Tuesday April 4, 2017
CHUV, César Roux Auditorium, BH 08
Rue du Bugnon 46, CH-1011 Lausanne

7th LIMNA Symposium

Organizers: Prof. J. Auwerx, Prof. Bart Deplancke, Prof. B. Desvergne, Prof. L. Fajas, Prof. F. Pralong, Prof. B. Thorens, Prof. K. Sakamoto, Prof. K. Schoonjans and Dr. L. Descamps.

Guest organizers: Isabelle Chareyron (NIHS), Caterina Collodet (NIHS), Mukul Girotra (Unil), Mary Gonzalez Melo (CHUV), Elena Katsyuba (EPFL), Darko Maric (Unil), Laia Martinez Carreres (Unil), Omid Mashinchian (NIHS), Erica Reggi (Unil), Tanja Sonntag (NIHS), Laura Steinbusch (Unil), Magda Zachara (EPFL).

Invited Speakers

Prof. Etienne Meylan, Swiss National Science Foundation Professor & Tenure-track Assistant Professor, School of Life Sciences, ISREC Institute, EPFL, Lausanne, Switzerland.

Prof. Olaia Naveiras, School of Life Sciences, Institute of Bioengineering, Laboratory of Regenerative Hematopoiesis, EPFL and Department of Medicine and Oncology (Hematology), Centre Hospitalier Universitaire Vaudois (CHUV), Lausanne, Switzerland.

PRIZES WINNERS:

Cédric Gobet, PhD, in the laboratory of Frédéric Gachon (NIHS) co-directed with Prof. Felix Naef (EPFL), received the prize awarded by the LIMNA direction committee for is talk entitled: *"Circadian and feeding rhythms differentially affect rhythmic mRNA translation in mouse liver".*
Ebru Aras, PhD, in the laboratory of Prof. Roberto Coppari (UNIGE) received the prize awarded by students and post-docs who attented the symposium for her talk: *"SIRT1 in SF1 Neurons Links Light Inputs to Circadian Function in Gastrocnemius".*
Tamara Marik, a PhD student in the laboratory of Prof. Elena Dubikovskaya (EPFL), reveived the poster prize awarded by the guest organizers for her poster: *"Development of novel optical imaging probe for quantification of glucose uptake in cells and living animals".*
Xavier Brenachot, a Post-doc in the laboratory of Prof. Roberto Coppari (UNIGE), received the prize awarded by the guest organizers for his poster: *"Protein Tyrosine Phosphatase Receptor Gamma is a target of NF-κB that negatively regulates insulin sensitivity".*

Agenda

8h30-9h00 Welcome and distribution of badges

Opening

9h00 **Kei Sakamoto**
Welcome

Morning session

Chairman: Tanja Sonntag

9h10 **Etienne Meylan**
"Glucose transporters in non-small cell lung cancer: regulation and function"

9h45 **Tung Chao**
PhD, Laboratory of Prof. Ping-Chih Ho, Department of Fundamental oncology, Immunometabolic Regulation Group, Unil
"Impaired mitochondrial homeostasis demolishes T cell anti-tumor responses"

10h05 **Maria Donaldson**
PhD, Laboratory of Prof. Orrichio, ISREC, EPFL
"Deciphering the impact of metabolic alterations in central nervous system primitive neural ectodermal tumor (CNS-PNET) development and progression"

10h25 Coffee Break

Chairman: Mary Gonzalez Melo

10h45 **Laura Steinbusch**
Post-doc, Laboratory of Prof. Thorens, CIG, Unil
"Brainstem glucokinase regulates glucagon section during hypoglycemia in male and female mice"

11h05 **Shehata Saifeldin**
PhD, Laboratory of Prof. Sakamoto, NIHS
"Physiological regulation of the CDK16/PCTAIRE-1 protein kinase and its proposed role in the brain"

11h25 **Cédric Gobet** **Best talk jury LIMNA**
PhD, Laboratory of F. Gachon (NIHS) co-directed with Prof. F. Naef (EPFL)
"Circadian and feeding rhythms differentially affect rhythmic mRNA translation in mouse liver"

11h45 **Ebru Aras** **Best talk students**
PhD, Laboratory of Prof. Coppari, Unige
"SIRT1 in SF1 Neurons Links Light Inputs to Circadian Function in Gastrocnemius"

12h05 Lunch

Afternoon session

12h50 Poster session

Chairman: Omid Mashinchian

14h30 **Olaia Naveiras**
"Bone Marrow Adiposity: an emerging tissue with metabolic and regulatory functions"

15h05 **Laura Velazquez – Villegas**
Post-Doc, Laboratory of Prof. Schoonjans, EFPL
"TGR5 signaling axis induces beiging of subcutaneous white adipose tissue"

15h25 **Pierre-Damien Denechaud**
Post-doc, Laboratory of Prof. Fajas, CIG, Unil
"E2F1 participates in liver cholesterol metabolism and protects against liver fibrosis development in response to high cholesterol diet"

15h45 Coffee break

Chairman: Laura Steinbsuch

16h05 **Alice Parisi**
Post-doc, Laboratory of Dr. P. Gut, NIHS
"Dynamics of mitochondrial biogenesis in zebrafish skeletal muscle"

16h25 **Pooja Jha**
Post-doc, Laboratory of Prof. Johan Auwerx, EPFL
"Identification of plasma lipid species as biomarkers of NAFLD using a systems genetics approach"

16h45 Concluding remarks and prizes distribution

ABSTRACTS - INVITED SPEAKERS

Etienne Meylan
Swiss National Science Foundation Professor & Tenure-track Assistant Professor, School of Life Sciences, ISREC Institute, EPFL, Lausanne, Switzerland.

"Glucose transporters in non-small cell lung cancer: regulation and function"

The rewiring of cellular energetics is an emerging hallmark of cancer. In particular, tumor cells have an increased demand for glucose, which is used for energy production, growth and proliferation. Our laboratory focuses on the modulation of glucose transporter activities in lung cancer. These transporters mediate the first and rate-limiting step for glucose usage, so deciphering their contribution to cancer development may lead to a better understanding of alterations in glucose metabolism, and to the identification of metabolic-based vulnerabilities for new treatment options. To investigate these crucial aspects of tumors, we use a combination of cell lines derived from human tumors, tumor tissue material, bioinformatics analyses and genetically engineered mouse models of lung cancer. During my talk, I will present our latest data and main research directions on the characterization of glucose transporters in this disease.

