Master thesis projects within Section for Clinical Microbiology, Department of Veterinary and Animal Sciences

qPCR for mastitis diagnostics

qPCR is used increasingly for mastitis bacteriology, including microbiology samples needed for dry udder therapy. Due to the very low number of bacteria needed to obtain a positive reaction, there is a higher risk that use of qPCR leads to false positive reactions than traditional bacteriology, in the sense that the bacteria detected may just be contaminants from the environment or the skin of the udder. To investigate this, we wish to perform a small trial, where qPCR and traditional bacteriology are used in parallel to evaluate presence of mastitis pathogens in milk samples from acute cases of mastitis in cow and in milk samples send for investigation prior to dry off treatment with antimicrobials. The study is performed in collaboration with a Danish veterinary practice in Jutland, and the student(s) must expect to spend time there.

Contact: John Elmerdahl Olsen, jeo@sund.ku.dk

Improved diagnostic methods for MRSA

So far it has not been possible to construct quantitative PCR methods for MRSA, and thus it is very cumbersome to quantify this bacterium in samples from pigs and pig stables. Having such a method would make it much easier to investigate the spread of the bacterium, and it could be used to certify absence of MRSA in herds. This project is designed for a student with a desire to become better at bioinformatics. Suitable DNA target sequences will be obtained by analysis of available genome sequences MRSA CC398 on the net – we know already that it needs to be a combination of targets. The methods will be preliminarly evaluated in the laboratory for specificity, sensitivity and ability to quantify MRSA..

Contact: John Elmerdahl Olsen, jeo@sund.ku.dk

Can UV light be used to inactivate pathogenic bacteria in stables

A small Danish company has developed a light system, which one one hand can funcition as the day light for people working in the stables, and at the same time provide a non-harmfull UV light, which maybe can be used to inactivate pathogenic bacteria and virus. In this project we wish to establish a proof of concept in the laboratory for this idea, by measuring inactivation of selected pathogens in dust treated with this light and not treated with light.

Contact: John Elmerdahl Olsen, jeo@sund.ku.dk

Mastitis and biofilm treatment.

Streptococcus uberis is a major cause of intramammary infections in dairy cows and these infections impact negatively on animal welfare and lead to milk loss and severe costs for the farmers. However, lack of knowledge of *S. uberis* virulence has hampered disease control. Many pathogenic bacteria are able to form biofilm, which protects the bacteria against antibiotic-treatment and the immune response and may result in recurring infections. Since antibiotic-treatment of pathogens in a biofilm is difficult, we will in this project identify antimicrobial peptides which can destroy the biofilm. In the project it can also be possible to create mutant strains, which lacks genes involved in biofilm formation and survival under stressfull conditions.
Contact: Line Elnif Thomsen leth@sund.ku.dk

Persister cells.

Some bacteria can survive antibiotic treatment by creating persister cells, which is a dormant antibiotic-tolerant bacterial population. The change into persister is not caused by genetic changes. These persisters can ”hide” from the immune-response and survive treatment and may explain recurring infections. It has been found that, depending on the bacterial species, between 0.1 and 10% of a sensitive population is able to convert into persisters and thereby survive treatment. The aim of this project is to investigate the prevalence of Streptococcus persisters and isolate persister cells to analyse their response to different antibiotics, host stress (pH, immune response, heat) and their ability to form biofilm. Even though persister cells are important, in particular due to their antibiotic tolerance and ability to lead to recurring infections, the phenotypic switch responsible for persistence remains largely unknown. Does the host induce persister cells by accident or is treatment actually increases the risk for development of persister cells?. Understanding persister cells will in the future aid in identifying new compounds to prevent persister cell formation and/or suggest improved treatment protocols.
Contact: Line Elnif Thomsen leth@sund.ku.dk

How do treatment with 3rd and 4th generation cephalosporin induce the spread of antibiotic-resistance plasmids in E. coli?.

