Established in 1871

SMW

Swiss Medical Weekly

Formerly: Schweizerische Medizinische Wochenschrift

Supplementum 196

ad Swiss Med Wkly
2012;142
November 17, 2012

The European Journal of Medical Sciences

44th Annual Meeting of the Swiss Society of Nephrology

Zurich (Switzerland), December 5–7, 2012

EMH FMH Schwabe
Editores Medicorum Helveticorum

Supported by the Swiss Academy of Medical Sciences (SAMS), the FMH (Swiss Medical Association) and by Schwabe AG, the long-established scientific publishing house founded in 1488

Official journal of the Swiss Society of Infectious Diseases, the Swiss Society of Internal Medicine and the Swiss Society of Rheumatology

Suppl. 196
ad Swiss Med Wkly
2012;142
November 17, 2012

Abstracted / indexed in
Index Medicus / MEDLINE
Web of science
Current Contents
Science Citation Index
EMBASE

Guidelines for authors
The Guidelines for authors are published on our website www.smw.ch
Submission to this journal proceeds totally on-line:
www.smw.ch → For Authors

All communications to:
EMH Swiss Medical Publishers Ltd.
Swiss Medical Weekly
Farnsburgerstrassse 8
CH-4132 Muttenz, Switzerland
Phone +41 61 467 85 55
Fax +41 61 467 85 56
office@smw.ch

Head of publications
Natalie Marty, MD (nmarty@emh.ch)

Managing editor
Nadine Leyser, PhD (nleyser@emh.ch)

Papers administrator
Gisela Wagner (gwagner@emh.ch)

Language editors
Thomas Brink, MD; Kirsten Clift; Judith Lutz-Burns, MD; Clark Stoppia; Roy Turnill, MA

ISSN printed version: 1424-7860
ISSN online version: 1424–3997

Regular subscription price for 2012: CHF 150.– (shipping not included)

12 print issues per year; continuous online publication

OC01

Frequency and Determinants of Pregnancy-Induced Child-Specific Sensitization

G. Hoenger[1], I. Fornaro[1], C. Granado[1], J.-M. Tiercy[2], I. Hoesli[1], S. Schaub[1]
[1]Basel, [2]Geneva

Purpose: Pregnancies are a major cause of sensitization in females awaiting organ transplantation. The aim of this study was to define the frequency and determinants of pregnancy-induced child-specific sensitization shortly after delivery.
Methods: Three hundered and one pregnancies were analyzed using sensitive single HLA-antigen beads (SAB) and high resolution HLA-typing of the mothers and their children (n = 301). A positive child-specific SAB result was defined by a background normalized ratio >1 or a mean fluorescence intensity (MFI) >300, using ten negative control sera.
Results: The overall frequency of child-specific sensitization at the HLA-A/B/C/DRB1 loci was 38% (ratio cut-off), 34% (MFI >300 cut-off), 31% (MFI >500 cut-off), and 28% (MFI >1000 cut-off), respectively. If sensitization had occurred, there were on average two child-specific HLA-antibodies. The number of live birth was associated with a higher frequency of sensitization, while miscarriages were not. There was a clear hierarchy of sensitization among the investigated loci (B-locus: 31%; A-locus: 26%; DRB1-locus: 20%; C-locus: 15%; p <0.0001). Some mismatched paternal HLA-antigens led to a significantly higher rate of sensitization than the average (e.g. HLA-A2, HLA-B49, HLA-B51, HLA-C*15). Furthermore, the mother's own HLA-phenotype – especially HLA-A/B homozygosity – was associated with a higher rate and broadness of sensitization.
Conclusion: This information can be useful to estimate the likelihood of occurred pregnancy-induced sensitization, even if no HLA-antibodies are detectable at the time of evaluation for transplantation.

OC02

Complement-Fixation Is Strongly Related to High Mean Fluorescence Intensity Values Of HLA-Antibodies

P. M. L. Amico, G. Hönger, S. Schaub
Basel

Purpose: The aim was to investigate determinants of complement-fixation of HLA-antibodies in the single HLA-antigen bead assay.
Methods: In this analysis, pre-transplant sera of 73 patients were screened for HLA-antibodies by the regular single HLA-antigen (SA) bead assay, a modified SA bead assay detecting complement-fixation (C1q-assay) and IgG1-4 subclasses. Results of the C1q-assay were then correlated with mean fluorescence intensity (MFI) values of the regular SA bead assay and IgG subclass assay.
Results: Out of 8,535 bead results, 1,895 (22%) bead results were positive by regular SA beads (MFI >500). Of these, 407 (21%) bead results were also positive by the C1q-assay (MFI >500). C1q-positive HLA-antibodies had significantly higher SA bead MFI than C1q-negative HLA-antibodies (median MFI: 10,828 [r: 3,316–19,214] vs. 2,127 [r: 501–12,134]; [p <0.0001]). C1q results were then correlated with MFI values of the IgG1-4subclasses and the SA bead assay (i.e. pan IgG). By multivariate logistic regression, higher MFI values of pan IgG (OR 20; p <0.0001), higher MFI values of the subclasses IgG1 (OR 29; p <0.0001) and IgG3 (OR 3; p <0.0001) were significantly associated with complement-fixation, whereas the subclasses IgG2 and IgG4 were not (p >0.2).
Conclusion: Complement-fixation is strongly associated with strength of HLA-antibodies (here defined by MFI value). This association is mainly driven by high level HLA-antibodies of the subclasses IgG1 and IgG3, which are known to be complement-activating. Thus, the C1q assay does not provide substantial value beyond the MFI assessed by the regular SA bead assay.

OC03

Pathology of Resolving Polyomavirus Nephropathy

T. Menter, S. Schaub, M. Mayr, H. Hirsch, M. J. Mihatsch, H. Hopfer
Basel

Purpose: Polyomavirus nephropathy (PVN) is a common complication after renal transplantation. Virus control is achieved by a reduction of immunosuppression allowing an effective T cell-mediated antiviral immune response. The morphology of resolving PVN has not been investigated.
Methods: 99 protocol biopsies of 35 patients with BK viremia treated by reduction of immunosuppression only were included and scored according to Banff criteria. The extent of interstitial inflammation was estimated as % of cortex area. The number of tubular cross sections with SV40+ cells per mm of biopsy length was counted. Findings were grouped as pre-, increasing, decreasing, and post-viremia.
Results: During the phase of decreasing viremia, we found a significant increase in the tubulitis score, the extent of tubules with intraepithelial lymphocytes, and interstitial inflammation (p <0.001). These, to a lower extent, persisted after virus clearance. The number of SV40+ tubules correlated with the virus load in the serum, but SV40 immunohistochemistry was frequently negative (33/55 cases), especially if viremia was below log 6 copies/ml.
Conclusion: Resolving PVN is characterized by a self-limiting acute interstitial nephritis. Our findings are important because the diagnosis of interstitial rejection depends on the same morphological criteria. Therefore, acute interstitial rejection cannot be diagnosed with certainty during PV viremia.

OC04

Modulation of Lymphocyte Apoptosis to Induce Mixed Chimerism and Tolerance Without Myelosuppression

P. Cippà, J. Chen, A. K. Kraus, R. P. Wüthrich, T. Fehr
Zurich

Purpose: Despite encouraging results of the first clinical studies, a broad application of tolerance induction strategies based on combined solid organ and hematopoietic stem cell transplantation to induce mixed chimerism is hampered by the toxicity of the conditioning therapy.
Methods: We investigated the role of the apoptosis pathway in a mixed chimerism induction protocol including costimulation blockade and fully MHC-mismatched bone marrow in mice.
Results: Using Bim-/- mice we found that the pro-apoptotic factor Bim was critically required to induce mixed chimerism. Conversely, by boosting the role of Bim with the small-molecule BH3-mimetic ABT-737 we were able to induce mixed chimerism with moderate doses of bone marrow cells and without any myelosuppressive conditioning. This protocol resulted in a complete deletion of peripheral donor-reactive CD8 T cells within one week after bone marrow transplantation. A stable myeloid-biased chimerism was detected over time in peripheral blood and in the thymus resulting in robust systemic donor-specific tolerance. Donor-type skin grafts were indefinitely accepted (observation time >200 days), donor-reactive antibodies and reactivity towards donor-cells in mixed lymphocyte reaction experiments were absent while transplantation of secondary donor-derived skin grafts confirmed the maintenance of robust tolerance.
Conclusion: In summary, we identified the apoptosis pathway as a new pharmacological target to induce mixed chimerism. Based on these findings we developed a new protocol that leads to tolerance across full MHC barriers in a manner that is completely independent of irradiation or myelosuppression. This approach represents a substantial advance towards a broader clinical application of tolerance as an ideal solution to prevent allograft rejection.

