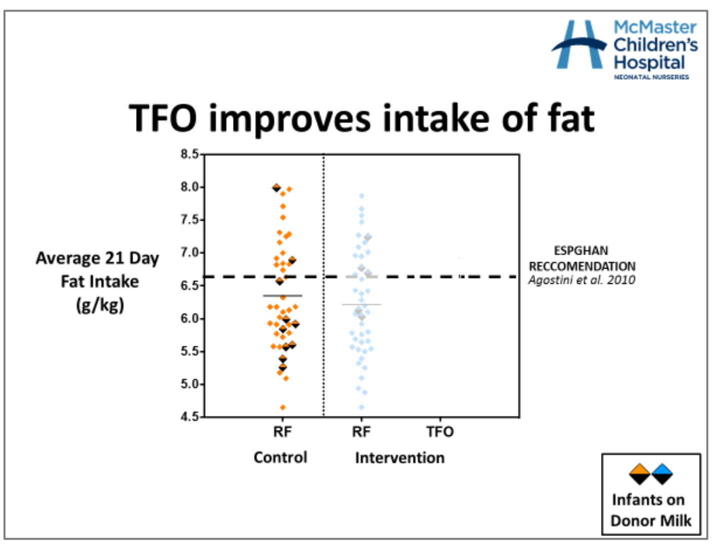
Slide 50

You see that the TFO (target fortification) improves intake of protein. So, this is the protein intake after standard fortification in the control group. This would have been in the TFO group, but with the extra fortification, we indeed achieved 4.5 g/kg/d.



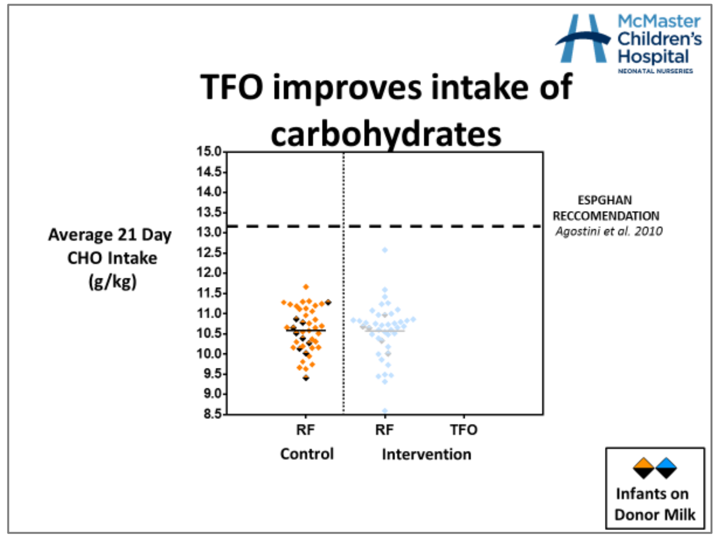
Slide 51

The same happened with fat, but there was not a big effect, because I told you that North American fortifiers already contain so much fat that we came close to the recommendations (was a little bit more).



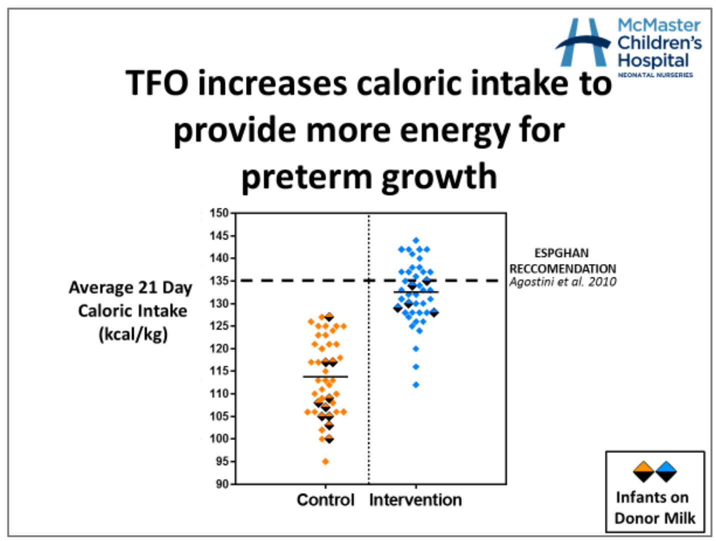
Slide 52

For carbohydrates, it was a big difference.