Antimicrobial resistance is a global health problem. We have demonstrated that treatment with 3rd and 4th generation cephalosporin affects transfer efficacy of many different R-plasmids encoding resistance to these drugs, but the underlying mechanism largely remains unknown. We have recently identified seven genes in *E. coli* where knock out of the gene abolish the antibiotic induced increase in transfer of the plasmids. The student will take responsibility for investigating the mechanism behind the effect one or two of these genes have on conjugative transfer. Elucidating the mechanisms behind antibiotic-induced spread of resistance plasmids, can lead to the identification of new targets to prevents the antibiotic induced spread.
Contact: Line Elnif Thomsen leth@sund.ku.dk

Identification of virulence factors in uropathogenic E. coli (UPEC) using TraDIS and Proteomics

UPEC cause >90% of cases of urinary tract infection (UTI) in humans.accounting for ~150 million cases globally/year representing an important health problem with significant costs. Of particular concern is the alarming increase in resistance among UPEC strains towards most of the relevant antimicrobials,. Thus, it is crucial to find new gene targets for antimicrobials to treat UTIs. For this purpose, it is essential to understand which bacterial genes are required for UPEC to infect the urinary system during a UTI. The project uses TraDIS (Transposon-Directed Insertion Site sequencing, combining traditional random transposon (Tn) mutagenesis with DNA sequencing and allowing the identification of essential genes under particular infection conditions) and preoteomics, The TraDIS library generated in a UPEC strain has been tested during growth in human urine and infection of the mouse bladder. Bioinformatic analysis has revealed a list of genes required for UPEC to cause UTI. UPEC proteins up-regulated during UTI have been also identified.The aim of this project is to construct the mutants for the genes and validate them under UTI conditions (growth in urine, infection of bladder cell lines and animals models of UTI infection) . This project provides the students with good possibilities to practice molecular microbiology techniques including mutagenesis techniques for inactivation of selected genes (i.e via CRISPR) and to carry out virulence characterization of mutants through the infection of cell lines and animals models. The student will work with two to five mutants that seem to be required for UTI in the initial in silico screening and confirm their importance for UPEC virulence.

Contact: Ana Herro-Fresno, ahefr@sund.ku.dk

Metabolsim of Avian Pathogenic E. coli (APEC) as a target for development of new antimicrobials

APEC is the infectious agent of a wide variety of avian diseases, which causes substantial economic losses to the poultry industry worldwide. In a recent project on APEC metabolism, by using a proteomic approach (APEC salpingitis proteome) and a genome scale metabolic model (GSMM), we have identified/predicted metabolic pathways (focus on redundant metabolic pathways) and virulence factors that seem to be involved in APEC growth and/or pathogenicity. These pathways might be potential targets for development of new antimicrobials against APEC. In this project, students have to disrupt the selected genes (resulting from modelling predictions –redundant reactions- or from the proteome study) and obtain the derived mutants which will be further validated over gowth (in rich and mimimal media) or across different infection conditions (i. e infection of chicken). The student carrying out this project will develop skills on molecular microbiology (i.e. mutagenesis techniques), infection of cell lines and the chicken model of infection.

Contact: Ana Herro-Fresno, ahefr@sund.ku.dk

Vaccine development for prevention of post-weaning driarrhea (PWD) in pigs

PWD is costly to swine production and affects welfare of pigs. Vaccination is suggested as the best solution for prevention of the disease. In a current project, antigens (vaccine candidates) from main PWD-associated pathogens; *Escherichia coli, Lawsonia intracellularis* and *Brachypira pilosicoli* have been selected and obtained following a cutting-edge approach for vaccine development. The approach is based on the use of capside-like particles (CLP) from pahges where several antigens can be simultaneously attached. The project involves the study of the efficacy of the potential vaccines (CLP-derived antigens) in several animal models as well as other related studies (i.e neutralization assays via infection of cell lines).
The student will get knowledge on vaccine development and will be familiar with assays required as part of the vaccine validation process.
Contact: Ana Herro-Fresno, ahefr@sund.ku.dk

The role of dedA, wabN and galE in colistin resistance of Klebsiella pneumoniae

Colistin is a last resort drug for treatment of severe hospital infections caused by *Klebsiella pneumoniae* and other Gram-negative pathogens. We have identified nonessential genes required for colistin-resistant *K. pneumoniae* to grow in the presence of therapeutic concentrations of this antibiotic (Jana et al. 2017). Individual deletion of three selected genes (*dedA*, *wabN* and *galE*) restored colistin susceptibility, resulting in a reduction of the MIC from 8 to 1 µg/ml. However, it is unknown how the proteins encoded by these genes influence the colistin resistance phenotype. The objective of this project is to understand whether these mutants can re-acquire resistance following exposure to colistin and which mutations are involved. The student will be trained to select mutants with increased MIC of colistin and to detect changes in their genome by Whole Genome Sequencing (WGS). The information generated by this project will be useful to understand i) how DedA, WabN and GalE interfere with the mechanism of colistin resistance and ii) if and how resistance would develop if one of these proteins was inhibited by a putative colistin helper drug.