Olaia NAVEIRAS, M.D./Ph.D.
SNSF Professor
Laboratory of Regenerative Hematopoiesis
ISREC & Institute of Bioengineering
Ecole Polytechnique Fédérale de Lausanne (EPFL)

"Bone Marrow Adiposity: an emerging tissue with metabolic and regulatory functions"

The epidemic of diabetes and obesity has motivated very significant advances in the understanding of adipocyte regulation, the cellular complexity of the adipose tissue, and the plasticity of subcutaneous and retroperitoneal white and brown adipose depots.
Less attention has been paid to bone marrow adiposity (BMA), which in humans constitutes on average 1.3 kg of adipose tissue and 8% of the total fat mass, ranging from 1-30% depending on age and pathophysiological condition. Indeed, BMA increases with age, glucocorticoid treatment, obesity, cold exposure and upon moderate caloric restriction. Bone marrow adipocytes contribute to circulating adiponectin and are thought to play a role in skeletal muscle adaptation upon caloric restriction. Thus, BMA is now thought of as an endocrine organ on its own right.
Being the main production site for blood cells in mammals, the bone marrow harbors an intricate crosstalk between bone-forming cells, specialized sinusoidal vasculature, hematopoietic cells and bone marrow adipocytes. Although an increase in bone marrow adipocytes has been long associated with osteoporosis and constitutes the hallmark of hematopoietic failure syndromes, the causal relationship and molecular mechanisms for such negative association have been elusive.
In 2009 we determined a net negative effect of bone marrow adipocytes in hematopoiesis both in homeostasis (mostly reflecting the effect of constitutive BMA) and upon stress hematopoiesis following irradiation-mediated aplasia (mostly reflecting the effect of regulated BMA). Others have since validated the potent effect of PPARg inhibitors in accelerating hematopoietic recovery. Recent data indicating that preadipocytes support hematopoiesis while fully mature adipocytes inhibit rapid HSC proliferation will be discussed. To further dissect this phenomenon, we have also developed an *in vitro* system to model the red-to-yellow-to-red marrow transition, and an image-recognition tool that allows for unbiased quantification of BM adipocytes in vivo (MarrowQuant Plug-In).

SHORT TALKS ABSTRACTS

Last Name: Chao

First Name: Tung

Affiliation: Department of Fundamental Oncology, UNIL-LICR

Presentation: Abstract submission

Title of presentation: Impaired mitochondrial homeostasis demolishes T cell anti-tumor responses

Authors list: Tung Chao, Ping-Chih Ho

Abstract: Cytotoxic tumor-infiltrating CD8+ T cells (CD8+ TILs) account for the major arms of anti-tumor immunity. However, the immunosuppressive tumor microenvironment (TME) hampers infiltration and functions of CTLs through unclear regulations. Here we show that CD8+ TILs accumulate defective mitochondria, a phenotype which is associated with reduced effector functions. We then identified that activated CD8+ TILs with high PD-1 expression display more severe mitochondrial dysfunction. However, activated exhausted CD8+ T cells with high PD-1 expression under chronic viral infection did not show accumulation of dysfunctional mitochondria. Interestingly, we found that PD-1 signaling is able to promote mitochondrial activity in T cells but inhibition of mitophagy in conjunction with PD-1 signal promotes mitochondrial dysfunction. Therefore, our results suggest that the TME impede anti-tumor functions of TILs by suppressing mitophagy, a process for PD-1+ T cells to clean up damaged mitochondria.

Last Name: Donaldson

First Name: Maria

Affiliation: EPFL ISREC

Presentation: Abstract submission

Title of presentation: Deciphering the impact of metabolic alterations in central nervous system primitive neural ectodermal tumor (CNS-PNET) development and progression

Authors list: Maria Donaldson, Mor Mishkovsky, Natalya Katanyeva, Sadegh Saghafinia, Stephanie Sungalee, Hikari Yoshihara, Giovanni Ciriello, Elisa Oricchio

Abstract: Central nervous system (CNS) tumors are the leading cause of cancer-associated death in children. Primitive neural-ectodermal tumors (PNETs) are a particularly aggressive subtype of embryonal CNS-tumor, with a 5-year overall survival rate in less than 50% of patients. Despite sharing a similar cell of origin with medulloblastomas, CNS-PNETs have a different anatomical location, unique genetic features, and significantly worse clinical outcome (Jakacki et al., 2015). Lack of good models for CNS-PNETs challenges the opportunity to dissect the genetics of this tumor. Here we present a new CNS-PNET mouse model using neural precursors derived from human iPS cells, which allows us to perform genetic, biochemical and therapeutic studies in vivo. Through DNA methylation and RNA sequencing we find that the neural precursor (NPC)-derived model accurately recapitulates key features of primary CNS-PNETs. In addition, mRNA expression profiling of primary CNS-PNET samples indicate that the deregulation of genes involved in glucose and nucleotide metabolism directly impact CNS-PNET pathogenesis. Using our NPC-derived CNS-PNET model and a patient-derived cell line, PFSK-1, we conducted in vivo metabolic analyses of these tumors and identified unique metabolic features of CNS-PNETs. We aim to interrogate these metabolic lesions in CNS-PNETs with the ultimate goal of developing new therapeutic and diagnostic approaches.

Last Name: Steinbusch

First Name: Laura

Affiliation: CIG Unil

Presentation: Abstract submission

Title of presentation: Brainstem glucokinase regulates glucagon section during hypoglycemia in male and female mice

Authors list: Laura KM Steinbusch, Davide Basco, Alexandre Picard and Bernard Thorens* Center for Integrative Genomics, University of Lausanne, Lausanne, Switzerland.

Abstract: Specific brain regions contain glucosensing neurons, most importantly the hypothalamus and the brain stem. Specific nuclei in the brain stem, the nucleus tractus solitarius (NTS) and the dorsal vagal nucleus (DMNX), are implicated in the regulation of glucose homeostasis. Both these regions express glucokinase (Gck) which is known for being the rate-limiting enzyme in pancreatic beta-cell glucose-stimulated insulin secretion. Here, we investigated the role of Gck in the NTS and DMNX in glucose homeostasis. We studied mice with genetic inactivation of the Gck gene in Phox2b neurons of the brain stem (Phox2bGck-/- mice). As compared to control littermates, both male and female Phox2bGck-/-mice displayed a reduction in glucopenia-induced vagus nerve activation and hypoglycemia-induced glucagon secretion. Strikingly, Phox2bGck-/- and control mice were similar with respect to body weight, body composition, glucose tolerance and insulin secretion. Thus, Gck in Phox2b neurons of the brain stem plays a role in the glucose-dependent control of vagus nerve activity and hypoglycemia-induced glucagon secretion in male and female mice. Altogether, our current study and a recently published study imply that both hypothalamic glucokinase as well as brainstem glucokinase are implicated in hypoglycemia-induced glucagon secretion.