OC05

Outcome of Expanded Criteria Donor Kidney Transplants in an Immunological Low-Risk Population

C. Praehauser, P. Hirt-Minkowski, K. Saydam Bakar, P. Amico, E. Vogler, S. Schaub, M. Mayr
Basel

Purpose: Outcome studies of kidneys from expanded criteria donors (ECD) are poorly controlled for immunological factors. This study investigated the outcome of standard and ECD kidney recipients with low immunological risk, defined by the absence of donor-specific HLA-antibodies (HLA-DSA).
Methods: We retrospectively analyzed death censored graft survival and graft function in a cohort of 265 recipients transplanted from 1/1999 to 12/2010.
Results: 112 (42%) kidneys derived from ECD and 153 (58%) from SCD. In multivariate Cox regression ECD status was the only significant risk factor for graft failure (HR 2.82 [CI 1.27–6.26], p = 0.01). Overall, the one-, three- and five-year graft survival rates for ECD kidneys (94%/92%/80%) were lower compared to standard criteria donors (SCD) (97%/94%/93%) (p = 0.004). Stratified by immunosuppression (IS) graft survival of ECD kidneys treated with tacrolimus-mycophenolate (Tac-MPA) was comparable to graft survival of SCD kidneys (p = 0.3), whereas survival rates of ECD kidneys treated without Tac-MPA were significantly lower (88%/83%/72%) (p <0.001). Overall, ECD kidneys had a lower median eGFR (37 [5–102] ml/min) than SCD kidneys (58 [5–137] ml/min) at three years (p <0.001). This difference remained consistent after stratification for IS (p <0.001). Within the ECD group, recipients treated with Tac-MPA had a higher median eGFR at three years (43 [5–102] ml/min) and a preserved graft function from one to three years (median change –0.2 ml/min, p = 0.7) compared to those treated without Tac-MPA (34 [5–67] ml/min) (p = 0.002), who showed a significant decrease in eGFR (median change –2.2 ml/min) (p = 0.004).
Conclusion: In the absence of HLA-DSA, outcome of kidneys derived from ECD is favourable. Tac-MPA seems to improve graft survival and to preserve graft function.

OC06

NEP- Syndrome: A New Genetic Condition with Nephrotic Syndrome, Epidermolysis Bullosa and Pulmonary Disease Based on Integrin A3 Mutation

G. Spartà[1], C. Has[2], D. Kiritsi[2], L. Weibel[1], A. Moelle[1], V. Vega-Warner[3], A. Waters[4], Y. He[2], Y. Anikster[5], P. Esser[2], B. K. Straub[6], I. Hausser[6], D. Bockenhauer[5], B. Dekel[7], F. Hildebrandt[4], L. Bruckner-Tuderman[2], G. Laube[1]
[1]Zurich, [2]Freiburg/DE, [3]Ann Arbor/US, [4]London/UK, [5]TelAviv/IL, [6]Heidelberg/DE, [7]Tel Aviv/IL

Purpose: Integrin-α3 (ITGA3) is a transmembrane integrin receptor subunit mediating signals between cells and their microenvironment. Mutations in integrin genes are associated with different human disorders. We report 3 infants with congenital nephrotic syndrome, skin fragility and interstitial lung disease, who were homozygous for mutations in the ITGA3-gene.
Methods: Patient 1 was the index case in whom the genetic defect was discovered, detailed histological evaluation performed and the complex phenotype described. Subsequently, two other children with similar clinical features and ITGA3 mutations were identified. From all 3 patients and their parents genetic analysis for ITGA3 and other candidate genes was performed.
Results: Patient 1 revealed a homozygous mutation c.1173_1174del in exon 8 of ITGA3 gene, histologically leading to a loss of Integrin-α3 in the kidney, skin and lung accompanied by profound abnormalities of the basement membrane in all affected organs. Although skin fragility initially was mild, it provided clues to the diagnosis. Patients 2 and 3 were homozygous for the ITGA3 mutations c.1538-1G>C, in intron 11, and c.1883G>C,p.Arg628Pro in exon 14, respectively. The ITGA3 mutations in all patients were associated with congenital nephrotic syndrome accompanied by end stage renal failure, worsening epidermolysis bullosa and severe interstitial lung disease (NEP-Syndrome). Although patients survived neonatal period, severe multi-organ involvement led to a lethal course.
Conclusion: We identified 3 patients with homozygous mutations of ITGA3-gene associated with disrupted basement membrane structures clinically leading to NEP-syndrome. These new mutations reflect the impact and indispensability of Integrin-α3 concerning the organization of basement membrane and its clinical impact.

OC07

Tumor-Associated FGF-23 Induced Hypophosphatemic Rickets in an Eight Year Old Boy

M.-A. Burckhardt, A. Schifferli, A. Krieg, D. Baumhoer, G. Szinnai, C. Rudin
Basel

Purpose: Tumor-associated Fibroblast Growth Factor 23 (FGF-23) induced hypophosphatemic osteomalacia has primarily been described in adults. In rare occasions this entity may also cause renal phosphate wasting and rickets in children, resulting from local production of phosphatonins by various benign and malignant mesenchymal tumors.
Methods: An eight year old boy was investigated for suspected unilateral painless limping. Radiographic evaluation showed a large and polylobulated osteolysis in the left iliac bone and acetabulum. Further typical clinical signs of rickets and the respective radiographic and laboratory signs including severe renal phosphate wasting were detected.
Results: Biopsy of the iliac lesion suggested a primary solitary bone cyst overlain by a secondary and solid aneurysmal bone cyst. Laboratory findings, i.e. hypophosphatemia, renal tubular phosphate wasting, normal parathormone and normal calcitriol levels were not compatible with common forms of rickets in childhood. Tumor-associated rickets was therefore suspected and further investigated with various methods, including a PET-Scan and measurement of FGF-23 plasma levels. A causal lesion other than the iliac tumor or clearly abnormal FGF-23 plasma levels could not be found. A complete curretage and stabilization of the iliac lesion were therefore performed and local FGF-23 expression could finally be proven by immunohistochemistry in solid portions of the lesion. After surgery, tubular phosphate absorption normalized immediately and rickets quickly resolved without any further substitution of phosphate or other interventions.
Conclusion: Tumor-associated rickets have only rarely been described in children. Nevertheless this diagnosis has to be considered in pediatric patients with acquired hypophosphatemic rickets beyond infancy.

OC08

Severe Hyperkalemia in a Patient With Chronic Hemodialysis Following Colon Diversion Surgery

N. Kononowa, M. Dickenmann, M. J. Kim
Basel

Purpose: Potassium (K+) homeostasis in healthy subjects is maintained mainly by urinary excretion of K+, which is almost equal to the amount of dietary K+ingestion. In patients with end-stage renal disease (ESRD), the capacity of the colon for K+secretion increases to the extent that it makes a substantial contribution to K+homeostasis.
Methods: We report on a chronic hemodialysis (HD) patient developing severe hyperkalemia following colon diversion surgery.
Results: A 56 year-old-woman with ESRD undergoing HD suffered from ischaemic colitis, leading to ileocaecal resection and temporary ileoascendostomy. She made a good recovery and her dietary intake was normalized in the following weeks. Three weeks later, a routinely measured pre-HD serum K+ was 7.2 mmol/l, which was much higher than her usual K+ level (range 4.9–6.1). There was no evidence of metabolic acidosis and any remarkable hyperkalemia-related symptoms or signs, including ECG. Despite a dietary restriction of K+ and use of oral cation-exchange resin and low K+ dialysate, serum K+ level remained high (6.1–7.9). Six month later, the bowel continuity was successfully restored and serum K+ decreased to the previous level (5.2–5.9). The measurement of fecal K+ level before and after restoration of bowel continuity revealed a remarkable difference between the values: 23 mmol/l and 60 mmol/l, respectively. We therefore assume that the severe hyperkalemia in our patient was caused by the failing colonic secretion of K+ due to the colonic diversion.
Conclusion: To our knowledge, this is first report on severe hyperkalemia following colonic diversion in patients with ESRD undergoing HD and demonstrates the importance of colonic K+ secretion for the maintenance of K+ homeostasis in these patients.

OC09

Dialysis for Two – The Zurich Dialysis Pregnancy Experience In 2012

M. Bonani, S. Wassmer, L. Schäffer, G. Andreisek, R. P. Wüthrich, S. Segerer
Zurich

Purpose: Pregnancies in women on hemodialysis are rare and the outcome is hampered by a high number of pregnancy-related complications.
Methods: Case-report of a successful pregnancy in a hemodialysis patient.
Results: A 28-year old woman was referred for the evaluation of renal failure in the 7th week of pregnancy. She presented without clinical symptoms, but with an eGFR of 17 ml/min, proteinuria of 3.5 g/day, bicarbonate of 15 mmol/l, and a hemoglobin of 8.0 g/dl. Renal ultrasonography demonstrated a shrunken kidney on the left side and a dilated pyelon on the right side. A suspected pyeloureteral stenosis and a toxic injury by streptomycin (in the childhood for tuberculosis) were the presumed causes of the renal insufficiency. She was treated with bicarbonate, iron, as well as erythropoietin and received a peripheral native AV fistula. Aspirin (100 mg per day) was given for preeclampsia prophylaxis. Once the urea levels exceeded 16 mmol/l and metabolic acidosis worsened, dialysis was started in the 17th week of pregnancy. With the advancement of the pregnancy, dialysis was intensified to 21 hours/week divided into six sessions per week. The median dose of erythropoietin to reach the target Hb level was 16700 U/week. From 22 weeks, a moderate polyhydramnion was present. In the last trimester, slight growth retardation was noted. At week 37 an elective cesarean section was performed with delivery of a healthy boy weighing 2080 grams.
Conclusion: Despite the improvements in hemodialysis therapy, only half of the dialysis pregnancies result in healthy children. Uncertainties remain in the decision when to start dialysis. Our case illustrates that intensive dialysis and a very close interdisciplinary monitoring can result in a successful pregnancy and delivery of a healthy child.