Key reference: Jana B, Cain AK, Doerrler WT, Boinett CJ, Fookes MC, Parkhill J, Guardabassi L. 2017. The secondary resistome of multidrug-resistant *Klebsiella pneumoniae*. *Sci Rep* 7:42483.

Contact person: Luca Guardabassi (lg@sund.ku.dk).

The effects of different β-lactam antibiotics on ESBL selection in pig fecal microbiota

Preliminary *in vitro* data generated by competition experiments using two isogenic strains with and without ESBL have indicated that ESBL-producing *Escherichia coli* are selected by low concentrations of amoxicillin-clavulanic acid but not by the same concentrations of amoxicillin alone. The objectives of this project are i) to confirm this finding *ex vivo* in pig feces and ii) to determine whether the selective effect of clavulanic acid is mainly due to selection of the resistant strain or plasmid transfer. The student will be trained to perform experiments using a fluorescence-based method specifically designed to track the fate of an ESBL-producing strain in complex microbial communities (Anjum et al. 2018). The chromosome and the ESBL-carrying plasmid of this strain are labeled with different fluorescence markers, allowing counts of donor and transconjugants by flow cytometry. Strain survival and plasmid transfer in pig fecal samples under exposed to different antibiotic concentrations will be monitored using fluorescence-activated cell sorting (FACS). The results of this project will contribute to understand whether the use of amoxicillin in combination with clavulanic acid implies a higher risk for selecting ESBL-producing strains in comparison to amoxicillin alone.

Key reference: Anjum M, Madsen JS, Espinosa-Gongora C, Jana B, Wiese M, Nielsen DS, Sørensen SJ, Moodley A, Bortolaia V, Guardabassi L. 2018. A culture-independent method for studying transfer of IncI1 plasmids from wild-type *Escherichia coli* in complex microbial communities.

Contact person: Luca Guardabassi (lg@sund.ku.dk).

Mapping of diagnostic microbiology practices among veterinarians in Denmark

It is unknown how often veterinarians in Denmark send samples to diagnostic laboratories, what kind of samples they send and to which laboratories. The aim of this project to gain a better insight into the diagnostic practices used by Danish veterinarians for different animal species and production systems. The student will be trained to design and conduct an electronic survey targeting different types of veterinary practices (e.g. pig, cattle, horse and small animal practices) in liaison with the Danish Veterinary Association. The results of the survey will allow identification of differences associated with animal species, production systems, as well as geographical differences. Moreover, it will provide information on the factors that influence the choice of submitting samples to diagnostic microbiology laboratories and on the frequency of in house diagnostic laboratories. It is expected that the results of the survey will be published in an international scientific journal and a Danish veterinary magazine (Veterinær Tidsskrift).

Key reference: De Briyne N, Atkinson J, Pokludová L, Borriello SP, Price S. Factors influencing antibiotic prescribing habits and use of sensitivity testing amongst veterinarians in Europe. *Vet Rec*. 2013;173(19):475. doi:10.1136/vr.101454

Contact person: Luca Guardabassi (lg@sund.ku.dk)

Development of diagnostic tests for typing of Mannheimia haemolytica

Problem: *Mannheimia haemolytica* is contributing to important respiratory diseases in cattle and the bacterium is responsible for major economic loss, excess use of antibiotics and animal welfare problems. In order to use killed vaccines the serotype needs to be determined. Traditional serotyping is no longer performed and PCR methods are only available for serotypes 1, 2 and 6. There is a need to develop new diagnostic test for the other serotypes. Project: The aim of the project is to extend the prediction of the serotypes based on whole genome multi-locus sequencing (wgMLST). All reference strains for the serotypes have been whole genome sequenced. The student will need to plan and analyse sequencing of more strains and to extend the wgMLST system already established on the BIGSdb platform (ivsmlst.sund.ku.dk). The outcome of the project will be a tool that can predict the serotype from the whole genomic sequence via the internet. The project is carried out in collaboration with a German diagnostic laboratory. Knowledge of bioinformatics is beneficial, but not necessary, as training can take place during the project period.

Contact person: Henrik Christensen hech@sund.ku.dk

Detection of bacteria comparing different MALDI-TOF platforms.