Slide 53

Overall, the caloric intake also improved.



Slide 54

These are the data: control = 2280 g, intervention = 2510 g, a difference of 230 g. Growth velocity was also different.

Individualized Fortification of Breast Milk for Preterm Infants

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NEONATAL NURSERIES

TFO improves growth outcomes

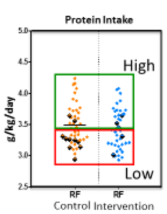
	Control (n=43)	Intervention (n=42)	*p*-value
Weight (g)	2280 ± 340	2510 ± 290	0.001
Growth Velocity (g/kg/d)	19.4 ± 2.3	21.2 ± 2.3	<0.001
Nutritive Efficiency (g/dL)	12.6 ± 1.6	13.9 ± 1.7	<0.001
TFI (mL/kg/d)	155 ± 4	153 ± 4	0.008

$$\text{Nutritive Efficiency} = \frac{\textit{Growth Velocity}}{\textit{TFI}}$$

Slide 55

Then we did a subgroup analysis in the high and low protein group. High and low protein group means the protein content in native, unfortified breast milk. You'll see that in those individuals that receive breast milk from mothers with high-protein content, there's no effect of the intervention. They grow equally well. Why? Because mother has already enough protein in there. In the low-protein group, there is a huge difference, of about 370 g. Also, the growth rates are significantly different.

McMaster Children's Hospital
NEONATAL NURSERIES



Improved growth outcomes in Low Protein group

	High Protein			Low Protein		
	Control (n=22)	Intervention (n=21)	p-value	Control (n=21)	Intervention (n=21)	p-value
Weight (g)	2400 ± 331	2480 ± 265	0.35	2170 ± 316	2540 ± 312	<0.001
Growth Velocity (g/kg/d)	19.7 ± 2.0	21.3 ± 2.0	0.011	19.2 ± 2.7	21.0 ± 2.6	0.030
Nutritive Efficiency (g/dL)	12.7 ± 1.4	14.0 ± 1.6	0.009	12.4 ± 1.9	13.8 ± 1.8	0.019
TFI (mL/kg/d)	155 ± 4	153 ± 3	0.087	155 ± 3	153 ± 4	0.044
	Intake after RF > Median of 3.41 g/kg/day			Intake after RF < Median of 3.41 g/kg/day		

Slide 56

Babies who received breast milk with low-protein content will definitely benefit from this approach. It is relatively logical, because it is physiology, but we could show it in this trial.

Interestingly, there were also a trend to better outcomes for all NICU outcomes in the TFO group. We had less NEC, less death, less PDA [patent ductus arteriosus]. Feeding intolerance was also lower (maybe because we had a more constant intake for the babies, no variation).

Perinatal characteristics and NICU Outcomes

	Control Group		TFO Group	
	Randomized	Completed	Randomized	Completed
N -	89	51	90	52
Maternal Diabetes	6 (7)	3 (6)	7 (8)	6 (12)
Hypertension/Preeclampsia	16 (18)	12 (24)	25 (28)	17 (33)
Suspected Chorioamnionitis	32 (36)	15 (29)	25 (28)	13 (25)
Antenatal Corticosteroids	84 (94)	49 (96)	78 (87)	47 (90)
Died	5 (6)	2 (4)	0 (0)	0 (0)
NEC all cases	5 (6)	2 (4)	2 (2)	0 (0)
NEC Bell stage 3	3 (3)	1 (2)	0 (0)	0 (0)
Sepsis clinical	41 (46)	19 (37)	30 (33)	14 (27)
Sepsis culture positve	17 (19)	7 (14)	22 (24)	10 (19)
PDA	57 (64)	30 (59)	51 (57)	29 (56)
PDA treated	35 (39)	21 (41)	31 (34)	14 (27)
BPD mild	22 (25)	17 (33)	20 (22)	15 (29)
BPD moderate/severe	32 (36)	18 (35)	29 (32)	16 (31)
feeding intolerance		14 (27)		8 (15)**