Last Name: Shehata

First Name: Saifeldin

Affiliation: NIHS/EPFL

Presentation: Abstract submission

Title of presentation: Physiological regulation of the CDK16/PCTAIRE-1 protein kinase and its proposed role in the brain

Authors list: Saifeldin N Shehata, Maria Deak, Roger W Hunter, Nicholas A Morrice, Vera M Kalscheuer, Kei Sakamoto

Abstract: Cell signalling, mediated to a large extent by protein kinase phosphorylation, plays a vital role in regulation of cellular function. PCTAIRE-1/CDK16, is a Ser/Thr kinase that has been implicated in many cellular processes, including spermatogenesis, neurite outgrowth, vesicle trafficking, and most recently, X-linked intellectual disability (XLID). To understand the molecular regulation of PCTAIRE-1, we previously showed that PCTAIRE-1 preferentially phosphorylated peptide motifs that differed from the classical CDK family preference. We also showed that cyclin Y robustly binds and activates PCTAIRE-1 > 100-fold. Moreover, we have identified two phosphorylation sites on cyclin Y that are essential for binding the well-known adaptor protein 14-3-3, which we propose stabilizes cyclin Y in a favourable PCTAIRE-1-binding conformation. Mutation of these sites abolished PCTAIRE-1 binding and activation. Furthermore, we have cloned human PCTAIRE-1 mutants identified in XLID patients, and confirmed their failure to bind the cyclin Y-14-3-3 activating complex. To understand the physiological relevance of PCTAIRE-1 activity, we have utilised a chemical genetics approach that exploits the ability of an engineered PCTAIRE-1 mutant to selectively modify its substrates in complex lysate. We have identified three putative PCTAIRE-1 substrates (AAK1, dynamin 1 and synaptojanin 1) in mouse brain that regulate crucial steps of receptor endocytosis, a fundamental process in neuronal synaptic transmission, and have further validated AAK1 as a genuine PCTAIRE-1 substrate in mouse brain. Collectively, this work has provided key molecular regulatory mechanisms of PCTAIRE-1, which has laid the foundations for future studies of its role in specific cellular functions and physiological pathways.

Last Name: Gobet

First Name: Cédric

Affiliation: NIHS/EPFL

Presentation: Abstract submission

Title of presentation: Circadian and feeding rhythms differentially affect rhythmic mRNA translation in mouse liver

Authors list: Cédric Gobet, Florian Atger, Julien Marquis, Eva Martin, Benjamin Weger, Grégory Lefebvre, Patrick Descombes, Felix Naef, and Frédéric Gachon

Abstract: Rhythmic gene regulation in mouse liver originates from an intricate interplay between the circadian clock and feeding rhythm. While such interactions have been extensively studied, a complete picture of the diurnal transcription-translation process is still lacking. Therefore, we performed RNA-Seq and Ribosome profiling in mouse liver under physiological light–dark conditions. Notably, we quantified temporal mRNAs transcription, accumulation, and translation in wildtype and Bmal1-deficient animals in ad libitum or night-restricted feeding. We found that rhythmic transcription is the main driver underlying rhythmic mRNA accumulation and translation for a majority of genes. On the other hand, translation efficiency is rhythmically regulated for genes with 5′-Terminal Oligo Pyrimidine tract (5′-TOP) sequences or a Translation Initiator of Short 5′-UTR (TISU) motif. Remarkably, the diurnal translational regulation is mainly driven by feeding rhythms although Bmal1 deletion slightly affects amplitude and phase for some genes. Secondly, we developed computational methods to infer ribosomes decoding rates of the different codons from the ribosome profiling datasets. We found that ribosome residence times span a threefold range from the fastest to slowest codon and exhibit positive and negative interactions between the different sites of the ribosome. Finally, we performed high-throughput tRNA profiling in mouse liver to decipher the regulation of translation elongation and explain the variation in the inferred ribosome residence times.

Last Name: Aras

First Name: Ebru

Affiliation: UNIGE-CMU-Phyme Department

Presentation: Abstract submission

Title of presentation: SIRT1 in SF1 Neurons Links Light Inputs to Circadian Function in Gastrocnemius

Authors list: Ebru Aras, Giorgio Ramadori, Roberto Coppari

Abstract: Daily light/dark cycles influence brain and peripheral tissues' function to align the organism with circadian environmental changes. This alignment is physiologically relevant as its disturbance increases the risk for several illnesses including insulin resistance, obesity, diabetes and cancer. At the central level, the hypothalamic suprachiasmatic nucleus (SCN) is a well-established component as it receives optical signals from the retina and is required for synchronization of circadian gene expression in peripheral tissues. Although the identity of the neurons lying downstream of the SCN is largely unknown, recent findings suggest that steroidogenic factor (SF) 1-expressing neurons within the hypothalamic ventromedial nucleus (VMH) are important components, as SF1 neurons have been reported to be involved in coupling light inputs to normal cyclic brown adipose tissue (BAT)'s activity and whole-body energy expenditure. We recently reported that mice lacking SIRT1 in SF1 neurons are more prone to develop diabetes due to impaired insulin action in gastrocnemius skeletal muscle. Our recent data confirmed that the impaired insulin action is rescued by light removal. Also, our data indicate that mice lacking SIRT1 in SF1 neurons have altered circadian gene expression selectively in gastrocnemius; a defect that is also rescued by light removal. Overall, these findings led us to hypothesize that SIRT1 in SF1 neurons links light inputs to circadian function in gastrocnemius skeletal muscle tissue.