Problem: MALDI-TOF (matrix assisted laser desorption / ionization time of flight) has become a standard method for detecting bacteria within clinical microbiology as well as food and environmental microbiology. Two platforms with different equipment and databases are used: MALDI Biotyper (Bruker) and VITEK MS (BioMérieux). Species identification is not always reliable and might differ between the systems.

Aim: The project includes a comparison of the reliability of detection with the two systems as well as analysis of data including work with the databases included in the detection systems. In addition the aim is to set up a common dataformat for both systems. The student needs to investigate selected bacterial species on both platforms and to use control methods such as DNA sequence-based detection. The student will be able to influence the choice of model organisms that are included in the study. Fungi of importance in skin infections can be investigated. The project will be carried out in collaboration with a larger Danish biotec company.

Contact person: Henrik Christensen hech@sund.ku.dk

Selection of probiotic bacteria to prevent pathogenic E. coli.

Problem: Antibiotic resistance is a problem in relation to the control of infections with pathogenic *E. coli* in production animals and alternative strategies to control such infections are needed. Probiotic bacteria are used as feed additives to improve the intestinal health and increase growth performance of production animals. Unfortunately, little has been done to investigate the effect of probiotics on pathogenic *E. coli*. Project: The student will need to screen a collection of probiotic candidate bacteria for their inhibitory effects on *E. coli*. Preliminary results are available for a collection of isolates obtained from chicken but the student is also welcome to isolate probiotic candidate bacteria in the project. The project will have to focus on the production of bacteriocins and similar secondary metabolites. The project will provide the student with a background to work with probiotics in the biomedical industry and the candidate will be trained in the major methods used in microbiological characterization.

Contact person: Henrik Christensen hech@sund.ku.dk

 

In vitro studies of G+ (and G-) organisms and the development of serum amyloid (SAA) and the association with amyloid isolated from birds with amyloid depositions

Systemic amyloidosis is a pathological manifestation causes by a chronic inflammatory response. The condition is mainly observed in adult poultry and in some years with a high prevalence. Recent investigations have shown a positive correlation with chronic infections (Gr+ cocci in particular) and pronounced amyloidosis. The depositions are due to misfolding of the circulating serum amyloid making it unsoluble. The misfolding of amyloid has been suggested to be caused by bacteria also producing amyloid-like proteins which may induce the misfolding. The project aims at elucidating the pathogenesis of the condition by investigating the association between common bacterial infections in poultry and their ability to induce amyloid depositions.

Contact persons: Jens Peter Christensen jpch@sund.ku.dk and Ida Thøfner icnt@sund.ku.dk

 

Experimental infections of the oviduct and the resulting immune response

Eschericia coli is the main cause of salpingitis in laying hens. We know from previous investigations that there is a major individual variation in the response to infection even if birds are infected with the same number of bacteria of the same isolate– some birds will clear the infection fast while others may die from the same infection. Poultry differ from mammals concerning the lymphatic tissues as they do not possess regional lymph nodes from where immunocompetent cells can be recuited. However, they do have a strong mucosal response which often is highly effective. Few investigations have been performed where detailed information concerning the mucosal response has been obtained. From previous experimental *E. coli* infections of the oviduct we have obtained serum, circulating immuno-cells and tissue samples which can form the basis for further investigations.

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The establishment of a primary salpinx cell culture

For the investigation of the interaction between *Eschericia coli* and the mucosa of the salpinx by in vitro methods, we would like to develop a in-house protocol for isolation, cultivation and infection of epithelial cells from the salpinx. In relation to this, bacterial and host specific factors concerning infection of the cell cultures should be investigated. The results obtained will add to the understanding of the variable outcome of infections.

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Effect of artemisinine on the immunesystem during experimental coccidia infections

Coccidia are the most important parasites affecting poultry and are controlled primarily by the use of coccidiostats in the feed. However, the compounds are similar to some growth promoters and resistance may develop. Consequently, it is necessary to identify new ways to control coccidial infections. A compound use to control malaria, artemisinine, has been shown to have a positive effect on the outcome of infections but the mode of action is still unknown. In relation to a experimental study using artemisinine for chickens infected with Eimera tenella, histological investigations showed a diverse cell response depending on the specific treatment. Further investigations of tissues sampled during infection will elucidate wether the observed positive effect of artemisinine is due to a modification of the immune response or due to a direct anticoccidial effect.