OC10

Hemodialysis Reduces the Calcification Propensity of Serum

A. Pasch[1], S. Farese[2], J. Floege[3], D. E. Uehlinger[1], W. Jahnen-Dechent[3]
[1]Berne, [2]Solothurn, [3]Aachen/DE

Purpose: Vascular calcification is a major cause of death in hemodialysis (HD) patients. We have developed an in vitro test, which measures serum calcification propensity by detecting the spontaneous transformation of colloidal primary calciprotein particles (CPPs) to crystalline secondary CPPs. The effect of hemodialysis on serum calcification propensity has not been determined yet.
Methods: The intrinsic calcification propensity of pre- and post-HD sera obtained from 98 prevalent HD patients were analyzed with our novel test. Calcium, phosphate, magnesium, fetuin-A, albumin, and total protein concentrations were related to the test results and integrated into a multivariate model with stepwise selection.
Results: HD reduced serum calcification propensity by delaying transformation time (T50 pre-HD 244 ± 112 min., post-HD 340 ± 114 min., p <0.0001) and reducing precipitation intensity (relative nephelometric units, RNU50 pre-HD 6892 ± 2404, post-HD 5234 ± 1789, p <0.0001). A multivariate model showed, that the T50 of pre-HD sera depended mainly on magnesium (transformation delay, p <0.0001) and fetuin-A (delay, p <0.0001), and the HD-induced delay of T50 on phosphate (acceleration, p <0.0001), magnesium (delay, p = 0.0094) and fetuin-A (delay, p <0.0001) serum concentrations. In contrast, the reduction of precipitation intensity RNU50 induced by HD depended on the change of the total serum protein concentration (p = 0.0407), which was closely correlated to the albumin and fetuin-A concentration changes induced by HD (p <0.0001).
Conclusion: HD vastly improves the intrinsic calcification propensity of sera, with phosphate, magnesium and fetuin-A as major influencing factors. Monitoring serum-inherent calcification propensity may help improve morbidity and mortality of HD patients in the future.

OC11

Klotho as a Prognostic Marker in Patients on Maintenance Hemodialysis

P. Ambühl[1], A. Starke[1], I. Pavik[1], R. P. Wüthrich[1], M. Miozzari[2], D. Kiss[3], D. Aerne[4], H.-R. Räz[5], T. Kistler[6], M. Hersberger[1], A. Serra[1]
[1]Zurich, [2]Schaffhausen, [3]Liestal, [4]Lachen, [5]Baden, [6]Winterthur

Purpose: Circulating Klotho has been identified as one of the key hormones regulating calcium-phosphate metabolism, and, potentially, as a protective factor against atherosclerotic vasculopathies. The aim of this study was to investigate the relationship of Klotho with survival on chronic maintenance hemodialysis (HD) therapy.
Methods: Baseline Klotho levels were measured in the year 2006 from 150 chronic maintenance HD patients, aged 67 ± 13 years with a dialysis vintage of 6.4 ± 3 years, enrolled in the *monitor!* study, a prospective cohort of HD patients. Klotho baseline levels were correlated with epidemiological and biochemical outcome data.
Results: Sixty two (41%) patients died during follow-up of 855 ± 429 days. Survivors were younger, had a higher body weight, a higher number of comorbidities, and were more likely to be on vitamin D supplements. Klotho plasma levels were similar between survivors and non-survivors (404 ± 173 versus 380 ± 204 pg/ml, respectively; P = 0.439). Klotho was negatively correlated with age, number of comorbidities, and LDL-cholesterol, and positively correlated with serum alkaline phosphatase and PAPP-A. In a multivariate logistic regression model including Klotho along with the covariates mentioned before, only age, PAPP-A and vitamin D therapy were independently predictive for survival. The lowest tertile of Klotho levels was associated with a trend towards a higher vitamin D supplementation rate, whereas Klotho did not correlate with serum calcium, phosphate, PTH and cardiovascular risk biomarkers such as IL-6, BNP, and CRP.
Conclusion: Klotho does not predict survival in chronic HD patients. Klotho is inversely related to age, and may be modified by vitamin D treatment, but is unrelated to biomarkers of bone mineral metabolism, except for alkaline phosphatase.

OC12

Stimulated Sweating as a Therapy To Reduce Interdialytic Weight Gain and Improve Potassium Balance in Chronic Hemodialysis Patients: A Pilot Study

M. Pruijm[1], Y. El-Housseini[1], H. Mahfoudh[2], F. Jarraya[2], J. Hachicha[2], D. Teta[1], M. Burnier[1]
[1]Lausanne, [2]Sfax/TN

Purpose: Controlling the extracellular volume in hemodialysis patients is a difficult task. The aim of this study was to evaluate the capacity of different methods of stimulated sweating to reduce mean interdialytic weight gain (IWG), to improve blood pressure regulation and potassium/urea balance.
Methods: Two center, cross over pilot study. In Lausanne, hemodialysis patients took four hot water baths a week of 30 minutes each, on non-dialysis days during one month. In Sfax, patients visited four times a week the local hammam-center. Hemodynamic parameters were recorded, and weekly laboratory analysis performed. Results were compared with a preceding one-month control period.
Results: In Lausanne, five patients (all men, median age 55y) participated. Bathing temperature was (mean ± SD) 41.2 ± 3 °C and sweating-induced weight loss 600 ± 500 g. Mean IWG (control versus intervention period) decreased from 2.3 ± 0.9 to 1.8 ± 1 kg (p = 0.004), SBP from 139 ± 21 to 136 ± 22 mm Hg (p = 0.4), and DBP form 79 ± 12 to 75 ± 13 mm Hg (p = 0.08); antihypertensive therapy could be reduced from 2.8 ± 0.4 to 1.9 ± 0.5 antihypertensive drugs per patient (p = 0.01). In Sfax (n = 9, median age 46y), weight loss per hammam session was 420 ± 100 g. No differences were found in IWG or BP, but pre-dialysis serum potassium level decreased from 5.9 ± 0.8 to 5.5 ± 0.9 mmol/l (p = 0.04) and urea from 26.9 ± 6 to 23.1 ± 6 mmol/l (p = 0.02).
Conclusion: Hot water baths appear to be a safe way to reduce interdialytic weight gain in selected haemodialysis patients. Hammam visits reduce serum potassium and urea levels, but not IWG. More data in larger patient groups are necessary before definite conclusion can be drawn.

OC13

The Spectrum of Podoplanin Expression in Encapsulating Peritoneal Sclerosis

N. Braun[1], D. M. Alscher[1], P. Fritz[1], J. Latus[1], I. Edenhofer[2], F. Reimold[1], S. L. Alper[3], M. Kimmel[1], D. Biegger[1], M. Lindenmeyer[2], C. D. Cohen[2], R. P. Wüthrich[2], S. Segerer[2]
[1]Stuttgart/DE, [2]Zurich, [3]Boston/US

Purpose: Podoplanin is a glycoprotein expressed by mesothelial cells, lymphatic endothelial cells, and myofibroblasts in peritoneal biopsies from patients with encapsulating peritoneal sclerosis (EPS). To further evaluate podoplanin as a marker of EPS we measured podoplanin mRNA levels and described the morphological patterns of podoplanin-positive cells in EPS.
Methods: Included were 20 peritoneal biopsies from patients on PD with the diagnosis of EPS (n = 5), patients on PD without signs of EPS (n = 5), and control patients (n = 10). In 24 peritoneal biopsies with EPS, podoplanin and smooth muscle actin (SMA) were localized by immunohistochemistry.
Results: EPS patient biopsies revealed a significantly elevated levels of podoplanin mRNA (p <0.05). The most common podoplanin pattern (8 of 24) consisted of organized, longitudinal layers of podoplanin-positive cells and vessels in the fibrotic zone ("organized" pattern). 7 of 24 biopsies demonstrated a diffuse distribution of podoplanin-positive cells. Five biopsies exhibited a "mixed" pattern. These contained cuboidal podoplanin-positive cells within SMA-negative epithelial structures embedded in extracellular matrix. Less frequently observed was the complete absence of, or only focal accumulations of podoplanin-positive fibroblasts outside of lymphatic vessels (podoplanin "low", 4 of 24 biopsies). Patients in this "low" group exhibited a lower index of systemic inflammation and a longer symptomatic period than in EPS patients with biopsies of the "mixed" type (p <0.05).
Conclusion: In summary we describe a novel cell differentiation process involved in EPS, confirm the increased expression of podoplanin in EPS, and distinguish EPS biopsies according to different podoplanin expression patterns which are associated with clinical parameters.

