

## Program: Specialedag.

- 13:00 – 13:15**    **Theresa June Winther Pedersen:** Glucose metabolism in *Python regius*
- 13:15 – 13:30**    **Line Hermanssen:** Effects of shipping noise on harbor porpoises in Aarhus Bay
- 13:30 – 13:45**    **Jesper Madsen:** Measurements of evoked potentials and behavior in the transition to cold hypoxia in the turtle brain.
- 13:45 – 14:00**    **Kristina Ydesen:** Prey engulfment in phocid seals studied with high speed cameras and accelerometry tags
- 14:00 – 14:15**    **Christian Bech Christensen:** From underwater larvae to terrestrial adults: Hearing across the metamorphosis in salamanders
- 14:15 – 14:30**    **Pause**
- 14:30 – 14:45**    **Inge G. Revsbech:** Increase in blood oxygen affinity during hibernation in brown bear *Ursus arctos*
- 14:45 – 15:00**    **Simon Uphill:** Lack of metabolic depression during aestivation in the Asian Swamp eel *Monopterus albus*
- 15:00 – 15:15**    **Anete Dudele:** The role of low grade chronic inflammation on development of cardiovascular disease and epigenetic programming of offspring disease
- 15:15 – 15:30**    **Karina Vincents Fisker:** No costs of adaptation on life-history traits, population growth rate and freeze tolerance in genetically copper adapted earthworm populations
- 15:30 – 15:45**    **Anders Eriksen:** Seasonal migration of roach (*Rutilus rutilus L.*) in a shallow Danish lake: The role of sex, morphology and body-condition
- 15:45 – 16:00**    **Pause**
- 16:00 – 16:15**    **Gudrun Winther:** Magnesium deficiency induces anxiety- and depression-like behavior and metabolic dysfunction in C57Bl/6J mice
- 16:15– 16:30**    **Denise Lyager Jensen:** Aerobic scope in summer and winter acclimated perch (*Perca fluviatilis*)
- 16:30 – 16:45**    **Annemette Møller-Sørensen:** Glucose metabolism and phenotypic flexibility during fasting in rats
- 16:45 – 17:00**    **Sanne Enok:** The role of gastric digestion on specific dynamic action and postprandial organ growth in pythons (*Python regius*)
- 17:00 – 17:15**    **Tine Østergaard Larsen:** Arterial extracellular matrix changes in a murine model of type 1 diabetes

## **Theresa June Winther Pedersen: Glucose metabolism in *Python regius***

The reptilian pancreas contains insulin and glucagon, however regulation of glucose is poorly understood and differences between fasting and digestion has received little attention. The effect of digestion and surgery on blood glucose and effects of fasting and digesting on glucose tolerance in *Python regius* was investigated. To evaluate glucose tolerance, snakes (n=3) fasted for 8-10 weeks and snakes (n=3) digesting a large meal (10% of body mass), received a glucose bolus (1g/kg) and the return to initial blood glucose values was monitored. Surgery led to a 37% increase in blood glucose. Blood glucose was unaffected by digestion. Both fasting and digesting snakes experienced prolonged hyperglycemia upon glucose administration, and took 18 and 10 h respectively, to normoglycemia. The faster glucose clearance during digestion could be attributed to increased metabolic rate and/or improved control. The underlying glucoregulatory mechanisms will be the subject of my MSc research.

## **Line Hermanssen: Effects of shipping noise on harbor porpoises in Aarhus Bay**

Danish waters are heavily ship-trafficked, and shipping noise may affect marine mammals negatively by impeding their use of sounds for echolocation and communication. In this study I recorded noise in Aarhus Bay to investigate the effect of shipping noise on ambient noise levels. I find that both conventional and fast-ferrries emit significant levels of high-frequency noise in the range of harbor porpoise echolocation (100-150kHz). Contrary to common beliefs the slower conventional ferry emitted the highest levels of high-frequency noise, potentially causing masking for porpoises more than 1000m away. To obtain a better assessment of ship noise levels, and thereby a more exact estimate of the impacts on harbor porpoises, noise recordings with longer timeframes and at varying distances from ferrries approximating the attenuation coefficient, are needed to address the scale of impact of anthropogenic noise, with relevance for the ongoing planning of an improved future for the Baltic Sea.