Contact person: Ida Thøfner icnt@sund.ku.dk

Keel bone fractures in laying hens

Keel bone fractures are observed with a high prevalence in laying hens from any producyion system (barn, organic and enriched cages). We have more ongoing projects within this area. Prevalences studies in different production systems, risk factor analysis and pathogenesis studies. The latter studies are performed together with Department of forensic medicine at Rigshospitalet where CT scanning and advanced histopathology techniques are used. It is possible to do a project within these areas of research.

Contact. Jens Peter Christensen jpch@sund.ku.dk

Assessment of microbiological diagnostics performed in veterinary practices

Veterinary practitioners regularly culture and perform antimicrobial susceptibility testing (AST) in their own practice. This saves time and money compared to sending samples to professional diagnostic laboratories. However, there are many pitfalls associated with microbiological diagnostics, and if not performed according to standards, animals may suffer from inappropriate treatment resulting in treatment failure – or overtreatment resulting in risks of toxicity and selection of antimicrobial resistance. **In this project**, you will map how veterinarians practice microbiological diagnostics in their practice. This can be done in different ways, for example by a questionnaire-based online survey or by interviewing veterinarians. At the same time, you may assess methods and interpretive criteria by processing animal specimens in duplicate in veterinary practices and in our diagnostic laboratory. Alternatively, you may set up a ring trial sending out "fictive samples" to veterinary practices and assessing their subsequent results from culture and AST. All in all, this project can be twisted in different directions towards microbiology and/or epidemiology depending on your wishes.

Contact person: Peter Damborg pedam@sund.ku.dk

Active surveillance of antimicrobial resistance in pigs

In early 2020, a new project on surveillance of antimicrobial resistance (AMR) in pigs will start. The project aims to provide a detailed overview of AMR in the Danish pig population, both by assessing existing diagnostic data, and by actively searching for resistant bacteria of high clinical relevance for human health. This may – for example - be done by selective screening of samples using antibiotic-supplemented agar, and subsequent analysis of isolates using various techniques including maldi-tof, antimicrobial susceptibility testing, PCR, and whole-genome sequencing. We will also develop a novel point-of-care snaptest for rapid identification of resistant enterotoxigenic *Escherichia coli* in farms. **As a student on this project**, you may join in sampling of farms and lab-based screening of bacteria, or you may join the development and validation of the new snaptest.

Contact person: Peter Damborg pedam@sund.ku.dk

Antimicrobial resistance in dogs and cats

Our diagnostic laboratory processes thousands of samples from dogs and cats every year. This results in gigabytes of antimicrobial susceptibility data and hundreds of resistant bacterial isolates stored in our freezers. **In this project**, you will analyze isolates that we have stored due to a suspicion that they may be methicillin-resistant staphylococci or extended-spectrum betalactamase (ESBL)-producing *Escherichia coli*. Isolates can be investigated by PCR- and sequence-based identification of resistance genes and bacterial lineages. At the same time, you may retrieve and analyze prevalence data on antimicrobial susceptibility. The project is "home safe" with guaranteed results, which will help us in optimizing antimicrobial treatment guidelines for companion animals. Most likely, your results can be published, for example in DANMAP, as the editors of this surveillance system have requested data for companion animals.

Contact person: Peter Damborg pedam@sund.ku.dk

European Network for optimization of Veterinary antimicrobial Treatment (ENOVAT)

At the end of 2019, a large European network of more than 30 countries aiming to optimize antimicrobial treatment of animals will commence. Over a 4-year period, the network will optimize microbiological diagnostic practices and antimicrobial treatment guidelines for animals through various workpackages. As part of this project, young scientists and veterinary students will be granted scholarships and sent to other research groups in order to learn microbiological techniques and to conduct studies such as MIC testing of local bacterial isolates. MIC results will contribute to the development of breakpoints used for antimicrobial susceptibility testing of veterinary pathogens. So, if you wish to travel in Europe, be part of a large international network, learn more about veterinary microbiology, and do research for the sake of animal health – then contact me and hear what is possible.

