



# NEOMUNE research platform – work package synopses

## WP 3.2: Probiotics in newborn mice

**1. Related WPs, MG contact:** Synergy with WPs 1.3a,1.4b,1.5,1.6b,2.2,3.1. MG contact: Hanne Frøkiær

**2. Key involved personnel, their institution, mail address (project leader + main study site underlined):**

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**3. Main aim and sub-aims:**

We aim to establish whether administration of probiotic bacteria peri- and postnatally will accelerate maturation of the gut epithelium and affect early hemapoiesis.

Sub-aims are:

a) To identify relevant probiotic bacteria or mixtures hereof.

b) To test whether administration of probiotics may compensate for effects of antibiotics treatment on development of the immune system.

**4. Background and a central hypothesis:**

If perinatal antibiotic treatment of mice as anticipated shows to compromise immune development due to a decrease in the diversity of the microbiota and/or the absence of specific genera, one way to compensate may be administration of one or more probiotic strains during antibiotics treatment.

*We hypothesize that administration of probiotic bacteria from birth, especially concomitant with antibiotic treatment may lead to a faster maturation of GI epithelium and well-balanced maturation of immunity.*

**5. Key analyses and methods:**

Perinatal antibiotics and probiotics treatment of dams and offspring mice.

To eradicate major populations of the gut microbiota, dams are treated with an antibiotics, identified in WP3.1 as a potent manipulator of immune maturation, perinatally, and probiotics are administered concomitantly.

Flow cytometry: to investigate the proportion and composition of CD11b+ cells in spleen during the first weeks of life.

Microscopy: to assess the efflux of differentiating HSC from liver and influx of cells to spleen and other organs.

RT-PCR: assess effects of microbiota on maturation of gut epithelium and cell migration in liver and spleen (up- and down regulation of chemokines, specific enzyme markers such as arginase, elastase).

16s sequencing and RT-PCR of gut contents

ELISA: e.g. cytokine/chemokine measurement of ex vivo stimulated spleen cells to assess the responsiveness to microbial stimuli.

Western blotting: e.g. assessment of enzyme (elastase, myloid peroxidase) production in cells.

Optical Projection Tomography Scanning: to assess localization of HSC in liver, spleen and gut and to identify cells in different organs expressing specific proteins, e.g. Cxcl2R.

**6. Expected results:**

To establishment if probiotic administration early in life improves immune development in antibiotic treated mice pups.



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### 7. Estimated time frame

Task	2013			2014			2015			2016			2017			2018		
Planning, protocol							x											
Sample collection								x	x									
Flow cytometry								x	x									
OPT-scanning								x	x									
16s seq/rtPCR											x	x						
rtPCR, microscopy, etc														x	x			
Publication work													x	x				

### 8. Estimated budget from NEOMUNE:

1.2 mio DKK

### 9. Estimated budget from elsewhere:

0.4 mio DKK. Salaries and equipment.

### 10. Additional comments:

- These activities will be based on results from WP 3.1. Therefore, protocols and planning are initiated late in project.