## **Jesper Madsen: Measurements of evoked potentials and behavior in the transition to cold hypoxia in the turtle brain.**

The exposure of brain tissue of hypoxia or anoxia can cause severe neurological damage in many animals. However, several species of freshwater turtles and fish are known to survive extended anoxia with no adverse effects. The turtles have been reported as entering a non-responsive comatose state while the fish remain vigilant, but is this really the case, and how responsive are turtles in transition into coma? To test the hypothesis of comatose during cold anoxia, turtles will be exposed to vibration stimuli, during which evoked potential measurements are carried out in the transition into and during cold anoxia. Preliminary results indicate that as temperature drops the amplitude of measured potentials also drop. It is expected that exposing the turtles to anoxia will lower the amplitude even further. This is in accordance with my expectations that turtles display gradual lowering of stimulus response which finally disappear completely, if turtles do enter coma.

## **Kristina Ydesen: Prey engulfment in phocid seals studied with high speed cameras and accelerometry tags**

Quantification of the foraging behaviour of marine mammal predators in the wild is fundamental for understanding their ecological role, but no reliable means to get information on prey capture success are available. Thus, a new technique is needed to quantify prey captures over long time series in the wild. Specific movements of the head and jaws may provide reliable feeding cues by involving fast specific changes in acceleration (jerk signatures) that might even be prey or context specific. To test this, underwater prey captures of two trained harbour seals wearing Dtag3, with triaxial accelerometers sampling at 500Hz, on their head, will be filmed with two high-speed video cameras. The hypothesis is that feeding produces prey or context specific jerk signatures that can be related to actual prey captures. If verified, the prey capture success and energetic turnover of pinnipeds in the wild can be measured over a year.

**Christian Bech Christensen: From underwater larvae to terrestrial adults:  
Hearing across the metamorphosis in salamanders**

Since animals went on land in the Carboniferous period the tympanic middle ear has evolved independently at least five times in all the major tetrapod lineages and is believed to play a crucial role in hearing of most terrestrial animals. But how does the change from an underwater to a terrestrial lifestyle affect the hearing of animals that lack a tympanic ear? In this study I wish to investigate the hearing and vibration detection in air and water of the axolotl (*Ambystoma mexicanum*) before, during and after the metamorphosis as it emerges from a completely underwater to a partly terrestrial lifestyle. Further, I wish to examine how the anatomy of the ears changes during the metamorphosis using CT and MRI scanning techniques. The results may give new perspectives on the role of the tympanic ear and hearing of animals with atympanic middle ears.

**Inge G. Revsbech: Increase in blood oxygen affinity during hibernation in brown bear *Ursus arctos***

Blood samples were taken in the wild from 6 individuals of Scandinavian brown bear during winter hibernation and summer active states. Oxygen binding curves performed at a constant pH (7.40) on the thawed red blood cells (37 and 30°C for both winter and summer samples) showed increased oxygen affinity during hibernation, also when compared at the same temperature. The increase in oxygen affinity was associated with a significant decrease in red cell cofactor 2,3-DPG during hibernation, approximately to half the summer values (means  $1.06 \pm 0.10$  vs  $0.49 \pm 0.19$  DPG/Hb). Hemoglobin (Hb) cellular concentration and chloride concentration in the RBCs did not show any significant change with hibernation. Isoelectric focusing showed the presence of a single Hb component in both summer and winter samples, indicating that no alternative Hb isoforms are upregulated during hibernation. Nitric oxide metabolites measured in RBCs and plasma showed no change with hibernation. These results indicate an increase in the blood oxygen affinity of brown bears during hibernation that can be attributed to a combination of changes in red cell DPG and in body temperature. Reduced oxygen offloading during hibernation may thus contribute to the 75% reduction in basal metabolism during hibernation, as recently described in black bears.

## **Simon Uphill: Lack of metabolic depression during aestivation in the Asian Swamp eel *Monopterus albus***

The air-breathing Swamp eel *Monopterus albus* aestivates in mud during the Asian hot and dry season to escape unfavourable conditions. Previous reports are unclear concerning burrow construction and whether animals are metabolically depressed, but it has been suggested that environmental hypoxia acts to lower metabolism. We fitted *M. albus* with interperitoneal heart rate ( $f_H$ ) transmitters and measured aerial oxygen uptake ( $VO_2$ ) and breathing frequency ( $f_V$ ) during stepwise increasing hypoxia. *M. albus* compensated by increasing  $f_V$  but maintained constant  $VO_2$  down to an ambient 5%  $O_2$ .  $f_H$  decreased gradually between 20% and 5%  $O_2$ . At 2%  $O_2$ ,  $f_H$  and  $VO_2$  dropped sharply to 33% and 19% of normoxic values, respectively, while  $f_V$  increased more than 23 times. All parameters quickly returned to normoxic levels after 2 h at 2%  $O_2$ , and there was no apparent  $O_2$  debt. These animals subsequently burrowed into mud for estivation.  $VO_2$  measurements were no longer possible, but all fish maintained a small opening to the air.  $PO_2$  was measured in the fish chamber using an optode to be 13%, well within the range where *M. albus*'s could maintain SMR.  $f_H$  was slightly reduced during aestivation, possibly due to the state of torpor. We conclude, that *M. albus* is able to cover its metabolic requirements with the  $O_2$  available during estivation and has no need of a significant metabolic depression. The effects of the prolonged fasting on metabolism and the fate of metabolically produced  $NH_3$  should be the focus of further experiments.