OC14

An Eye-Catching View of the Glomerulus: In vivo Imaging of Glomeruli Transplanted into the Anterior Chamber of the Mouse Eye

A. Kistler[1], A. Caicedo[2], J. Reiser[2], A. Fornoni[2]
[1]Zurich, [2]Miami/US

Purpose: Advanced in vivo imaging technologies allow to study complex physiological and pathological processes. Multiphoton microscopy enables live imaging of the kidney. However, longitudinal in vivo imaging of glomerular structure and function independent of the effects of parietal epithelial and proximal tubular cells are currently limited. Here, we utilized the anterior chamber of the mouse eye as a natural body window to image transplanted glomeruli engrafted on top of the highly vascularized iris.
Methods: Mouse glomeruli were isolated by sequential sieving, handpicked and microinjected into the anterior chamber of recipient mice. For allogeneic transplantation, immune deficient recipient mice were used.
Results: Within 3 weeks after transplantation 10% of transplanted glomeruli gained access to the iris vasculature, and became perfused. By transplanting glomeruli from podocyte-specific CFP expressing mice, we were able to visualize transplanted podocytes and demonstrated their survival for several months. Electron microscopy confirmed preservation of interdigitating foot processes. Both donor and recipient endothelial cells contributed to the glomerular filtration barrier, as revealed by endothelial cell specific GFP expression in recipient mice. Intravenous injection of fluorescence-labeled dextrans confirmed functionality of transplanted glomeruli as 10 kDa dextran was filtered into the subpodocyte space that restricted further passage of the dextran. 70 kDa dextran particles were not filtered through the glomerular capillaries and were detected in the subpodocyte space only after disease induction in an inducible transgenic model of podocyte injury.
Conclusion: We established a novel technique for non-invasive and longitudinal study of glomerular physiology and pathophysiology in living animals.

OC15

Non-Invasive Arterial Spin Labelling and T1 Relaxation Time Magnetic Resonance Imaging Provides Insights into the Pathophysiology of Cardiorenal Syndrome

T. Breidthardt[1], E. Cox[2], I. Squire[3], A. Odudu[4], T. Eldehni[4], S. Francis[2], C. McIntyre[4]
[1]Basel, [2]Nottingham/UK, [3]Leicester/UK, [4]Derby/UK

Purpose: The pathophysiology of the chronic cardiorenal syndrome is not fully understood. Recently Arterial Spin Labelling (ASL) and T1 relaxation time measurements (non-invasive, contrast-free, free-breathing MRI techniques) became available and allow us to assess true tissue perfusion and renal structur in vivo.
Methods: 41 participants were enrolled in 4 groups according to their HF and renal impairment status [groups 1/2: healthy volunteers (1: <40 years; 2: HF age matched); groups 3/4 stable HF (3: eGFR >60 ml/min; 4: eGFR <60 ml/min)]. The association of ASL-MRI and T1 relaxation time ($T1\rho$) with renal function was the primary endpoint.
Results: Renal cortical perfusion correlated with eGFR values ($r = 0.52$, $p < 0.01$) and was lower in HF patients vs. volunteers (161 ml/mg ± 42 vs. 267 ml/mg ± 91; $p < 0.01$). There was no significant difference in renal perfusion between HF patients with and without renal dysfunction ($p = 0.27$). $T1\rho$ correlated negatively with eGFR values ($r = –0.41$; $p > 0.01$) and was higher in HF patients vs. volunteers (1121 ± 102 ms vs. 1054 ± 65 ms; $p = 0.03$). There were no $T1\rho$ differences between groups 1, 2 and 3 (1080 ± 68 ms vs. 1029 ± 55 ms; $p = 0.10$; 066 ± 78 ms; p vs Group 2 = 0.27). $T1\rho$ was selectively prolonged in HF patients with renal dysfunction (1170 ± 100 ms, p vs. HF = 0.04). In linear regression analyses only history of coronary artery disease (OR 82 [95%CI 18–147] $p = 0.01$), hypertension (OR 59 [95%CI 1–117]; $p = 0.04$) and diabetes mellitus (OR 113 [95%CI 47–179], $p < 0.01$) were associated with $t1\rho$.
Conclusion: Renal dysfunction in HF is not primarily mediated by decreased renal perfusion. Contrastingly, prolonged $T1\rho$ reflecting chronic structural renal changes might be the primary culprit in the chronic cardiorenal syndrome. These structural changes appear to be associated with classical cardiovascular risk factors.

OC16

A Common Variant in UMOD, Associated With the Risk Of Chronic Kidney Disease and Hypertension, Influences the Urinary Excretion Of Uromodulin

S. Youhanna[1], J. Weber[1], R. Glaudemans[1], C. Hayward[2], M. Bochud[3], O. Devuyst[1]
[1]Zurich, [2]Edinburgh/UK, [3]Lausanne

Purpose: Uromodulin is exclusively produced in the thick ascending limb of the Henle's loop and is the most abundant protein secreted in normal urine. Mutations in the *UMOD* gene that codes for uromodulin are responsible for autosomal-dominant kidney diseases characterized by hyperuricemia and gout, interstitial fibrosis and progressive renal failure. Genome-wide association studies (GWAS) have shown that variants in *UMOD* are associated with the risk of developing hypertension and chronic kidney disease (CKD) in the general population. The biological mechanism of these associations remains unknown.
Methods: In these studies, we developed a specific ELISA to determine urinary uromodulin levels and characterized the optimal conditions of handling and storage for a stable uromodulin. We generated the first large database of uromodulin levels in more than 10,000 samples collected from 4 genetic isolates and a large urban population. We then performed a GWAS to find loci associated with urinary uromodulin. Finally, we characterized the biological relevance of the top variant identified as modifier of uromodulin excretion.
Results: A common variant (rs4293393) located in the promoter of *UMOD* appears to be the most important in regulating the level of uromodulin in urine. This variant acts as a potent regulator of the transcriptional activity of *UMOD*, as evidenced from luciferase reporter gene assays in renal epithelial cells. The major allele of rs4293393, which is consistently associated with the risk of CKD and hypertension in GWAS, is associated with a dose-dependent increase in urinary uromodulin levels in these cohorts.
Conclusion: These results give insights into the regulation of uromodulin excretion and the association between *UMOD* variants and the risk of CKD and hypertension.

OC17

Rapid Homeostatic Effects of Oral Potassium Loading on the Kidney

M. Sorensen[1], S. Grossmann[1], M. Rösinger[1], D. Loffing-Cueni[1], G. Barmettler[1], U. Ziegler[1], A. Odermatt[2], O. Staub[3], J. Loffing[1]
[1]Zurich, [2]Basel, [3]Lausanne

Purpose: A large dietary potassium (K+) load is a homeostatic challenge for mammals. It is known to induce a rapid kaliuretic and natriuretic response. These renal effects are reported to occur even before plasma K+ and aldosterone levels increase. Here we elucidate the underlying molecular mechanisms of K+ induced kaliuretic and natriuretic response.
Methods: We analyzed in mice the time course (15', 30', 2h, and 6h) of the effect of a gastric K+ load on plasma ion concentrations, aldo levels, urinary ion excretion, and expression and/or phosphorylation of renal ion transport proteins.
Results: Following a gastric gavage of 2% KCl, plasma K+ concentrations rose rapidly (at 15'), followed by a significant rise of plasma aldo (at 30'). Enhanced urinary K+ and Na+ excretion was detectable as early as spot urines could be collected (~30'). The functional changes were accompanied by a rapid and sustained dephosphorylation of the NaCl cotransporter (NCC) (15'-6h) and a later up-regulation of proteolytic activated epithelial sodium channels (ENaC) (6h). The rapid effect on NCC and the late effects on ENaC were independent from the co-administered anion (same effect with KHCO3; no effect with NaCl). In contrast to the proteolytic ENaC regulation, NCC dephosphorylation was independent of plasma aldo as indicated by experiments in aldo-deficient mice. The observed urinary Na+ loss was likely related to NCC, as it was not seen in NCC-deficient mice.
Conclusion: Rapid down-regulation of NCC contributes to the early kaliuresis and explains the natriuresis in response to an oral K+ load. Enhanced activation of ENaC occurs quite late and might be more important for the long-term control of K+ homeostasis.

OC18

Daytime Sleepiness Associated with Immunosuppressive Non-Adherence in Renal Transplant Recipients: A Cross-Sectional Multi-Center Study

H. Burkhalter[1], A. Wirz-Justice[1], C. Cajochen[1], T. Weaver[2], J. Steiger[1], T. Fehr[3], R. M. Venzin[4], S. de Geest[1]
[1]Basel/CH, [2]Chicago/US, [3]Zurich/CH, [4]Bern/CH

Background: Medication non adherence (NA) is common in renal transplant (RTx) recipients and is associated with negative clinical and economic outcomes. The aim of this study was to determine the prevalence and to assess a potential association between NA and daytime sleepiness (DS) in RTx.
Methods: Using a cross-sectional design, a convenience sample of 927 home dwelling RTx recipients who received their transplant at one of three Swiss transplant centers were enrolled in the study. Data on NA, DS and depression were collected by self-report. Non-adherence was assessed using the Basel Assessment of Adherence Scale for Immunosuppressives, DS using the Epworth Sleepiness Scale and depression with the Depression, Anxiety and Stress Scale. Binary logistic regression controlling for depression, co-morbidities, gender, age and years since Tx was used for the analysis.
Results: The prevalence of DS was 52%, taking NA 16%, timing NA 42% and overall NA 35%. Taking and timing NA were positively associated with more DS and longer time since Tx. The multivariate model showed that DS is a significant (p <0.001) predictor for taking [1.06 (1.01–1.11)], timing [1.07 (1.03–1.11)] and overall NA [1.09 (1.05–1.13)]. Further, greater time since transplantation increased the odds of taking by 28%, timing 18% and overall NA by 18%.
Conclusion: There is an association between DS and immunosuppressive NA in transplantation. This a novel finding that provides better understanding of NA, especially in view of non-intentional NA where forgetfulness is a driving factor.