Last Name: Velazquez Villegas

First Name: Laura Alejandra

Affiliation: UPSCHOONJANS

Presentation: Abstract submission

Title of presentation: TGR5 SIGNALING AXIS INDUCES BEIGING OF SUBCUTANEOUS WHITE ADIPOSE TISSUE

Authors list: Laura Velazquez-Villegas1*, Alessia Perino1*, Norman Moullan1, Andréane Fouassier1, Kristina Schoonjans1 1 Metabolic Signaling, Ecole Polytechnique Fédérale de Lausanne, Lausanne, Switzerland * Equal contribution

Abstract: Obesity represents one of the major worldwide health problems and is associated with a complex array of metabolic perturbations. Bile acids (BAs) are signaling molecules that control a plethora of metabolic processes, including insulin resistance and obesity. One of the main mediators of BA signaling is the G protein-coupled receptor TGR5. TGR5 is expressed in a wide variety of metabolic tissues, including the adipose tissue. Activation of BA-TGR5 signaling in obese mice increases energy expenditure and reduces fat mass by activating brown adipose tissue (BAT). Whether these effects are only mediated through the activation of BAT is unknown. Recently, a new fat cell type called “beige” with characteristics similar to BAT has been described. In this study, we show that chronic treatment with specific TGR5 agonists (the bile acid analog INT-777) of diet-induced obese Tgr5 wild-type (Tgr5+/+) mice enhances energy expenditure and promotes the up-regulation of various beige adipocyte markers in the subcutaneous WAT (subWAT). In contrast to the Tgr5+/+ mice, no browning effect could be observed in the TGR5 knockout mice (Tgr5-/-). Prolonged cold exposure also induced browning of subWAT in Tgr5+/+, but again not in Tgr5-/- and adipose tissue-specific Tgr5 Adipoq-/- mice. The browning effect could be recapitulated in pre-adipocytes derived from the subWAT stromal vascular fraction upon differentiation and exposure to INT-777 or BAs. In addition, administration of TGR5 agonists to Tgr5+/+ differentiated cells resulted in improved mitochondrial function. Taken together, these data identify BA-TGR5 as a novel pathway to promote beige cell formation and activity. A better understanding of these mechanisms may lead to novel therapeutic approaches to prevent obesity-related diseases.

Last Name: Denechaud

First Name: Pierre-Damien

Affiliation: CIG UNIL

Presentation: Abstract submission

Title of presentation: E2F1 participates in liver cholesterol metabolism and protects against liver fibrosis development in response to high cholesterol diet

Authors list: Pierre-Damien Denechaud, Qiuwen Lai, Albert Giralt, Cédric Le May, Lianjun Zhang, Bertrand Cariou and Lluis Fajas

Abstract: Cholesterol accumulation in the liver is an early event in nonalcoholic fatty liver disease (NAFLD). We found that E2F1, known for its implication in cell cycle regulation, plays a crucial role in maintaining cellular cholesterol homeostasis. This transcription factor regulates cholesterol uptake via PCSK9 (proprotein convertase subtilisin/kexin type 9), an enzyme that promotes LDLR (low density lipoprotein receptor) degradation upon activation. E2F1-/- mice display reduced total plasma cholesterol levels and increased cholesterol content in the liver. In this study, we show that E2F1 deletion in cellular and mouse models leads to a marked decrease in PCSK9 expression and increase in LDLR expression. In addition to the up regulation of LDLR, we report that E2F1-/- hepatocytes exhibit increased LDL uptake. ChIP-Seq and PCSK9 promoter reporter experiments confirmed that E2F1 binds to and transactivates the PCSK9 promoter. Interestingly, E2F1-/- mice fed a high cholesterol diet (HCD) display a fatty liver phenotype and early liver fibrosis, which is reversed by re-expression of PCSK9 in the liver. Collectively, these data indicate that E2F1 regulates cholesterol uptake and that the loss of E2F1 leads to abnormal cholesterol accumulation in the liver and the development of fibrosis in response to a high cholesterol diet.

Last Name: Parisi

First Name: Alice

Affiliation: NIHS

Presentation: Abstract submission

Title of presentation: Dynamics of mitochondrial biogenesis in zebrafish skeletal muscle

Authors list: Alice Parisi, Laura Strohm, Joy Richard, Eugenia Migliavacca, Jerome Feige, Bruce Spiegelman, Philipp Gut

Abstract: Mitochondria biogenesis in response to exercise is a critical event in increasing skeletal muscle endurance. To identify novel signals that govern mitochondrial plasticity, we probe mitochondrial biogenesis in zebrafish through a combination of genetic and physiological perturbations. We developed an aerobic exercise protocol sufficient to potently induce the generation of new mitochondria. Using swim-performance spirometry, histology and gene profiling, we compared exercise adaptations of untrained and trained wildtype zebrafish with that of transgenic zebrafish overexpressing PGC1A (PPARGC1A) the key regulator of mammalian mitochondrial biogenesis. Strikingly, untrained Tg(actc1b:PPARGC1A) zebrafish present a complete fiber type switch to slow-twitch, consume more oxygen and have a similarly elevated exercise endurance as trained wildtype zebrafish. GO analysis shows a broad overlap of transcriptional programs of trained wt and untrained Tg(actc1b: PPARGC1A) zebrafish as well as a high evolutionary conservation of exercise-related genes. In addition, we profiled the generation of new mitochondria at early stages. We find that exogenous human and zebrafish ppargc1a efficiently induce transcriptional activation of mitochondrial biogenesis at 6 dpf, but not at 4 dpf. We furthermore developed customized gene expression arrays of exercise-induced genes, and found that at larval stages ppargc1a mainly induces components of the respiratory chain whereas the reprogramming of genes related to fiber metabolism occurs later during early adulthood. In summary, our findings provide unique insights into the timing of mitochondrial biogenesis in larval zebrafish, and pave the way to use adult zebrafish for studying plasticity of mitochondria in response to exercise.

Last Name: Jha

First Name: Pooja

Affiliation: NCEM, EPFL

Presentation: Abstract submission

Title of presentation: Identification of plasma lipid species as biomarkers of NAFLD using a systems genetics approach

Authors list: Pooja Jha, Pedro M. Quiros, Molly McDevitt, Dave Pagliarini, Johan Auwerx

Abstract: As genome-wide association studies (GWAS) may be reaching their limit for discovering genetic determinants of non-alcoholic fatty liver disease (NAFLD), alternative genetic strategies, such as the mouse genetic reference populations (GRPs) may provide both novel and complementary information to identify clinically and biologically important biomarkers of NAFLD. Since accumulation of specific lipids in the liver is the pathophysiologic hallmark of many liver diseases including NAFLD, we aimed to find the lipid biomarkers of NAFLD in serum using the BXD mouse GRP. We performed targeted lipidomics analysis from serum and liver of chow diet (CD) and high fat diet (HFD) fed cohorts of 50 BXD strains. Over 120 lipids from different lipid classes were measured. Using genetic and bioinformatic approaches we identified 4 triglyceride (TG) species as biomarkers of NAFLD, which we further validated in human cohorts with NAFLD. Additionally, we identified multiple novel lipid QTLs (mQTLs) for ~70% of the lipids measured and validated a handful of the genetic loci for triglycerides (FADS1-3, LRP1, LIPC, IRS1, FRMD5, AGPAT1, GCKR, TOMM40, APOE, PLD2 etc.) identified in GWAS to their specific TG species. The biomarkers identified from our BXD GRP holds the potential to be used in clinical diagnosis of NAFLD. Additionally, the lipidomics profiling in the BXDs, along with the previously curated additional omics datasets provides a robust resource to the scientific community for in-silico data analysis.