## **Anete Dudele: The role of low grade chronic inflammation on development of cardiovascular disease and epigenetic programming of offspring disease**

Despite the extensive research within obesity related diseases and their inheritance, the underlying pathophysiological mechanisms remain largely unknown. There is ample evidence that overeating can induce a chronic low grade inflammation – a reaction that illustrates the close evolutionary relationship of animal metabolic and immune systems. In the current conditions where availability of calories to human population is vast and increasing urbanisation reduces overall physical activity, formerly advantageous adaptation has become detrimental. Chronic inflammation leads to insulin resistance, increased food intake and depressive like behaviour. The aim of the study is to investigate the role of chronic low grade inflammation in development of cardiovascular disease in adult rats and its importance in maternal programming of the offspring towards development of metabolic syndrome in later life.

**Karina Vincents Fisker: No costs of adaptation on life-history traits, population growth rate and freeze tolerance in genetically copper adapted earthworm populations**

For around three centuries the area around Gusum (Sweden) has been highly copper polluted, which affect the earthworm populations living there. Earthworms are known to cope with high copper burdens; however, the distinction between adaptive responses, evolutionary heritable changes, and associated fitness consequences of the adaption to long-term copper exposure has rarely been studied. To investigate adaptation of earthworm populations (*Dendrobaena octaedra*) to copper contamination, three populations from polluted soil near Gusum were studied, and compared to three unpolluted populations. A transplant experiment and a combined freeze and copper experiment were performed on the laboratory F1-generation to study if any costs were occurring due to adaptation. The overall conclusions of this study are that worms from polluted sites seem to be better in handling copper, and that no costs, in terms of reduced population growth rate and/or reduced cold tolerance, were identified in connection to genetic adaptation in these populations.

**Anders Eriksen: Seasonal migration of roach (*Rutilus rutilus L.*) in a shallow Danish lake: The role of sex, morphology and body-condition**

A number of studies have described how cyprinid fishes in shallow lakes migrate to connected streams in autumn and overwinter outside the lake. It's not the whole population which participates in the migration and the migration is therefore partial. It's believed that the individuals' decision to migrate is a trade-off between food resources and the risk of predation. This study investigates the differences between resident and migrating individuals of a common Cyprinid, Roach (*Rutilus rutilus L.*), in proportion to: sex, body condition and morphology, in a shallow and eutrophic Danish lake, from October 2010 to March 2011. There were no differences in the sex-distribution between the habitats. The roaches inhabiting the two habitats had different morphology (body-depth). The muscle water content increased and body-condition decreased from November to March in the roaches caught in the stream, which indicate that they're using up body reserves.

**Denise Lyager Jensen: Aerobic scope in summer and winter acclimated perch (*Perca fluviatilis*)**

Recent studies have shown that a temperature increase of only a few degrees can have dramatic effects on the performance of aquatic ectothermic animals; it is likely that aquatic ecosystems and commercial fisheries will be affected by current and future climate change. There is, however, at present an insufficient scientific basis to allow for a qualified prediction of the consequences of the predicted global warming, and it is also largely unknown to what extent acclimation can buffer the impact of temperature change. To contribute to a better understanding of this issue, it is the aim of this master to study the interaction between oxygen transport capacity and temperature tolerance of the European perch (*Perca fluviatilis*). Specifically, the relationship between cardiac performance and thermal tolerance will be studied by measuring aerobic scope, temperature tolerance, heart rate, and force development of the heart at different temperatures in two groups of fish acclimated to summer and winter conditions, respectively.