No difference in perinatal characteristics and demographics

Trend towards better NICU outcomes

Better tolerance of enteral nutrition (more balanced intake??)

Slide 57

The clinical chemistry is here [Slide 58], 2.5 BUN vs 4.2 in both groups, a slight increase but not of clinical significance. Interestingly, the triglycerides went down, maybe because we gave more carbohydrates, so we could burn the fat a little better.

Individualized Fortification of Breast Milk for Preterm Infants

Clinical chemistry

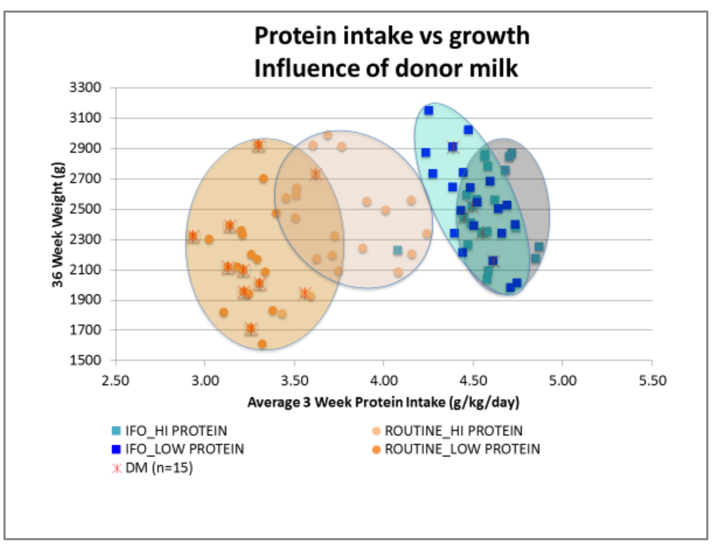
Outcome	All		High protein group		Low protein group	
	Control Group	TFO Group	Control Group	TFO Group	Control Group	TFO Group
Glucose Day 14	4.1±0.8	4.6±0.9*	4.3±0.9	4.6±1.1	4.0±0.8	4.5±0.7*
Glucose Day 21	4.6±2.3	4.3±1.1	4.8±3	4.3±1.2	4.3±0.9	4.2±1
BUN Day 14	2.5±1.1	4.2±1.5***	2.8±0.9	4.1±1.6**	2.3±1.2	4.2±1.4***
BUN Day 21	2.5±1.1	4.7±1.5***	2.8±1.2	4.8±1.1***	2.3±0.8	4.6±1.8***
TG Day 14	0.9±0.4	0.7±0.2	0.8±0.3	0.7±0.2	0.9±0.6	0.7±0.2
TG Day 21	0.8±0.5	0.7±0.3	0.8±0.3	0.7±0.3	0.9±0.7	0.7±0.2

Mild increase in glucose and BUN levels without clinical significance
Drop in triglyceride levels