OC19

Rôle de l'infirmière dans la prise en charge et le suivi de l'observance thérapeutique chez des patients dialysés: expérience tirée d'une étude clinique au CHUV

C. Zweiacker, V. L. Forni, M. Burnier
Lausanne/CH

Introduction: L'objectif est d'évaluer si un suivi intensif de l'adhérence des patients associé à des entretiens de motivation conduits par une infirmière améliore le contrôle de l'hyperparathyroïdisme dans un service de dialyse.
Méthode: Les patients dialysés chroniques traités par cinacalcet sont randomisés en 2 groupes: un groupe contrôle sans intervention et un groupe avec une prise en charge intensive de l'observance. Le rôle de l'infirmière comprend: la gestion du médicament, du pilulier électronique, l'exécution des entretiens motivationnels chaque 2 mois avec lecture du pilulier, l'identification et la résolution des obstacles à la prise du médicament.
Résultats: 13 patients sont suivis par l'infirmière. Deux cas illustrent son travail: 1) Femme 42 ans avec une PTH élevée. Les doses de cinacalcet sont augmentées de 60 mg/j à 120 mg/j mais la PTH reste élevée. A 6 mois, on découvre une adhésion thérapeutique de 40%. Un entretien motivationnel est réalisé dès la fin de l'étude. 2) Homme 30 ans avec une PTH initiale à 417 ng/L. A 2 mois, la PTH est à 1881 ng/L mais l'adhésion thérapeutique est à 30%. L'entretien met en évidence plusieurs obstacles: peur, superstition culturelle face au médicament, accessibilité des médicaments. Après 2 entretiens visant à comprendre ses représentations et à expliquer les bénéfices du traitement l'adhésion s'améliore à >70% et la PTH chute à 205 ng/L. La dose du médicament est réduite.
Conclusion: L'infirmière joue un rôle très important dans la prise en charge des problèmes d'adhésion thérapeutique des dialysés. Le contact fréquent avec le patient lui permet d'identifier les sujets à risques, d'aider les patients à verbaliser leurs problèmes et trouver de nouvelles ressources pour réintégrer le médicament dans leur structure de vie.

OC20

Factors associated with quality of ambulatory blood pressure monitoring in the population-based SKIPOGH study

M.-O. Levy[1], S. Tremblay[2], U. Schupbach[3], G. Gok-Sogut[2], S. Estoppey-Younes[2], M. Bochud[2], O. B. O. The Skipogh Study[2]
[1]Geneva, [2]Lausanne, [3]Berne

Purpose: Ambulatory blood pressure monitoring (ABPM) has limited success rates which impacts population-based studies. We hypothesized that certain participants' characteristics can be identified, which are associated with quality of ABPM.
Methods and results: Families were randomly selected from the general adult population in Lausanne, Geneva, and Bern. Validated Diasys Integra devices were used for ABPM. We used generalized estimating equations with a logit link and binomial distribution to explore predictors of having less then 80% valid measurements. We included 320 men and 325 women. The mean (SD) age was 47.6 (17.8) / 49.8 (16.9) years respectively, the mean BMI was 26.2 (4.4) / 24.1 (4.5) kg/m^2, and the mean (SD) number of measurements over 24-hour was 69 (13). The prevalence of overweight and obesity were 33% and 13%, respectively. Twenty-five percent of participants had less than 80% valid ABPM measurements. Main predictors of having less than 80% valid measurements were age older than 60 years (OR [SE] = 1.69 [0.41], P = 0.03), being obese (OR [SE] = 1.87 [0.55], P = 0.03), and having low pulse pressure (OR [SE] = 0.96 [0.01] per mm Hg, P <0.001). The association with obesity became non-significant upon adjustment for arm circumference.
Conclusions: In conclusion, selected characteristics of participants were identifiable that limit the accomplishment of an integral ABPM. Specifically, this was older age, obesity, and low pulse pressure. The effect of obesity was largely explained by large arm circumference. These results can help guiding nurses when instructing participants before monitoring.

OC21

Functional dependence related to physical activity in hemodialysis patients analyzed in a large prospective Swiss dialysis cohort

R. Winzeler[1], F. Barnert[2], L. Walther[3], C. Studer[1], M. Stücheli-Morssinkhov[4], B. Sam Aka[5], G. Herfs[3], D. Kiss[6], P. Ambühl[1]
[1]Zurich, [2]Lachen, [3]Baden, [4]Schaffhausen, [5]Winterthur, [6]Liestal

Purpose: Functional impairment in hemodialysis (HD) patients has been suggested to severely impact on survival and quality of live (QoL). The aim of the present study was to analyze physical capacity and bodily activity in relation to functional status in a Swiss HD population.
Methods: 375 patients were evaluated from the *monitor!* project, a prospective dynamic hemodialysis cohort assessing a wide range of clinical, laboratory and anthropometrical data. Functional status was determined by the indices for activity of daily living (ADL, "Barthel score"), the instrumental activity of daily living (IADL) and the social adaptability index (SAI). Physical capacity was measured by three-minute walk test (3MWT), upper body strength (UBS) by a handgrip dynamometer and 24-hour step count by an armband motion detector (sensewear®, Bodymedia).
Results: Mean scores for ADL, IADL and SAI were 92 ± 14, 5.4 ± 2, and 6.7 ± 2, respectively, indicating relevant impairment in functional activities of daily live and social adaptability. All 3 scores are closely and significantly correlated to each other.
Conclusion: Functional dependence is high in this Swiss HD cohort and directly related to physical impairment and physical quality of life. Measures should be taken to improve physical capacity and activity in order to reduce functional impairment and, thereby, improve outcomes in quality of life and survival in HD patients.

ADL score, median	Age, yr	CCI*	3MWT, m	UBS, kg	Steps/day, n	QoL physical
Low	70.5 ± 13	4.1 ± 2	130 ± 62	21 ± 10	2299 ± 2446	33.2 ± 10
High	66.3 ± 14**	3.6 ± 2**	179 ± 55**	27 ± 10**	3313 ± 3168**	41.6 ± 11**

*) Charlson comorbidity index; **) P <0.05 vs. "low ADL"
By multivariate analysis, ADL was independently correlated with age, 3MWT, UBS and 24-hour step count, but not CCI. Physical QoL was positively correlated with ADL scores.

P01

Soluble CD30 Correlates With Clinical but Not Subclinical Renal Allograft Rejection

P. Hirt-Minkowski, M. Roth, G. Hönger, P. Amico, H. Hopfer, S. Schaub
Basel

Purpose: Soluble CD30 (sCD30) has been proposed as a promising non-invasive biomarker for clinical renal allograft rejection, but its diagnostic characteristics have not been assessed in subclinical rejection.
Methods: We investigated sCD30 in 146 consecutive kidney allograft recipients under tacrolimus-based immunosuppression having 250 surveillance biopsies at 3 and 6 months as well as 52 indication biopsies within the first year post-transplant. Allograft histology results were classified as (i) acute Banff score zero or interstitial infiltrates only, (ii) tubulitis t1, (iii) tubulitis t2-3, and (iv) isolated vascular compartment inflammation.
Results: sCD30 correlated well with the extent of clinical ($p < 0.0001$) but not subclinical tubulo-interstitial rejection ($p = 0.06$). To determine diagnostic characteristics of sCD30, histological groups were assigned to two categories: no relevant inflammation (i.e. acute Banff score zero and interstitial infiltrates only) versus all other pathologies (tubulitis t1-3 and isolated vascular compartment inflammation). For clinical allograft inflammation, AUC was 0.87 (sensitivity 89%, specificity 79%; $p = 0.0006$); however for subclinical inflammation, AUC was only 0.59 (sensitivity 50%, specificity 69%; $p = 0.47$).
Conclusion: In conclusion, sCD30 correlated with clinical but not subclinical renal allograft rejection limiting its clinical utility as a non-invasive rejection screening biomarker in patients with stable allograft function.

P02

Lack of Contribution of Interferon Gamma Release Assays (Igras) to the Diagnosis of Latent Tuberculosis Infection after Renal Transplantation

K. Hadaya, P.-O. Bridevaux, P. Roux-Lombard, A. Delort, P. Saudan, P.-Y. Martin, J.-P. Janssens
Geneva

Purpose: Renal transplant recipients, as all immune-suppressed patients, are at increased risk of reactivating latent tuberculosis infection (LTBI). Detecting LTBI in this population is therefore important to prevent active tuberculosis. The tuberculin skin test has a poor sensitivity in this setting.
Methods: The aim of this prospective study was to compare the diagnostic performance of the tuberculin skin test with 2 interferon gamma release assays (IGRAs): T-SPOT.TB (OxfordImmunotec,UK) and QuantiFERON-Gold-In tube (QGIT,Cellestis,Australia), performed simultaneously, for the detection of patients with a probable LTBI or a definite history of tuberculosis, among renal transplant recipients under stable immune-suppression.
Results: 205 patients (aged 59 ± 13 years, tested 10.4 ± 7.1 years post transplant) were studied. Positivity rate was 4.9% for tuberculin skin test, 20.2% for T-SPOT.TB and 23.8% for QGIT. Agreement between interferon gamma release assays was fair ($\kappa = 0.71$). Sensitivity of T-SPOT.TB and QGIT for detection of LTBI was 33.3% (95%CI: 19.6–49.5); specificity was 85.5% (78.9–90.7) and 80.1% (72.9-86.2), respectively. Combining interferon gamma release assays did not significantly improve either sensitivity or specificity.
Conclusion: Since their sensitivity for detecting probable latent tuberculosis infection in renal transplant recipients is very low, IGRAs cannot be used to exclude such an infection. It remains therefore mandatory to diagnose and treat latent tuberculosis infection before transplantation.