Contact person: Peter Damborg pedam@sund.ku.dk

Zoonotic aspects of swine influenza viruses

Some strains of swine influenza viruses have the capability to cross the species barrier and infect humans and by that provide a risk of starting a new human influenza pandemic as seen in 2009. We have received a major grant from Novo Nordisk Foundation to a project with the primary aim to identify and validate reliable molecular and/or immunological markers for the zoonotic potential of swine influenza viruses. In the frame of this project, there are a number of attractive master project available that will be performed in cooperation with PhD students and post docs at SSI, DTU and St. Jude Childrens Hospital in Memphis, USA..ie. a research stay in the US may be offered. The projects will involve laboratory work within virology, molecular biology, bioinformatics, immunology or pathology; or practical work with animals though participation in experimental trials in ferrets and swine. These projects are especially interesting for Biomedicine students that wants to work with research or in the medical industry after graduation. Examples of projects - in headlines - are listed below:

- Deep sequencing and bioinformatics analyses of influenza A virus strains collected from humans and swine
- Determination of receptor preferences of different swine influenza isolates
- Establishment of *ex vivo* organoids (trachea, lungs etc) of swine and humans for the study of influenza virus pathogenesis
- Participation in experimental infection of swine and ferrets with influenza virus and test of samples for virus and immunological mediators
- Participate in generation of genetic modified influenza virus

Contact: Lars Erik Larsen; lael@sund.ku.dk

Comparative study of influenza A virus infection in different animal species such as mink, swine and seals.

The aim of this project is to compare the pathology of influenza A infections in different animal species, and establish immunohistochemical methods for detection of influenza virus and receptors in tissues from a range of influenza A susceptible animals including swine, poultry and mink and wildlife species such as seals, wild birds and whales. The project is cooperation between the virus and pathology groups at IVH.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Characterization of Newcastle disease (ND) viruses from wild birds.

The purpose of the project is to clarify which variants of Newcastle disease (ND) virus, are circulating in wild birds in Denmark. ND viruses are causing devastating disease in poultry in unvaccinated populations world-wide. Different variants of the virus are known to cause varying degrees of disease, and new variants emerge as the result of the high mutation frequency of RNA viruses. Characterization of ND viruses is an ongoing activity in many of our surrounding neighbouring countries, while there is only a sparse knowledge about the ND viruses, circulating in Denmark. In this project, newer Danish ND viruses will be characterized by means of PCR, sequence analysis and phylogenetic analysis.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Avian influenza.

In the frame of a PhD project we are analyzing the diversity and pathogeneses of HPAIV . We have planned experimental infections in Pheasants to study the transmission dynamics, viral pathogeneses, immune responses etc. A range of master projects are available focusing on different aspects of the infection by the use of virological, pathological and immunological methods. The project is cooperation with SSI and a research group in the UK.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Vaccination responses to Newcastle Disease Virus (NDV)

The purpose of the project is to investigate if the present, mandatory vaccination program against NDV provide suefficient protection during the lifespan of layers. Layers are vaccinated against NDV prior to the start of laying and are expected to be protected against NDV infection thoughout the laying period. The lifespan of layers has, however, increased considerable during the last ten years, but there is a lack of studies that have documented the duration of immunity under these conditions. In this project, blood samples will be collected from layers at different ages and tested for antibody against representative isolates of NDV using hemagglutination inhibition assays and serum neutralization tests.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Vaccination strategies of horses against influenza virus

Most horses in Denmark are vaccinated against Influenza A virus using commercial available vaccines. In addition to the primary three vaccinations spanned by 3-5 months most vaccine companies recommend booster vaccination once yearly. In general, the duration of immunity after parential vaccination with an inactivated vaccination are shortlasting so it is not know if annual revaccination against influenza virus in horses are adequate. The aim of this project is to analyse the level of antibodies in horses one year after the last vaccination. In cooperation with horse practisioners, blood samples will be collected from horses in connection to their booster vaccination. The samples will be tested for antibodies against influenza A virus by ELISA, hemaagglutination test and serum neutralization tests

Contact: Lars Erik Larsen; lael@sund.ku.dk

Virus in dogs, cats and horses in Denmark

Only very limited diagnostics and research are performed on cats, dogs and horses in Denmark and therefore there is a profound lack of data on the prevalence and diversity of circulating viruses in these animals. Each master project may focus on one of the animal species and on a selected number of viruses such as parvo virus in dogs and cats, herpesvirus or equin arteritis virus in horses. The student will be involved in the establishment of real time PCR assays for detection of the viruses and methods for sequencing by either SANGER or on one of our NGS platforms. Samples will be collected from the small animal clinic at KU SUND, from other ongoing projects and/or from other veterinary practices.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Virus in chickens in Denmark