POSTERS ABSTRACTS

Poster 1

Last Name: Hafner

First Name: Jasmin

Affiliation: Laboratory of Computational Systems Biotechnology, EPFL

Presentation: Abstract submission

Title of presentation: Exploring chemodiversity in metabolism towards the integration of chemistry into biology

Authors list: Jasmin Hafner, Noushin Hadadi, Vassily Hatzimanikatis

Abstract: The availability of different levels of omics data help us to observe cells with higher resolution and from different perspectives. Consequently, the computational exploration of metabolism advanced in the last decade to make sense of newly available data from genomics, transcriptomics and metabolomics. However, our understanding of metabolism lags behind in explaining the chemodiversity observed in living organisms. Integrating experimentally measured metabolites into existing metabolic knowledge is a challenge we aim to address by extrapolating the known biochemistry towards selective integration of chemical compounds and their associated reactions. The “ATLAS of Biochemistry” is an ongoing effort to explore and expand our knowledge of metabolism. We apply the computational tool BNICE.ch to generate known and novel reactions and compounds using expert curated, generalized enzyme reaction rules. The first released version of ATLAS (http://lcsb-databases.epfl.ch/atlas) contains all possible reactions (known and hypothetical) between known biological compounds. We could also demonstrate that the selective integration of chemicals into metabolic networks is the key to complete the mechanism of poorly characterized reactions and to integrate orphan metabolites into metabolic networks. Starting with 16’000 biological compounds, we found biochemical reactions towards 60’000 unique PubChem compounds one reaction step away from known metabolism, and 120’000 PubChem compounds two steps away. The database of these compounds and the associated reactions can be used to create hypotheses about the origin of experimentally measured compounds and serve as a tool for metabolic engineers, synthetic biologists and other scientists working with metabolomics and secondary metabolism.

Poster 2

Last Name: Nasrallah

First Name: Anita

Affiliation: CIG - UNIL

Presentation: Abstract submission

Title of presentation: The Role of Novel Kinases in Adipose Tissue Biology

Authors list: Anita Nasrallah*, Isabel-Cristina Lopez Mejia*, Albert Giralt Coll, Alessia Spirli, Miriam Ejarque, Sonia Fernandez Veledo, Olivier Staub, Joan Vendrell Ortega, Francisco Tinahones Madueño, Lluis Fajas Coll

Abstract: It is well established that pro-inflamatory cytokines, such as IL-6 and TNF-α, affect insulin signalling, which in turn is essential to maintain glucose homeostasis and to regulate its metabolism in the liver, muscle, and adipose tissues. This leads to the stimulation of downstream protein kinases, thus activating and crosslinking numerous pathways, potentially resulting in insulin resistance. Consequently, insulin resistance status is determined by the type of activated inflammatory pathways, abnormalities of lipid metabolism, as well as in the type of activated kinases and their downstream targets. This project revolves around the role of novel kinases in SAT and VAT of patients that are insulin resistant (IR) or insulin sensitive (IS). Several of the known protein kinases involved in the onset of insulin resistant are AMP-activated protein kinase (AMPK), IκB kinase (IKK), protein kinase C (PKC), mitogen-activated protein kinases (MAPKs), etc. Identifying new and specific protein kinases involved in obesity-induced chronic inflammation may help in developing the targeted drug therapies to minimize insulin resistance in patients.

Poster 3 Poster Prize

Last Name: Brenachot

First Name: Xavier

Affiliation: University of Geneva/CMU/PHYME

Presentation: Abstract submission

Title of presentation: Protein Tyrosine Phosphatase Receptor Gamma is a target of NF-κB that negatively regulates insulin sensitivity

Authors list: Xavier Brenachot, Giorgio Ramadori and Roberto Coppari

Abstract: Insulin resistance underlies type 2 diabetes mellitus (T2DM). Yet, the molecular mechanisms underpinning insulin resistance are poorly understood. Here we show that hepatic expression of Protein Tyrosine Phosphatase Receptor Gamma (PTPR-γ) is induced by inflammation in obese and T2DM mice and positively correlates with indices of inflammation and insulin resistance in humans. PTPR-γ loss-of-function lowers glycemia and insulinemia and protects from development of T2DM by enhancing insulin-stimulated suppression of endogenous glucose production. Conversely, hepatic-specific overexpression of PTPR-γ is sufficient to cause insulin resistance in mice. These loss- or grain-of-function effects are concomitant with enhanced or decreased phosphorylation of the insulin receptor and several members of its signaling cascade, respectively. Furthermore, our results demonstrate that Nuclear Factor κB (NF-κB) binds to PTPR-γ promoter and is required for inflammation-induced PTPR-γ expression. These data establish that PTPR-γ negatively regulates insulin sensitivity and identify a new target for treatment of T2DM.

Poster 4

Last Name: CONTAT

First Name: Caroline

Affiliation: EPFL/ISREC/SV

Presentation: Abstract submission

Title of presentation: Role of glucose transporter Glut1 in non-small cell lung cancer

Authors list: Caroline Contat, Pierre-Benoit Ancey, Nadine Zangger, Nadine Stokar, Etienne Meylan

Abstract: Lung cancer is the leading cause of cancer-related deaths worldwide, with non-small cell lung cancer (NSCLC) representing 85% of the cases. Tumor cells have a high demand for glucose for growth and proliferation, the uptake of which is carried by specific members of the glucose transporter (Glut) family. Among them, Glut1 and Glut3, which have high affinity for glucose, show increased expression in various malignancies. However, their contribution to lung tumorigenesis is unknown. Here, we used autochthonous mouse models of NSCLC where tumors are initiated in adults upon Cre-mediated oncogenic Kras activation (K-RasLSL-G12D/wt), in combination (or not) with concomitant loss of one of p53 or Lkb1 tumor suppressors, to interrogate the role of Glut1 in lung tumor development. To do that, these mice were crossed to Glut1 conditional knockout mice, to enable tumor development in the presence or absence of this glucose transporter. Our current and future investigations are aimed to comprehend the regulation of Glut1 expression in normal and cancerous lung tissue, and its impact on tumor epithelial cells as well as the microenvironment of lung tumors.

