



<b>Algemene gegevens</b>	
PPS-nummer	TKI-AF-12190
Titel	PPS-Food4LiveSolutions
Thema	TKI Agrifood, Theme 6 (Health) and Theme 4 (sustainability of livestock breeding).
Uitvoerende kennisinstelling(en)	TNO
Projectleider onderzoek (naam en emailadres)	Jasper Kieboom (TNO)
Penvoerder PPS (namens private partij)	Harm Dijkstra & Laurien Ulfman (FrieslandCampina)
Contactpersoon overheid	
Werkelijke startdatum	08-07-2013
Werkelijke einddatum	31-06-2018

<b>Goedkeuring penvoerder / consortium</b>	
De eindrapportage dient te worden besproken met de penvoerder/het consortium. De TKI's nemen graag kennis van evt. opmerkingen over de rapportage.	
De penvoerder heeft namens het consortium de eindrapportage	<input checked="" type="checkbox"/> goedgekeurd <input type="checkbox"/> niet goedgekeurd
Evt. opmerkingen over de eindrapportage:	N.v.t.

<b>Mutaties ten opzicht van het oorspronkelijke projectplan en follow-up</b>	
Zijn er wijzigingen geweest in het consortium / de projectpartners? Zo ja, benoem deze.	Nee
Zijn er inhoudelijke wijzigingen geweest in het project?	Vooral voor het humane gedeelte is een alternatieve route ingezet om toch antwoorden te krijgen op de oorspronkelijke vragen door naast de analyse voor het oorspronkelijke Indonesisch cohort een Nederlands cohort te analyseren. Tevens is er besloten geen virus qPCRs uit te voeren.
Is er sprake van een octrooi-aanvraag (evt. first filing) vanuit deze PPS?	Nee
Is er sprake van spin-offs (contract-onderzoek dat voortkomt uit dit project, aanvullende subsidies die zijn verkregen of spin-off bedrijvigheid)	Ja
Binnen hoeveel jaar zullen de private partijen resultaten uit dit project gaan gebruiken in de praktijk?	Onbekend
In hoeverre heeft het project bijgedragen aan de ontwikkeling van de betrokken kennisinstelling(en)? (bijv. wetenschappelijk track record, nieuwe	In grote mate heeft dit project bijgedragen aan de kennispositie. Zowel op technologie gebied (sequencing, bioinformatica), publicaties en samenwerking met UMCU en nieuwe partijen in Indonesië.

technologie, nieuwe samenwerkingen)	
Krijgt het project een vervolg in de vorm van een nieuw project of een nieuwe samenwerking? Zo ja, geef een toelichting.	Nog geen concrete plannen