P03

Serum CXCL10 and Vascular Lesions in Surveillance Biopsies

P. Hirt-Minkowski[1], J. Ho[2], A. Gao[2], P. Amico[1], H. Hopfer[1], P. Nickerson[2], S. Schaub[1]
[1]Basel, [2]Winnipeg/CA

Purpose: Non-invasive biomarker correlating with subclinical allograft rejection would be very useful to identify patients who should be further investigated by surveillance biopsies. Previously, we have reported that urinary CXCL10 is a promising non-invasive biomarker for subclinical tubulointerstitial rejection, but it did not reflect vascular inflammation (i.e. glomerulitis, endothelialitis, peritubular capillaritis). The aim of this study was to investigate whether serum CXCL10 correlates with subclinical vascular inflammation.
Methods: For this pilot study, 42 surveillance biopsies were selected and grouped according to histology: (i) Banff acute score zero (n = 10), (ii) tubulitis t1-3 without vascular inflammation (n = 16), (iii) vascular inflammation (n = 16). Serum CXCL10 was measured by ELISA.
Results: There were no differences among the three histological groups regarding eGFR (49 vs 44 vs 44 ml/min; $p = 0.76$), urinary protein/creatinine ratio (13 vs 15 vs 12 mg/mmol; $p = 0.42$), urinary albumin/creatinine ratio (2.5 vs 3.6 vs 2.7 mg/mmol; $p = 0.55$), CRP ($p = 0.32$), and leucocyte count ($p = 0.99$). Urinary CXCL10 levels were significantly higher in the tubulitis t1-3 group than in the other two groups ($p < 0.01$). By contrast, serum CXCL10 levels were significantly higher in the vascular inflammation group than in the tubulitis t1-3 and the Banff acute score zero group (96 vs 50 vs 41 pg/ml; $p < 0.007$). Ten of 42 patients had concomitant infections (i.e. urinary tract infection, CMV- and/or BKV-viremia). Serum CXCL10 levels were not different between patient with/without concomitant infections (69 vs 65 pg/ml; $p = 0.67$).
Conclusion: In this small pilot study, serum CXCL10 levels correlated with subclinical vascular inflammation. These results require validation in a larger patient population.

P05

A Pilot Study in Kidney Transplant Recipients to Monitor EBV-Specific Cellular Immunity with Urea-Formulated BZLF1

L. Deml[1], S. Barabas[1], F. Zeman[1], H. Bendfeldt[1], A. Starke[1], A. Pfister[1], B. Krüger[2], W. Jilg[1], H. Wolf[1], M. Koller[1], B. K. Krämer[2], B. Banas[1]
[1]Regensburg/DE, [2]Mannheim/DE

Purpose: Epstein-Barr virus (EBV) infections are effectively controlled by cell-mediated immunity (CMI). Inadequate impairment of CMI by immunosuppressive treatment after transplantation (tx) occasionally causes EBV-reactivation potentially associated with serious complications (PTLD). The prospective pilot study aimed to investigate the suitability of (u)BZLF1 to monitor functional EBV-reactive CMI in kidney transplant recipients in the course of immunosuppressive treatment.
Methods: We performed a 2-year prospective observational study in 83 patients, of whom 92.8% had a positive EBV serostatus. Blood was collected pre and post tx to measure functional (u)BZLF1-reactive effector cells with an IFN-γ ELISpot. EBV load was determined from serum by quantitative PRC.
Results: Prior to tx, a significant (u)BZLF1-reactive CMI was detected in 63.8% of EBV-seropositive patients followed by a substantial decrease within the first 3 weeks post tx. In some patients, EBV-specific CMI started to recover in month 6 to 18. Transient and weak, clinically inapparent EBV-reactivations occurred in 53% of the patients. Individual patient courses revealed that reactivations appeared preferentially at time points with low numbers of (u)BZLF1-reactive leukocytes.
Conclusion: Monitoring of functional (u)BZLF1-reactive CMI may be an interesting novel strategy to assess the risk for EBV-reactivation in immunocompromised transplant recipients.

P06

Feasibility of T-Track® Cmv and T-Track® Ebv to Assess the Functionality of Virus-Specific Cell-Mediated Immunity (Cmi)

B. Banas[1], C. Böger[1], G. Lückoff[2], B. Krüger[3], A. Starke[1], J. Batzilla[1], M. Schemmerer[1], J. Köstler[1], L. Deml[1], J. Leicht[4], B. K. Krämer[3]
[1]Regensburg/DE, [2]Landshut/DE, [3]Mannheim/DE, [4]Schwandorf/DE

Purpose: Inadequate impairment of cell-mediated immunity (CMI) by immunosuppressive therapy is a major cause of severe clinical complications in solid organ transplantation. Assessment of CMI may help to predict the onset of complications, as well as to adjust immunosuppressive/antiviral therapy. The aim of this cross sectional multicenter study was to evaluate the feasibility of the novel tools T-Track® CMV and T-Track® EBV to assess the functionality of virus-specific CMI in a clinically relevant patient population. Applying urea-formulated viral stimulator antigens, T-Track® assays provide information about the functionality of a broad network of virus-reactive effector cells.
Methods: The test sensitivities of T-Track® CMV/EBV assays were examined in a cohort of 124 hemodialysis patients and compared with Quantiferon®-CMV and a panel of 6 preselected CMV tetramers.
Results: Herein, positive T-Track® CMV or T-Track® EBV results were obtained in 60/67 (89.6%) of CMV- and 104/118 (88.1%) of EBV-seropositive hemodialysis patients, respectively. In addition, also 12/57 (21.1%) of CMV- seronegative dialysis patients showed significant numbers of CMV-responsive cells. Quantiferon®-CMV and CMV tetramers revealed sensitivities of 72.6% (45/62) and 76.9% (40/52), respectively.
Conclusion: T-Track® CMV/EBV assays can be used in a broad population of hemodialysis patients neglecting their HLA-type. Thus, T-Track® assays may also represent valuable tools to assess functionality of CMI in transplant recipients and help to guide personalized therapy.

P07

Mother, Child and Graft Outcome after Pregnancy in Kidney Transplantation: A Retrospective Single–Center Study

F. Langner-Viviani, J.-P. Venetz, Y. Vial, M. Pascual, D. Golshayan
Lausanne

Purpose: The management of pregnant women after transplantation (Tx) remains a challenge. Although fertility is restored in many patients soon after kidney Tx, pregnancy remains a relatively rare event in most centres. The risks of complications for the mother and fetus are proportional to the degree of functional impairment of the grafted kidney, proteinuria and/or arterial hypertension at the time of conception, as described for the general population. Besides, pregnant transplant recipients are exposed to complications related to the immunosuppressive treatment as well as graft rejection and aggravation of renal function.
Methods: In this single-centre study, we retrospectively analyzed the effects of pregnancy in our cohort of kidney transplant recipients. The variables studied were: type of nephropathy, living donors (LD) vs. deceased donors (DD), age, time between Tx and pregnancy, immunosuppressive treatments, creatinine/blood pressure/urinary proteine:creatinine at conception and at different time-points during and after pregnancy, complications including infectious events, anti-HLA antibodies status and rejection episodes, gestation time, type of delivery, child birth weight.
Results: Overall, we describe in detail 14 pregnancies in 11 kidney transplant recipients in our centre between 1994 and 2011. Our results suggest that pregnancy can be successful if carried out under tight nephrological and obstetrical surveillance, as well as in optimal circumstances including stable allograft function for at least one year after Tx, good control of blood pressure, no/low level proteinuria and appropriate adjustment of immunosuppression prior to conception.
Conclusion: Thus, pregnancy after Tx should be carefully planned to limit risks iand insure best outcome for mother and fetus.