Slide 58

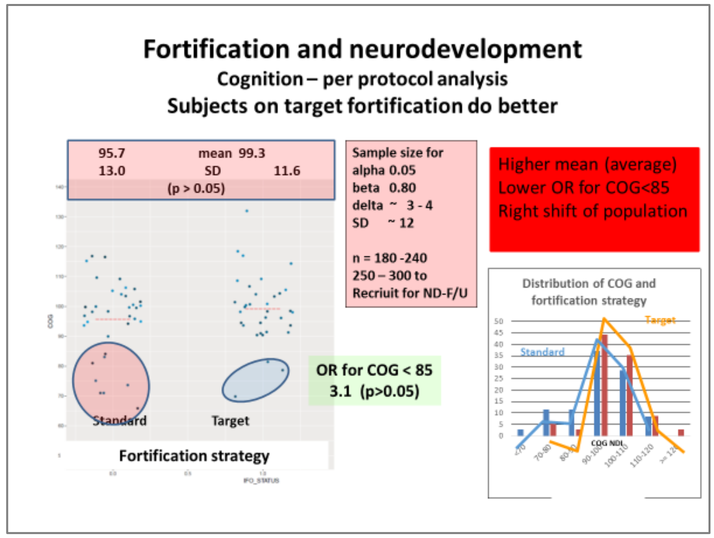
Here [Slide 59] you see all protein intakes vs weight; and these are the 2 groups with low-protein intake. This is without fortification. This is with fortification. You see that these children who do not grow well without target fortification, basically, are moved up here [higher weight].

You see here these babies also on donor milk intake, and they are now also up here. This again visualizes that the effect is mostly pronounced in this group of babies with low-protein content in native breast milk.



Slide 59

These are very recent data [Slide 60]. We got the neurodevelopmental follow-up. It's not shown, yet, at a conference like PAS [Pediatric Academic Societies]. And you'll see here that the intellectual outcome, the difference is about 4 points. The odds ratio for cognitive below 85 is 3.1.



Slide 60

Also, if you look here at the distribution of the IQs, you see a shift to higher IQs, and the effect size again is around 4. Unfortunately, we are not powered to detect this difference, so we didn't find statistical difference. If we wanted to have been powered on these levels, we would have needed to include 250 to 300 babies. So, that's something for the next step for a multicenter randomized control trial.

In summary, preterm formula makes babies grow with predictable and adequate growth rates, including neurodevelopment. Trade-off is microbiome and NEC.

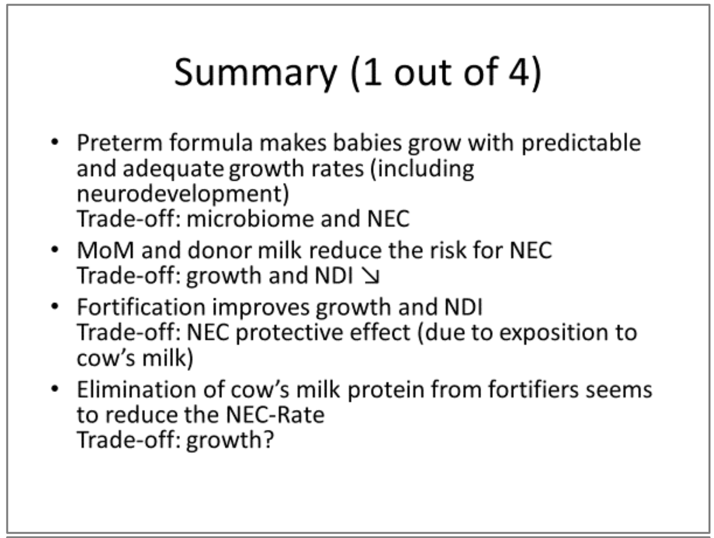
Mother's own milk and donor milk reduce risk for NEC. The trade-off is growth, and neurodevelopmental outcome might not be as good as it could be. Fortification improves growth and neurodevelopmental outcome. Trade-off is NEC protective effects due to the exposition of cow's milk decreasing. The elimination of cow's milk protein from fortifiers seems to reduce the NEC rate. Trade-off is growth. (I didn't show you the data on that.) Modern fortifiers should contain more protein, about 0.5–0.7 g/kg/d, and a more balanced mixture of fat and carbohydrates.