Poster 5

Last Name: Moyat

First Name: Mati

Affiliation: EPFL-GHI

Presentation: Abstract submission

Title of presentation: Metabolic changes associated with Helminths infection during diet-induced obesity

Authors list: Mati Moyat, Kathleen Shah, Audrey Chuat, Nicola Harris

Abstract: Throughout evolution both intestinal helminths and commensal bacteria have inhabited our intestines. This "ménage à trois" situation is likely to have exerted a strong selective pressure on the development of our metabolic and immune systems. Although intestinal helminths are generally accepted to possess potent immuno-modulatory activity and to modulate metabolic status, it is unknown whether these abilities require interactions with the intestinal microbiota. Recent work from our laboratory investigating allergic asthma determined that helminths can modulate host disease by altering microbial metabolism. The current study extends this work to investigate the impact of helminth-bacterial interactions in regulating host metabolism. Our first goal is to determine if chronic infection with the murine helminth Heligmosomoides polygyrus bakeri (Hpb) provides protection against diet-induced obesity (DIO). The second objective is to assess the effect of helminth-altered microbiota on DIO. Firstly, we looked for marker associated with metabolic improvement such as visceral adipose tissue (VAT) eosinophilia or brite adipocyte. Although, under normal condition Hpb infection was associated with VAT eosinophilia, the first results of DIO showed with different experimental settings that Hpb infection was not protective. However, the first fecal transfer experiment was much more promising. In this experiment, antibiotic treated mice recolonized with helminth-altered microbiota gained less weight under high fat diet compared to mice recolonized with naïve microbiota. Our plan is now to focus on the effects of helminth-altered microbiota on intestine and adipose tissue physiology to understand its protective mechanisms.

Poster 6

Last Name: Pimentel

First Name: Gregory

Affiliation: CHUV - Endocrinology Diabetology and metabolism

Presentation: Abstract submission

Title of presentation: Metabolic footprinting of fermented milk consumption in serum of healthy men

Authors list: Grégory Pimentel, Kathryn J Burton, Ueli von Ah, Ueli Bütikofer, François Pralong, Nathalie Vionnet, Reto Portmann, Guy Vergères

Abstract: Fermentation is a widely used method for natural food preservation and extension of the nutritional value. Therefore, fermented dairy products are more and more in the focus of studies for their ability to exert health benefits beyond their nutritional qualities. With the aim of providing new elements to the discussion, we have evaluated the metabolic footprint of fermentation in milk products, as well as changes in serum metabolome after the ingestion of a fermented dairy product. In a randomized crossover study, 14 healthy men consumed a non-fermented milk and a probiotic yogurt. Using untargeted metabolomics, we assessed the modulation of the serum metabolome postprandially (over six hours), as well as fasting after two weeks daily intake. Multivariate statistical analysis revealed that fermentation produced a detectable 'footprint' on the metabolomes of milk products as well as on the postprandial serum. In particular, yogurt intake was characterized by higher levels of seven free amino acids, reduced levels of five bile acids and modulation of four indole derivative compounds. Based on the postprandial results, fasting sera after semichronic intake of milk or yogurt could also be differentiated. This study follows the effects of milk fermentation from product changes to consequences on the serum metabolic profiles. Several metabolic pathways appeared to be modulated by yogurt and could be further investigated to explore potential novel health qualities of fermented dairy products.

Poster 7

Last Name: Groeneveld

First Name: Svenja

Affiliation: EPFL-ISREC-UPMEYLAN

Presentation: Abstract submission

Title of presentation: A mouse model to study metabolic alterations in non-small cell lung cancer

Authors list: Svenja Groeneveld Bernard Moret Simona Rossi Alessandra Piersigilli Paolo Angelino Nadine Zangger Etienne Meylan

Abstract: During epithelial-to-mesenchymal transition (EMT), the metabolism of a cancer cell changes, likely to meet the environmental challenges faced during the metastatic process. We recently reported that the glucose transporter GLUT3 is upregulated during EMT in non-small cell lung cancer (NSCLC). Furthermore, we found that Glutamine-Fructose-6-Phosphate Transaminase 2 (GFPT2), the rate-limiting enzyme of the hexosamine biosynthesis pathway (HBP), which produces a substrate for protein modifications, is correlated with GLUT3 and upregulated during EMT as well. To investigate the metabolic alterations accompanying the EMT process in vivo, we used a genetically engineered K-ras(LSL-G12D/+);p53(fl/fl) lung tumor model and overexpressed or silenced the EMT-inducing transcription factor Snail in the lung tumor cells. Comprehensive characterization of the resulting tumors, including histological and gene expression analysis, revealed that while Snail overexpression enhanced malignant progression, it was not sufficient to result in an overt EMT phenotype. In line with this, no changes in GLUT3 or GFPT2 expression occurred upon Snail overexpression. Interestingly, gene expression and flow cytometry analyses demonstrated a strong alteration of the intratumoral immune compartment in response to Snail overexpression. Furthermore, by integrating both Snail overexpression and silencing approaches we found that Snail repressed the imprinted Dlk1-Dio3 locus, which contains one of the genome's largest cluster of miRNAs. We conclude that in our mouse model of NSCLC, chronic Snail expression is not sufficient to trigger an EMT. However, Snail exerted several functions beyond its classical role as EMT inducer, which include a previously undescribed role in regulating the Dlk1-Dio3 locus.

Poster 8

Last Name: Masid Barcon

First Name: Maria

Affiliation: LCSB EPFL

Presentation: Abstract submission

Title of presentation: Understanding reprogramming of cancer cell metabolism using metabolic modeling

Authors list: Maria Masid Barcon Joana Pinto Vieira Meriç Ataman Vassily Hatzimanikatis

Abstract: Cancer cells have been observed to undergo a metabolic reprogramming where glycolytic fluxes are up-regulated whereas oxidative phosphorylation is down-regulated. This metabolic phenotype is known as the Warburg effect, and it is observed even under high oxygen conditions. Experimental disruption of the expression of glycolytic enzymes has shown to stimulate oxidative phosphorylation and to lead to a decrease tumor cell proliferation. These observations have generated interest in the metabolic modeling of tumor cell metabolism with the purpose of identifying possible targets for new cancer therapies. However, despite the many studies performed, the mechanistic details of the metabolic reprogramming of cancer cells are still not completely understood. In particular, the physiological conditions in the origin of the Warburg metabolic switch have not been properly characterized. Metabolic modeling has proven to be a valuable tool for the investigation of cell physiology. Therefore, based on a generic human genome-scale reconstruction, we generated a reduced human metabolic model to focus on a set of subsystems from the central carbon metabolism that is of interest for the physiology under study. We explore the mechanism behind this metabolic reprogramming integrating fluxomics and metabolomics data into the reduced model to simulate and analyze the metabolic phenotypes of cancer cells and their healthy counterparts.