## **Annemette Møller-Sørensen: Glucose metabolism and phenotypic flexibility during fasting in rats**

Many animals experience periods of food deprivation under natural conditions and respond at different levels to reduce metabolism. Given that the digestive organs are considered energetically expensive, considerable emphasis has been placed on the ability to down-regulate the size and function of these organs during fasting. The actual costs of maintaining structure and function, however, have not been studied directly. In the present study, we measured glucose metabolism of various organs in fasting rats. The rats were divided into five groups of different digestive state. The metabolism of the visceral organs was determined by the uptake of  $^{18}\text{F}$ -FDG; a radioactive tracer that is metabolically trapped in the cells. The  $^{18}\text{F}$ -FDG uptake is considered to be proportional to glucose metabolism. An hour after injection of the tracer the rats were sacrificed, and tissue samples were counted. The results so far indicate that the digestive system is responsible for 12 % of BMR irrespective of digestive state.

## **Gudrun Winther: Magnesium deficiency induces anxiety- and depression-like behavior and metabolic dysfunction in C57Bl/6J mice**

**Background:** Balance of magnesium (Mg) ions regulates mood and may influence mood disorders. Magnesium is also linked with the glucose metabolism and the inflammatory response in the gut. **Methods:** We examined the behavioral effects in magnesium deficiency in mice through depression- and anxiety phenotyping experiments. We determined the effects after treatment with imipramine (20 mg·kg<sup>-1</sup>), diazepam (2 mg·kg<sup>-1</sup>) and ketamine (3 mg·kg<sup>-1</sup>). **Results:** We found that, compared to control mice (n=10), mice receiving magnesium deficient diet (n=10) (10 % RDA) for 6 weeks, were more immobile in the Forced swim test ( $p<0.01$ ), which suggested depression-like behavior. There was a significant antidepressant-like effect in the Forced swim test. Administration of imipramine and ketamine reduced the duration of immobility compared to the vehicle group (Ket vs. MgD  $p<0.001$ ; Imi vs. MgD  $p<0.0001$ ). They also displayed anxiety-like behavior in the Light/Dark box ( $p<0.01$ ). The intraperitoneal glucose tolerance test showed an elevation in glucose response after 30 minutes compared to the controls ( $p<0.05$ ). **Conclusion:** Insufficient dietary Mg may contribute to depressive disorders and its supplementation may have therapeutic benefits. **Future plans:** The inflammatory responses will be examined in the gut. We will look for different cytokines and determine the gut microbiota. We will look for protein expressed in the frontal cortex and hippocampus using real-time quantitative polymerase chain reaction (PCR).

**Sanne Enok: The role of gastric digestion on specific dynamic action and postprandial organ growth in pythons (*Python regius*)**

Digestion is associated with an increased oxygen consumption in animals (SDA), but the specific contributions to the metabolic rise remain unclear. The importance of gastric acid secretion and gut hypertrophy has been emphasized in previous studies. Thus, gastric digestion has been reported to account for up to 55% of the SDA response. To investigate the role of gastric processes, we blocked the pyloric sphincter in snakes, to exclude any intestinal contribution to digestive processes and let the snakes eat voluntarily. Animals subjected to a pyloric block showed a lower oxygen consumption compared to un-operated controls. In addition, animals given an amino acid solution (peptone) directly into the intestine (gastric bypass) showed SDA responses similar to animals fed the same solution into the stomach, thus experiencing a complete digestion. No organ growth was found in either the pyloric block or gastric bypass group. These results indicate a much lower gastric contribution to the SDA response than previously reported and that organ growth is not initiated in the stomach or as a response to protein.

## **Tine Østergaard Larsen: Arterial extracellular matrix changes in a murine model of type 1 diabetes**

Diabetes in humans accelerates cardiovascular disease caused by atherosclerosis. However the underlying pathogenesis of accelerated development of atherosclerosis in diabetics remains poorly understood. The arterial wall often becomes diffusely damaged (diabetic arteriopathy) which induces structural and epigenetic changes of the arterial wall. Namely alterations in the extracellular matrix (ECM) have been suggested to play a role in the progression of atherosclerosis. Therefore, we wish to characterize the arterial ECM proteins in type 1 diabetes and further, to test whether these changes will be reversible in a non-diabetic milieu. Aortas from streptozotocin-induced diabetic mice are retrieved and ECM proteins are extracted and analysed by liquid chromatography mass spectrometry (LC-MS/MS). Further, changes in the ultrastructure of the arterial wall are examined by electron microscopy and histological staining. Lastly, vessel segments will be transplanted from diabetic to non-diabetic mice to investigate whether glucose-lowering will reverse the changes in the arterial ECM.