<p><b>Resultaten</b></p> <p>Wat is er aan de hand?  FrieslandCampina and VanDrie Group aim for a common goal; making the optimal food for the first phase in life to ensure optimal growth and development of a healthy newborn. Two interesting observations triggered this research initiative: 1) breastfeeding and suckling protects against the development of infections in both human and bovine newborns, respectively, and 2) the microbiota of breast and formula fed newborns has a different composition at various sites along the respiratory and gastrointestinal tract.</p> <p>Wat doet het project daaraan?  Since the host's microbiome is assumed important for pathogen-resistance and immune-modulation, and thus for a healthy development of newborns, we hypothesize that the composition of the microbiota of both human and bovine newborns plays an important role in resistance against infections and is steerable through diet. By comparing the microbiota of breastfed infants or traditionally fed calves with formula-fed infants or novel food intervention strategies in calves f insights were obtained on the impact of formula feeding on the microbiota composition of infants and calves. Data regarding health status of infants and calves were linked with the microbiota profiles as determined by advanced molecular technologies to understand how to steer to optimal microbiota profiles via food.</p> <p>Wat levert het project op?  Extensive microbiota profiles of calves and infants were generated and analyzed using an Illumina platform. Integrated models, that link microbiota composition with health outcomes, have provided us with the characterization and understanding of microbiota profiles that are associated with health parameters in infants. By combining both microbial ecology and newborn physiology in relation to health and disease, new leads will be obtained for the development of functional foods and/or ingredients for the infant formula market and the calf husbandry. The resulting reduction in infections will decrease antibiotic usage in calves due to a higher resistance. Optimal food in the first phase of life will set a fundament for health later in life.</p> <p>Wat is het effect hiervan?  Early-life environmental exposures, including infant feeding, have strong effects on respiratory microbiota development (Bosch et al., 2016 &amp; 2017). Moreover, our data show a strong resemblance between nasopharynx microbiota of Asian and European infants suggesting that the biological niche more than geographical location is determines the composition of the respiratory microbiota.</p>
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<p><b>Deliverables (geef een korte beschrijving per deliverable uit het projectplan)</b></p> <p><b>WP1 Evaluation and preparation</b></p> <ul style="list-style-type: none"> <li>• <i>Report and presentation on literature evaluation of existing data</i></li> <li>• <i>Report and presentation consisting of an overview of available data retrieved through bioinformatics</i></li> <li>• <i>In silico selection of molecular probes for relevant (micro)biological targets and aims human infant and calf NPX from literature and bioinformatics tools</i></li> <li>• <i>Report on technical outcome of feasibility of study with recommendations proposal for workplan</i></li> </ul> <p>A literature search was performed and presented within the consortium. While partner UMCU presented human aspects of respiratory microbiota – disease interactions, TNO presented this for the calves. State of art data was presented for both calf as well as human respiratory microbiota. Based on availability of data in the public domain and</p>
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sample collections available a strategy was proposed for further analytical steps.

Human part: From literature, few publications/data were identified on relations between infant nutrition, respiratory health and the respiratory microbiome. However, all available data sets were from Western infants and may not be relevant for an Asian population. Plans were put forward to establish better respiratory microbiota models and to explore acquiring data or samples from Asian infant populations.

Calf part: From literature it became clear that not sufficient data was available on the respiratory microbiome. In addition, sample techniques were not well established. We therefore planned to establish sampling techniques, establish a reference data base for the healthy and unhealthy respiratory microbiota. In a final study, effects were examined of a food intervention.

### **WP2: Proof of principle (infants) and lead identification (calves)**

- *Report and presentation on microbiota profile of breastfed vs formula fed infants of  $t=0$  and  $t=4$ .*
- *Identification of main differences in microbiota profile between groups, illustration of differences through pie charts, and to this purpose adapted cluster analysis as well as (multivariate) statistical analysis. Recommendation for specific microbial targets to be analyzed by qPCR.*
- *Report containing description of temporal dynamics of calf nasopharynx including analysis of associations between symptoms and antibiotic usage.*
- *MTA documentation approved by local authorities.*

Human part:

As the planned Indonesian intervention study was stopped, alternative plans had to be made. For this, we made use of a Dutch infant cohort set up by partner UMCU, for which samples were available for analysis. Analysis was presented and discussed within the consortium and published in scientific journals. In addition, samples could be retrieved from an Indonesian intervention study by extension of the Indonesian project. Difficulties in shipping Indonesian samples for analysis in the Netherlands were avoided by establishing protocols at the Indonesian study partner. Findings on differences between breast fed and formula fed infants and the dynamic development of the Indonesian and Dutch infant respiratory microbiota were presented within the consortium. A scientific publication is in preparation.

Calf part:

WP1 showed that research was required to establish methods for sampling and references for a healthy and unhealthy respiratory microbiota in calves. In collaboration with the Gezondheidsdienst voor Dieren and de vanDrie groep, we established a method for sampling a respiratory microbiota analysis in calves of different ages. Next, analysis was performed in veal and rearing calves in order to acquire a reference database for the dynamic development of the respiratory microbiota in veal and rearing calves, in association with antibiotics use and the incidence of infections. Results were presented and discussed within the consortium.