Poster 9

Last Name: CASTILLO ARMENGOL

First Name: JUDIT

Affiliation: CIG-UNIl

Presentation: Abstract submission

Title of presentation: CDK4, a new player in brown adipose tissue biology and adipose stem cell fate

Authors list: Judit Castillo Armengol, Isabel Lopez Mejia, Honglei Ji, Sylviane Lagarrigue, Lluis Fajas Coll

Abstract: White adipose tissue is known for its role in fat storage and whole body lipid/energy homeostasis. On the other hand, brown adipose tissue generates heat through the activity of uncoupling protein 1 (UCP1). More recently, attention has been placed into a third category of specialized heat-producing adipocytes, that can not only store lipids, but also prevent the onset of the metabolic phenoytpe. These adipocytes have been named brite (brown-in-white) adipocytes. The fact that the activities of brown and brite fat cells can limit metabolic diseases in mice, and correlate with leanness in humans underlines the importance of research in this recent field. Numerous genes and pathways that regulate brown and beige adipocyte biology have now been identified, however the role of cell cycle regulators in the development and the function of these oxidative adipose depots has not been thoroughly studied yet. We now aim to determine the role of the CDK4 in the function and differentiation of brown and brite adipocytes. Preliminary data suggests that CDK4 activity is inversely correlated with oxidative function in brown adipose tissue (BAT) and with browning in subcutaneous adipose tissue (scWAT). Indeed both morphological and gene expression data show that mice lacking CDK4 exhibit decreased lipid content and increased brown and oxidative gene expression in BAT. Under chow diet, these animals also display browning and increased oxidative gene expression in scWAT. Our preliminary data strengthen the need to study the molecular mechanisms by which CDK4 controls energy metabolism in brown and brite adipose cells. A better understanding of those processes might open up new therapeutic perspectives in the control of metabolic diseases such as diabetes or obesity.

Poster 10

Last Name: Weger

First Name: Benjamin

Affiliation: Nestlé Institute of Health Sciences SA - Diabetes & Circadian Rhythms

Presentation: Abstract submission

Title of presentation: Gut microbiota rewire diurnal sexually dimorphic metabolism

Authors list: Benjamin Weger, Cédric Gobet, Jake Yeung, Eva Martin, Aline Charpagne, Chieh-Jason Chou, Félix Naef, Frédéric Gachon

Abstract: Gut microbiota and the circadian clock are both key regulators of metabolic processes. Although recent evidence points to the impact of the circadian clock on microbiota, gut microbiota effect on diurnal host gene expression remains elusive. A transcriptome analysis of germ-free mice reveals subtle changes in circadian clock gene expression in liver, intestinal, and white adipose tissue. However, a lack of microbiome leads to the loss of liver sex-dimorphism and alters the rhythmic expression of sex-specific genes involved in lipid and amino acid metabolism, as well as xenobiotic detoxification. Consequently, rhythmic sex-dimorphic metabolism is also strongly altered in germ-free mice. These results emphasize the mutual interaction of gut microbiota and its host even on unexpected functions.

Poster 11

Last Name: Pandey

First Name: Vikash

Affiliation: LCSB, EPFL

Presentation: Abstract submission

Title of presentation: Context-Specific Cell Signaling Analysis using Logic Framework

Authors list: Vikash Pandey and Vassily Hatzimanikatis

Abstract: The description of signaling networks in textbooks and online resources is usually not context specific, because it is typically based on evidence from multiple experiments, performed for different cell types growing under various conditions. Thus, any analysis of such networks alone will lack all context-specific information that originates from context dependent signaling. Here, we developed a logic-based novel method to perform context-based analysis using signaling networks and context-specific transcriptomics and proteomics data. To understand how NOTCH1 interactions differ between different cancer types, we first integrated the cancer cell line encyclopedia (CCLE) data of 24 cancer types into NOTCH1 signaling network. Subsequently, we identified CNNT1 and HES1 as expressed only in blood, prostate, and salivary glands cancers. Furthermore, MYC was found in almost each cancer types. This result is in agreement with literature where MYC it known to be a cell proliferation regulator, and results in the development of cancer.

Poster 12

Last Name: Basco

First Name: Davide

Affiliation: Center for Integrative Genomics, UniL

Presentation: Abstract submission

Title of presentation: Alpha-cell glucokinase is required for the glucose-dependent suppression of glucagon secretion

Authors list: Davide Basco, Quan Zhang, Albert Salehi, Andrei Tarasov, Wanda Dolci, Xavier Berney, Pedro Herrera, David Tarussio, Patrik Rorsman, Bernard Thorens.

Abstract: Glucokinase (Gck) catalyzes the phosphorylation of glucose (G) into G-6-phosphate. Its role in pancreatic α-cells in the process of hypoglycemia-stimulated glucagon secretion is not yet established. Mice with selective inactivation of Gck in α-cells (αGckKO) were generated by crossing Gckflox mice and preproglucagon-Cre mice. Hormone secretion at different [G], ATP production, and membrane potential were measured on isolated islets. Whole body glucose homeostasis was assessed in Control (ctrl) and αGckKO mice of different ages. Glucagon secretion by αGckKO and ctrl islets was identical at 1mM G. However, at 6 and 20mM G, whereas glucagon secretion was suppressed in ctrl islets it remained uninhibited in αGckKO islets. Tolbutamide, a KATP-dependent channel blocker which mimics high [G], reduced glucagon secretion in islets of both genotypes. Thus, Gck inactivation affects glucose-dependent glucagon secretion upstream of the KATP channel by preventing ATP production at high glucose concentrations, as confirmed by [ATP]int measurements. In agreement with these observations, patch clamp analysis showed that the electrical activity of αGckKO α-cells was not suppressed by high [G], in contrast to the reduction observed in ctrl α-cells. Analysis of fed αGckKO mice showed higher plasma glucagon level, increased liver expression of P-CREB, Pepck and G6Pase as well as impaired pyruvate tolerance. 36 week-old αGckKO mice then developed a pre-diabetes phenotype with hyperglucagonemia, glucose intolerance, and higher insulin secretion. Collectively, these data show that Gck in α-cells is required for ATP production and suppression of glucagon secretion at high [G]. Absence of this control leads to hyperglucagonemia, increased hepatic glucose output, and development of pre-diabetes.