Only very limited diagnostics and research are performed on commercial chicken herds in Denmark and therefore there is a profound lack of data on the prevalence and diversity of circulating viruses in this segment. Each master project may focus on a selected number of viruses such as infectious bronchitis virus, Gallid herpes virus 2 (Mareks disease virus) or one of the retroviruses. The student will be involved in the establishment of real time PCR assays for detection of the viruses and methods for sequencing by either SANGER or on one of our NGS platforms. Samples for testing will be collected from commercial herds or from archived samples stored at KU-SUND. The project will be a cooperation between the virus group and the avian disease group (Professor Jens Peter Christensen)

Contact: Lars Erik Larsen; lael@sund.ku.dk

Important viruses in Danish swine

We have several ongoing projects on important viruses viruses in Danish swine, including PRRSV, swine influenza virus, PCV2, rotavirus etc. The projects are focusing on characterization of viruses by molecular techniques (PCR, sequencing), experimental trials, optimized vaccination protocols, transmission dynamics, but are also open for new ideas. The project may either be focused on laboratory work or field studies according to the intereste of the student.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Important viruses in Danish cattle

We have several ongoing projects on important viruses viruses in Danish cattle. Specific planned projects include studies on a rather new viruses in Danish cattle – influenza D virus, but we are also interested in studies on BRSV, Bovine coronavirus and rotavirus in the frame of a larger project – Rubuste kalve. The projects are focusing on characterization of viruses by molecular techniques (PCR, sequencing), optimized vaccination protocols, transmission dynamics, but are also open for new ideas. The project may either be focused on laboratory work or field studies according to the intereste of the student.

Contact: Lars Erik Larsen; lael@sund.ku.dk

Help make the final step toward overcoming current limitations with qPCR for diagnostic purposes in veterinary clinics

qPCR is used widely in clinical microbiology, however, it has the limitation that you cannot ask questions to which antimicrobials to use, when you detect and quantify a bacteria by this method. This limitation may be overcomed by running relevant qPCR methods for important resistance genes in parallel. We wish to test this idea in a test system, set up to build a warning system against multi-drug resistance (MDR) in pathogens associated with pneumonia in cattle (*Mannheimia haemolytica, Histophillus somni, Pasteurella multocida*). Currently, there is no treatment problem with these bacteria, but MDR is on the rise in other parts of the world. We have designed methods, and now we seek students how will test the system in the laboratory using tracheal samples from healthy calves, which we spike with sensitive and MDR bacteria.

Contact: Egle Kudirkiene egle@sund.ku.dk

Differentiation of blood samples collected from domestic swine and wild boar

African swine fever has become a major threat to the pig production industry across Europe and Asia. The disease is being spread, in part, by transmission between wild boar and from wild boar t domestic pigs. During summer months, there is an increase in the number of outbreaks in domestic pigs and this may be linked to an increase in insect activity. It has been shown that blood-feeding insects can contain sufficient virus to enable mechanical transmission of the virus to domestic pigs. Thus if blood feeding insects take blood from ASFV-infected wild boar and then enter a premises with domestic pigs then transfer of the virus may occur. It is important to be able to determine if blood fed insects, captured within a pig stable, contain blood from wild boar or from only the domestic pigs. In principle, it is possible to discriminate between blood from these two different sub-species on the basis on sequence differences within the mitochondrial cytochrome b gene. The project will aim to establish an assay suitable for the discrimination of blood samples from domestic pigs and wild boar using PCR and DNA sequencing. It can then be tested on samples of blood fed insects.

This project will be performed in collaboration with the Statens Serum Institut.

Contact: Graham J. Belsham (grbe@sund.ku.dk)

Bat species identification using PCR.

There are 17 different species of bats present within Denmark. Different species have their own types of viruses. Evidence for a variety of different bat coronaviruses (closely related to the porcine epidemic diarrhea virus) present within these distinct bat populations has been obtained. Using fecal samples from bats, it is possible to detect these different viruses, and the source of the samples, including the specific bat species, can be identified based on sequencing of PCR amplified fragments corresponding to the mitochondrial cytochrome b gene and also the mitochondrial 16S ribosomal RNA gene. A collection of bat samples, collected during passive surveillance, is available and the specific bat species represented by these samples needs to be established using these assays.

This project will be performed in collaboration with the Statens Serum Institut.

Contact: Graham J. Belsham (grbe@sund.ku.dk)