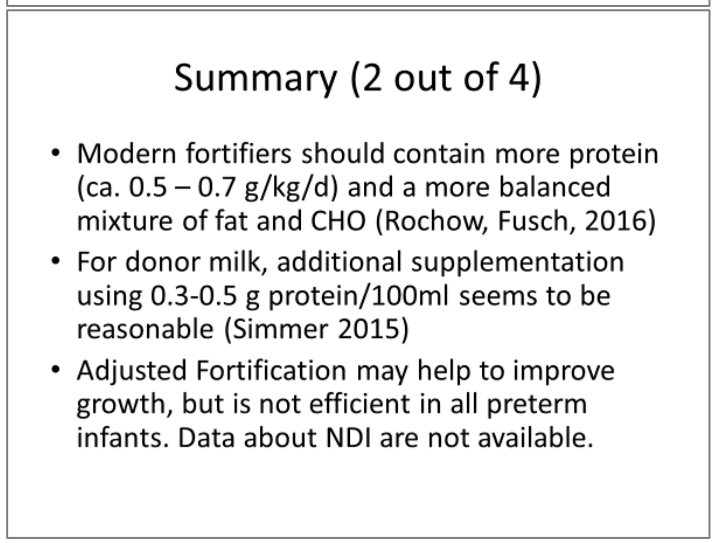
For donor milk, additional supplementation using 0.3–0.5 g protein/100 ml seems to be reasonable. I think that may be the most important message.

Adjusted fortification may help to improve growth but is not efficient in all preterm infants. Data about NDI are not available.

The 2 randomized controlled trials on TFO are charged with significant methodological limitations and cannot be generalized to a standard setting. Data from our double-blind randomized controlled trial showed that target fortification improves growth compared to standard fortification,[24] most likely including neurodevelopmental outcome. It's a kind of precision medicine.

For both high-end fortification strategies, modern modular components need to be developed to conserve the NEC protective effect of breast milk, ideally to make it cow's milk protein free and minimize the pro-inflammatory potential by using better lipids. More research and clinical studies are needed, and they need to apply rules of good laboratory practice.





Individualized Fortification of Breast Milk for Preterm Infants

Summary (3 out of 4)

- The 2 RCTs on TFO are charged with significant methodological limitations and cannot be generalized to a standard setting
- Data from our double-blind RCT show that target fortification improves growth compared to standard fortification (most likely including NDI).
  Precision medicine....

Summary (4 out of 4)

- For both "high-end" fortification strategies modern modular components need to be developed to conserve the NEC protective effect of breast milk (cow's milk protein free) and minimize the pro-inflammatory potential (omega 3 : 6, limited MCT).
- More research and clinical studies are needed and they need to apply rules of GCLP.

Abbreviations

BGA	blood gas analysis	**LISA**	less invasive surfactant administration
BM	breast milk	**MAMAS**	maternal adiposity, metabolism, and stress study
BSID	Bayley Scale of Infant Development	**MDI**	mental development index
BUN	blood urea nitrogen	**ME**	metabolize energy
BW	birth weight	**MM**	mother's milk
CHO	carbohydrate	**MN**	macronutrients
Cog	cognition	**NDI**	national death index
CPAP	continuous positive airway pressure	**NE**	nutritive efficiency
CV	conventional ventilation	**NEC**	necrotizing enterocolitis
DM	donor milk	**NIV**	non-invasive ventilation
DOHaD	developmental origins of health and disease	**NHFOV**	non-invasive high-frequency ventilation

Individualized Fortification of Breast Milk for Preterm Infants

ELBW	extremely low birth weight	**PDA**	patent ductus arteriosus
ESPGHAN	European Society for Pediatric Gastroenterology Hepatology and Nutrition	**PDI**	physical development index
F	fat	**PNGR**	postnatal growth restriction
FM	fat mass	**PTF (or PF)**	preterm formula
GCLP	Guidelines for Good Clinical Laboratory Practices	**SGA**	small for gestational age
GV	growth velocity	**TG**	triglyceride
HFOV	high-frequency oscillation ventilation	**TFI**	total fluid intake
HM	human milk	**TFO**	target fortification
HMF	human milk fortifier	**VLBW**	very low birth weight
HMO	human milk oligosaccharide		

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