Poster 13

Last Name: shah

First Name: kathleen

Affiliation: Global health institute (GHI) - EPFL

Presentation: Abstract submission

Title of presentation: Beyond Immunity: Exploring a role for eosinophils in the maintenance of intestinal homeostasis and function

Authors list: Kathleen Shah*, Jeremiah Bernier-Latmani*, Tiffany Bouchery, Mati Moyat, Tatiana Petrova, Nicola L. Harris *Authors with equal contribution

Abstract: Eosinophils are multi-functional granulocytes, commonly implicated in type II immune responses. Recent studies have shown these cells to be protective against diet-induced obesity and subsequent development of metabolic syndrome. Under homeostatic conditions, eosinophils represent 10-20% of total lymphocytes in the small intestinal lamina propria, and have been shown to promote intestinal homeostasis through its maintenance of immune cell populations (e.g. IgA+ B-cells, ILC3, CD4 T-cells) in this tissue. However, whether they regulate other intestinal functions such as nutrient uptake and barrier integrity has yet to be explored. Using a model of eosinophil-deficiency (i.e. dbl-GATA1 mice), we show that eosinophils are important for regulating villous architecture/size, in a manner dependent on its interactions with the stroma. In addition, we show that this interaction can have direct consequences on intestinal fat uptake. Future studies will focus on determining whether intestinal eosinophils protect against diet-induced obesity through their maintenance of structures within the villi (i.e. lymphatics, myofibroblasts) and barrier integrity – which is thought to preceed the development of metabolic abnormalities in obese individuals.

Poster 14

Last Name: Maric

First Name: Tamara

Affiliation: EPFL

Presentation: Abstract submission

Title of presentation: Development of novel optical imaging probe for quantification of glucose uptake in cells and living animals

Authors list: Tamara Maric1, Georgy Mikhaylov1, Pavel Khodakovskiy1, Aleksandra Konovalova1, Kei Sakamoto2 and Elena Dubikovskaya1

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Abstract: Glucose is one of the major sources of energy for cells and its uptake and metabolism are often altered in cancer cells. Therefore, quantification of glucose uptake in tumors plays important role for understanding of cancer biology as well as development of new diagnostics and therapies. Many different methods have been used to specifically image glucose uptake such as positron emission tomography, magnetic resonance imaging and fluorescence, however all have limitations. Here we have developed a novel reagent for noninvasive imaging and quantification of glucose uptake in cells and living animals based on bioluminescent readout. This approach differs from other currently in use molecular imaging methods by favorable properties of bioluminescence for in vivo studies that shows good tissue penetration, low background and biological compatibility. We demonstrate that our novel imaging reagent, called GAz4, possesses similar characteristics to regular D-glucose and it allows monitoring glucose fluctuations in real-time. Moreover, we show that our approach, is sensitive enough to study in vivo glucose uptake and can be used for studying different biological processes. The results of this study shows that suggested method has a great potential as a useful and cost efficient approach for studying glucose-based energy consumption in various in vivo models.

Poster 15

Last Name: Maciel Ioris

First Name: Rafael

Affiliation: University of Geneva/PHYM

Presentation: Abstract submission

Title of presentation: SIRT6 hinders stemness of tumors bearing PI3K activation

Authors list: Rafael Maciel Ioris, Mirco Gallig, Giorgio Ramadori, Roberto Coppari.

Abstract: Cancer stem cells (CSCs) have high tumorigenic capacity. Here, we show that stem-like traits of specific human cancer cells are reduced by overexpression of the histone deacetylase sirtuin 6 (SIRT6). SIRT6-sensitive cancer cells bear mutations that activate phosphatidylinositol-3-kinase (PI3K) signaling, and overexpression of SIRT6 reduces growth, progression, and grade of breast cancer in a mouse model with PI3K activation. Tumor metabolomic and transcriptomic analyses reveal that SIRT6 overexpression dampens PI3K signaling and stem-like characteristics and causes metabolic rearrangements in this cancer model. Ablation of a PI3K activating mutation in otherwise isogenic cancer cells is sufficient to convert SIRT6-sensitive into SIRT6-insensitive cells. SIRT6 overexpression suppresses PI3K signaling at the transcriptional level and antagonizes tumor sphere formation independent of its histone deacetylase activity. Our data identify SIRT6 as a putative molecular target that hinders stemness of tumors with PI3K activation.

Poster 16

Last Name: Bazhin

First Name: Arkadiy A

Affiliation: EPFL

Presentation: Abstract submission

Title of presentation: From test tube to animals: a universal assay for DPP-4 activity.

Authors list: Arkadiy A. Bazhin1, Marc Chambon2, Jonathan Vesin2, Julien Bortoli2, James W. Collins1, Gerardo Turcatti2, Chieh Jason Chou3, Elena Dubikovskaya1

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KEYWORDS: dipeptidyl peptidase-4, bioluminescence, high throughput screening (HTS).

Abstract: Incretins, hormones that are secreted by L-cells of the gut, play important role in regulation of postprandial glucose level in blood1-3. Strategy of preservation of endogenous incretins from inactivation by DPP-4 was clinically validated for treatment of people suffering from type 2 diabetes mellitus (T2DM) 2 which is one of the most prevalent non-communicable diseases in the world4, 5 Development of long-acting DPP-4 inhibitors is the current focus of many pharmaceutical companies and research institutes6-8. The most widely used method for DPP-4 quantification is based on in vitro fluorescent enzymatic assays9-11 but this assay is only limited to ex-vivo applications, time consuming, and not suitable for high throughput screening (HTS) 12, 13. We have developed a novel optical-based readout method that is bereft of these disadvantages and can be used for sensitive quantification of DPP-4 activity ex-vivo, in vitro and in vivo (in live animals). Moreover, the method is easily adoptable for high throughput screening. With this approach, we conducted HTS within 4000 compounds - a library of known widely used drugs. Several hits were identified from this in vitro HTS screen and all of them were subsequently checked for its levels of DPP-4 activity in living mice using the same optical-readout assay. Interestingly, only one compound, a cancer drug mitoxantrone, possessed significant DPP-4 inhibitory activity in vivo. The mechanism of this inhibition in mice was further confirmed by measurement of active GLP-1 concentration in plasma by ELISA from animals treated with this drug. In conclusion, this novel sensitive method represents a valuable tool for DPP-4 inhibitor drug discovery starting from ex-vivo assays all the way to validation of hits in living animals. It provides a substantial improvement over existing animal studies since it completely eliminates the need for catheterization, repetitive blood sampling, and extensive animal use.