P08

A Randomized Open-Label Clinical Trial Examining The Effect Of Denosumab on the Prevention of First-Year Bone Mineral Density (BMD) Loss after Renal Transplantation (POSTOP Study; NCT01377467)

M. Bonani, A. Serra, T. Fehr, J. Brockmann, M. Schiesser, D. Frey, R. P. Wüthrich
Zurich

Purpose: Renal allograft recipients are at risk for bone loss after transplantation. Denosumab is a humanized monoclonal antibody targeting RANK ligand, which is effective in the treatment for postmenopausal osteoporosis. Whether Denosumab is effective to prevent BMD loss after renal transplantation has not been evaluated.
Methods: POSTOP is a randomized study testing the efficacy and safety of Denosumab to prevent bone loss in the first year after kidney transplantation. Patients are recruited within 28 days after transplantation. After BMD measurement by DXA, study patients are randomized 1:1 to receive Denosumab 60 mg s.c. at baseline and after 6 months, or no treatment, in addition to standard treatment. The primary endpoint is the change in total hip BMD after one year. Secondary endpoints include changes in bone mineral metabolism parameters, fractures, and allograft function.
Results: Of the planned 100 patients 43 were recruited (21 Denosumab vs. 22 control), representing an exposure time of 145 patient-months for the 21 subjects randomized to Denosumab. Their mean age was 48 ± 14 years. Their eGFR at baseline was 54 ± 17 ml/min/1.73 m^2. The DXA T-score values for total hip were – 0.7 ± 0.8 and for lumbar spine –1.0 ± 1.1. The DXA BMD-values for total hip were 0.89 ± 0.12 g/cm^2 and for the lumbar spine 0.97 ± 0.13 g/cm^2. Among the 43 randomized patients, 53% had osteopenia. The calcium values were 2.30 ± 0.19 mmol/l, phosphate 0.61 ± 0.36mmol/l, 25-OH vitamin D 17.2 ± 9.6 µg/l, and PTH 180 ± 179 ng/l. The mean cumulative dose of steroids until randomization was 1123 ± 449 mg. Among the adverse events urinary tract infections appeared to be more frequent with Denosumab treatment.
Conclusion: The POSTOP trial represents the first study to investigate whether Denosumab prevents BMD loss in the first year after kidney transplantation.

P09

Renal Replacement Lipomatosis Mimicking Tumor After Renal Transplantation

M. Bonani, T. Fehr, D. Fischer, B. Bode-Lesniewska, R. P. Wüthrich, A. Gaspert
Zurich

Purpose: n/a
Methods: We present two patients who underwent nephrectomy due to suspicion of malignant renal tumors.
Results: Case 1: A 63-year-old man was on dialysis for 9 years due to chronic GN, until he was first transplanted in 1984. After one year, a cellular rejection occurred, and a therapy with PDN, ATG and irradiation of the graft was initiated. Due to graft failure, a second transplantation followed in 1996. Sixteen years after the second transplantation, a CT scan showed a tumorous growth in both native kidneys. At this time eGFR was 66ml/min. Immunosuppression consisted of CyA, Aza and PDN. Grossly the kidneys were shrunken, but the perirenal fat was massively hypertrophic. Histology revealed renal replacement lipomatosis (RRL) with end-stage kidneys and massively increased perirenal fatty tissue with focal inflammation. Case 2: A 62-year-old man was first transplanted in 1983 because of a mesangioproliferative GN. One year later, cellular rejection occurred and was treated with PDN, ATG and graft irradiation. Due to graft failure, the patient was on dialysis for 2 years, until a second transplant was realized in 1986. Twenty-eight years after the first transplantation, a tumor in the first graft was detected by MRI. At this time eGFR was 25 ml/min. Immunosuppression consisted of CyA and PDN. Grossly the kidney was shrunken and surrounded by predominantly myxoid tissue. Histology showed RRL with end-stage kidney and increased perirenal fatty tissue with extensive myxoid areas and fibrosis.
Conclusion: RRL is a rare condition described in renal grafts and native kidneys. RRL has been associated with kidney stones, recurring infections and rejection episodes. In immunosuppressed patients, the differential diagnosis between RRL and renal tumors or PTLD is crucial and can avoid unnecessary nephrectomies.

P10

The Tolerogenic Effect Of Bcl-2 Inhibition in Allo-Transplantation is Mediated by Enrichment of Regulatory T Cells

S. S. Gabriel[1], N. Bon[2], A. K. Kraus[1], J. Chen[1], P. Bardwell[3], A. Bushell[4], T. Fehr[1], P. Cippà[1]
[1]Zurich, [2]Nantes/FR, [3]Worcester, MA/US, [4]Oxford/UK

Purpose: Inhibition of anti-apoptotic Bcl-2 family members by the BH3-mimetic ABT-737 suppresses allogeneic immune responses and promotes the induction of immunological tolerance in combination with costimulation blockade. In this study, we investigated the contribution of classical CD4+CD25+FoxP3+ regulatory T cells (Tregs) to the tolerogenic effect of ABT-737.
Methods: FoxP3-GFP transgenic mice were used to assess the effect of ABT-737 on natural and induced Tregs *in vitro* and *in vivo*. Furthermore, we evaluated the contribution of Tregs to tolerance induction in a non-myelosuppressive, mixed chimerism protocol based on Bcl-2 inhibition.
Results: Natural and induced Tregs were 10 to 100x more resistant to Bcl-2 inhibition compared to naïve T cells, leading to a relative Treg enrichment after exposure to ABT-737 *in vitro* and *in vivo*. This effect was not inhibited by cyclosporine A, indicating that this resistance was not mediated by the calcineurin-NFAT pathway as previously described in activated T cells. *In vivo*, Tregs enrichment by ABT-737 potentiated the effect of an established induction protocol which includes donor specific transfusion and costimulation blockade, leading to long-term survival of fully MHC-mismatched skin grafts without maintenance of immunosuppression. Furthermore, we demonstrated that the favorable effect of ABT-737 to induce mixed chimerism and tolerance was mediated by Tregs, since tolerance was lost when either CD4-, CD25- or GITR- expressing cells were depleted during the induction phase.
Conclusion: Tregs enrichment can easily be achieved *in vivo* by exploiting the relative resistance of Tregs to Bcl-2 inhibition. In future, therapeutic protocols for immunosuppression or tolerance induction based on Tregs could take advantage of this fact.

P11

Strength and Limitations of Regulatory T Cells for Immunotherapy in Transplantation

L. Govender, J.-C. Wyss, M. Pascual, D. Golshayan
Lausanne

Purpose: In many experimental models, CD4+Foxp3+ regulatory T cells have been identified as key players in promoting peripheral transplantation (Tx) tolerance. We have been focusing on therapies based on natural Treg (nTreg) that can control effector T cells (Teff) and prevent graft rejection. The use of nTreg in therapeutic protocols for solid organ Tx is however limited by low numbers and polyclonality in a normal individual. Moreover, although we previously described robust protocols to generate and expand antigen-specific nTreg in vitro, the process requires selection of highly pure nTreg and cumbersome ex-vivo manipulations, rendering this strategy not easily applicable in clinical Tx.
Methods: In this study, we expanded Treg directly in vivo and determined their efficacy and stability in promoting donor-specific tolerance in a murine skin Tx model.
Results: Our data suggest that IL-2-based therapies lead to a significant increase of Treg in vivo. The expanded Treg suppressed

Teff proliferation and allowed prolonged survival of MHC-mismatched grafts in wild-type non-lymphopenic recipients. The expanded Treg alone were however not sufficient to induce tolerance in stringent conditions. The combination with rapamycine or costimulation blockade, given at the time of Tx, modified the alloreative T cell pool by proportionnally increasing Treg thus promoting long-term survival of grafts. In contrast, pro-inflammatory stimuli hindered the expansion of Treg and resulted in an increase in the frequency of Th1 and Th17 cells.
Conclusion: We propose an efficient method for expanding functional Treg in vivo, thereby favorably shifting the pool of alloreactive T cells towards regulation in response to an allograft. However, we also highlight potential limitations such as concomitant inflammatory events.

P13

Luminex Prozone Effect Demonstrated in Vitro and in Vivo: Clinical Implications

S.-R. Wassmer, M. Bonani, T. Fehr
[1]Zurich

Purpose: The prozone effect leads to false low antibody measurement. It occurs when excess of antibody is unable to bind to receptor sites. It can be unmasked by sample dilution. A prozone effect in solid phase anti-HLA antibody testing has rarely been described.
Methods: We present two cases, where a Donor Specific Antibody (DSA) was missed due to prozone effect and led to antibody-mediated rejection (AMR). We then reviewed our kidney waiting list for the frequency of prozone effect.
Results: A 59 y old patient with ADPKD received a 2ndkidney transplant. Because of DSA he received ATG induction and immunosupression with TAC, MMF and PDN. After 13 days AMR occured. Immunoadsorption (IADS) reduced all DSA, but a novel DSA to DQ7 paradoxically increased. Repeat biopsies showed persistent AMR and resolved only after bortezomib therapy. Because of paradoxical MFI increase of DSA DQ7 after IADS, the prozone effect was considered and retrospectively confirmed by serum dilution.
A 22 y old patient with primary FSGS required a 2ndkidney transplant. Because of low titer DSA, he received immunosupression as above. After 8 months AMR occurred. Despite IADS, a novel DSA to DQ5 appeared which increased from 2200 to 13000 MFI after treatment. The prozone effect was confirmed. Analysis of the kidney waiting list revealed that among 261 patients 86 were HLA-sensitized (76 for class I, 48 for class II, 38 for both). Among those 25 had a positive CDC lymphoscreen assay on class I. Sofar dilution has been done in 16 Serum. In 7/16 (44%) patients we found a prozone effect. Further analyses is ongoing.
Conclusion: The prozone effect is common in highly sensitized patients tested by Luminex technology. Failure to detect it can result in severe AMR. We show for the first time the prozone effect in vivo.