### **WP3: food/feed intervention trials**

- *Presentation of the effects of feed intervention study in calves*
- *Presentation with description of profile of nasopharyngeal microbiota of infants in Indonesia.*

A feed intervention study was performed by partner vanDrie. Samples were analyzed by TNO. Data was analyzed and presented to the consortium, description of dynamic profile of nasopharyngeal microbiota of calves of the feed intervention groups in relation to symptoms of upper respiratory tract infections.

Data acquired on the Indonesian study allowed studies on the correlations of infant feeding with incidence and/or duration of upper respiratory tract infections and re dynamic development of the upper respiratory microbiota of Indonesian infants. Comparison was

made with the respiratory microbiota of Dutch infants. The findings were presented and discussed with F4L consortium members.

**WP4: Development of predictive model for beneficial communities and design of food and feed solutions**

- *Report on the microbial network interactions that are the basis for a stable the ecology of the upper respiratory tract*
- *Report on the microbial network interactions that may be important for the levels of respiratory pathogens*
- *Provision of a computer model simulating microbial interactions of the upper respiratory tract in relation to feed ingredients and respiratory pathogens*

A microbial dynamic respiratory microbiota model has been achieved for both Dutch as well as Indonesian infants. On the level of respiratory pathogens this has not been achieved. Significantly more samples were analyzed than planned by including a Dutch as well as an Indonesian cohort. In addition, acquisition of Indonesian data based on sequencing protocols developed by partner TNO entailed technology transfer to the Indonesian study partners allowing local analysis of samples and required significant more resources. Furthermore, we established a biomarker framework (microbial health space model) to determine similarities of a feed intervention with the effects obtained for a healthy reference group. The calve respiratory microbiota under rearing conditions tested was found to be extremely complex and variable, impairing the definition of simple healthy biomarkers for a stable and healthy microbiota.

<b>Aantal opgeleverde producten</b>			
Wetenschappelijke artikelen	Rapporten	Artikelen in vakbladen	Inleidingen/ workshops
4	-	-	2

**Bijlage: Titels van de producten of een link naar de producten op een openbare website**

Early respiratory microbiota composition determines bacterial succession patterns and respiratory health in children. Biesbroek G, Tsvitvadze E, Sanders EA, Montijn R, Veenhoven RH, Keijser BJ, Bogaert D. *Am J Respir Crit Care Med.* 2014 Dec 1;190(11):1283-92. doi: 10.1164/rccm.201407-1240OC.

The impact of breastfeeding on nasopharyngeal microbial communities in infants. Biesbroek G, Bosch AA, Wang X, Keijser BJ, Veenhoven RH, Sanders EA, Bogaert D. *Am J Respir Crit Care Med.* 2014 Aug 1;190(3):298-308. doi: 10.1164/rccm.201401-0073OC.

Development of Upper Respiratory Tract Microbiota in Infancy is Affected by Mode of Delivery. Bosch AATM, Levin E, van Houten MA, Hasrat R, Kalkman G, Biesbroek G, de Steenhuijsen Piters WAA, de Groot PCM, Pernet P, Keijser BJF, Sanders EAM, Bogaert D. *EBioMedicine.* 2016 Jul;9:336-345. doi: 10.1016/j.ebiom.2016.05.031. Epub 2016 May 26.

Maturation of the Infant Respiratory Microbiota, Environmental Drivers, and Health Consequences. A Prospective Cohort Study. Bosch AATM, de Steenhuijsen Piters WAA, van Houten MA, Chu MLJN, Biesbroek G, Kool J, Pernet P, de Groot PCM, Eijkemans MJC, Keijser BJF, Sanders EAM, Bogaert D. *Am J Respir Crit Care Med.* 2017 Dec 15;196(12):1582-1590. doi: 10.1164/rccm.201703-0554OC.

Presentatie Agri Food top juni 2017 door LH Ulfman

Presentatie D. Bogaert op ESPGHAN congres als invited speaker voor FrieslandCampina (mei 2017)