Poster Presentations
Case Reports: Clinical Nephrology and Hypertension

P14

Urinary Neutrophil Gelatinase-Associated Lipocalin Does not Predict the Occurence of Contrast-Induced Acute Kidney Injury in Patients Undergoing Coronarography

T. N. U. Perrin, E. Descombes, S. Gobet, J.-L. Magnin, M. Gachet, O. M. Hemett, D. Hayoz, V. Stolt, G. Baeriswyl, J.-C. Stauffer, J.-J. Goy, M. Togni, S. Cook
Freiburg

Purpose: Diagnosis of contrast-induced acute kidney injury (CI-AKI) relies on a late marker, namely serum creatinine (SCr). Recently, urinary Neutrophil Gelatinase-Associated Lipocalin (uNGAL) has been used for early detection of AKI in the emergency department, after cardiopulmonary bypass or following contrast medium (CM) administration. The aim of this study was to evaluate the usefulness of uNGAL as an early detector of CI-AKI in a large cohort of patients undergoing percutaneous coronary procedures (PCP), and whether or not uNGAL correlates with the volume of CM used.
Methods: We enrolled 244 consecutive patients undergoing PCP with Iomeprolum at our institution. CI-AKI was defined as a ≥25% increase in SCr from baseline when measured 2 to 4 days after PCP. Urinary NGAL was measured at its peak (4–6 hours after PCP) with the Abbott ARCHITECT assay. Results are given as median [interquartile range].
Results: Among the 244 patients (age 66.6 [59.5–74.7] years, 70% male), 149 (61%) underwent a diagnostic PCP and 95 (39%) a therapeutic PCP, with a median CM volume of 123 [88–168] ml per procedure. Twenty-five (10%) patients developed CI-AKI. In our cohort, there was no significant difference in uNGAL levels between patients with and without CI-AKI, and patients developing CI-AKI tended even to have comparatively lower uNGAL levels (6.4 [3.4–10.4] versus 8.2 [4.1–15.1] ng/ml; p = 0.20). Also, we found no significant correlation between CM volume used during the procedure and uNGAL levels.
Conclusion: In our large cohort of patients mainly at low-risk for contrast-induced nephropathy, the incidence of CI-AKI was 10%. Urinary NGAL measured 4-6 hours after the coronarography did not predict renal toxicity, and did not correlate with the volume of contrast medium used during the procedure.

P15

Case Report: Plasma Exchange in Lupus-Like Syndrome With Very High MPO-ANCA

S. Kalbermatter[1], T. Öttl[1], K. König[1], I. Heijnen[2], D. Kiss[1]
[1]Liestal, [2]Basel

Purpose: A 43- year-old woman with a history of fatigue and swollen lymph nodes for three month, developed anemia and underwent a biopsy of a lymph node which revealed only a reactive lymphadenopathy. Due to increased serum creatinine and proteinuria in the nephrotic range she was transferred to our hospital. Pulse rate was 68 bpm and RR 117/77. C-reactive protein (CRP) was not elevated, S-Cr 174 µmol/l, albumine 27 g/l, hemoglobin 102 g/l, erythrocyte sedimentation rate 81 mm/h.
Methods: Renal biopsy revealed a diffuse segmental accentuated proliferative immune complex glomerulonephritis with crescents in four of 19 glomerula. Electron microscopy showed tubuloreticular structures within the endothelial cells. Myeloperoxidase Antineutrophil Cytoplasmic Antibodies (MPO-ANCA) were elevated to 3622U/ml (<5), anti-nuclear antibodies (ANA) titer 1:80, Anti-SS-A/Ro52 22U/ml (<10), Anti-SS-A/Ro60 >240 U/ml, complement C3c 0.2 g/L (0.8–1.8), C4 0.01 g/L (0.1-0.4); CH50 14U Eq/mL (70–180). The vasculitis was not to classify, because there was a typical clinical presentation and compatilbe histopathologic findings for a systemic lupus, but an atypical presentation of the antibodies. We started a treatment with pulses of intravenous cyclophosphamide and oral methylprednisolone (1 mg per kg) without any improvement of the kidney function or the proteinuria. To eliminate the MPO-ANCA suspected to play a major role in the pathogenesis we performed five high volume (5000 ml) plasma exchanges (PE). After the PE S-Cr decreased from 324 to 237 µmol/l and MPO-ANCA from 3622 to 114 U/ml.
Results: To our knowledge this is the first case with a lupus like syndrome and extremely high MPO-ANCA.
Conclusion: PE may improve outcome of lupus like syndrome with very high levels of MPO-ANCA.

P16

Associations of Diuretic Medication and Electrolyte Disorders on Osteoporotic Fractures: A Cross-Sectional Analysis of Elderly Patients Admitted to the Emergency Department

S. Arampatzis[1], G.-C. Funk[2], C. Schwarz[3], M. Mohaupt[1], H. Zimmermann[1], A. K. Exadaktylos[1], G. Lindner[1]
[1]Berne, [2]Vienna/AT, [3]Graz/AT

Purpose: Although hyponatremia is a well-recognized complication of treatment with diuretics and recently identified as a novel cause of osteoporosis, the impact of diuretic associated electrolyte disorders on osteoporotic fractures (OF) have rarely been studied in emergency department (ED) patients.
Methods: In this retrospective case series at Inselspital we identify 10823 adult outpatients (≥50 years) with a serum sodium measurement which were admitted between January 1, 2009 and December 31, 2010 at the ED.
Results: After exclusion of 573 patients with non-OF we identified 480 (5%) out of 10823 patients , with 547 OF. The OF group was characterized by higher mean age at presentation, smaller proportion of male patients, higher hospitalisation rates and longer hospitalization stay compared to controls (N = 9.769). The use of any diuretic agent (p <0.0001) and in particular loop, potassium sparing and amilorid (p values 0.02, 0.02 and <0.01 respectively) was significantly more common among OF patients. The prevalence of hyponatremia increased with the number of diuretics taken by the patients (p <0.0001). The use of SSRIs and antiepileptic drugs between both

groups was similar. In the multivariable analysis, advanced age (OR 1.04, p <0.0001), the presence of hyponatraemia (OR 1.3, p = 0.011) higher serum creatinin (OR 1.53, p = 0.0001), furosemide use alone (OR 1.40, p = 0.01) or diuretic co-treatment with amilorid (OR 2.22, p = 0.02) were all associated with a higher risk for OF.
Conclusion: The use of loop diuretics or diuretic co-treatment with amilorid was associated with an increased risk for osteoporotic fractures in elderly ambulatory patients. Moreover this condition could possibly be prevented by simply serum sodium monitoring.

P17

Acute Kidney Injury (AKI) in Cirrhotic Patients: Utility of New Biomarkers and Renal Resistive Indexes?

B. Ponte, L. Spahr, G. Berra, V. Poffet, N. Garin, P.-Y. Martin
Geneva

Purpose: AKI is frequent in cirrhotic patients. Pre-renal, acute tubular necrosis (ATN) or hepatorenal syndrome (HRS) are difficult to differentiate with plasma creatinine. We study new biomarkers and renal artery resistive indexes (RI) to evaluate their added value for better differentiate AKI etiologies.
Methods: We aim to include prospectively 100 adults with cirrhosis and ascitis. Exclusions' criteria are multifocal hepatocellular carcinoma, acute gastro-intestinal hemorrhage, severe CKD (eGFR <15 ml/min/1.73 m^2 or dialysis) and renal or hepatic transplant. AKI diagnosis is made according to AKIN criteria. Urine and blood samples are collected as soon as possible from the admission to analyze creatinine, cystatin c, NGAL and KIM1. A renal Doppler ultrasound is performed within the next days. We compare characteristics and biomarkers levels between AKI groups and use ROC curves to detect the best predictor.
Results: We include 77 patients in this intermediate analysis: 66.2% men, 58.3 ± 10.2 years old. AKI occurrs in 50.6% cases: 76.9% have pre-renal injury, 15.4% ATN and 7.7% HRS. Height patients are admitted to intensive care unit (ICU) and 6 died. Age, diabetes, sexe, infection, ICU admission or death are similar in the 3 etiological groups. RI is similarly high in all groups (p = 0.16). Patients with CKD have higher rates of ATN (p = 0.01). In univariate analyses new biomarkers (NGAl, KIM1, cystatine C) and creatinine are useful to differenciate ATN from pre-renal bu not from HRS. In the ROC analysis, urinary NGAL has the best AUC for ATN diagnosis [AUC 0.93 (0.78–100)].
Conclusion: New biomarkers can help in differentiating ATN from pre-renal AKI but not from HRS in cirrhotic patients with ascitis. RI has no added value in the differencial diagnosis. Urinary NGAL seems to be the best predictor to diagnose ATN.

P18

Determinants of Renal Artery Resistive Indexes in the Swiss Kidney Project on Genes in Hypertension (SKIPOGH)

B. Ponte[1], M. Pruijm[2], D. Ackermann[3], P. Vuistinier[2], U. Eisenberger[3], M. Mohaupt[3], B. Vogt[2], F. Paccaud[2], M. Burnier[2], P.-Y. Martin[1], M. Bochud[2]
[1]Geneva, [2]Lausanne, [3]Berne

Purpose: Recent evidence suggests that the renal resistance index (RI), defined as the percentage reduction of arterial end-diastolic flow as compared with systolic flow, is correlated with arterial stiffness and predicts cardiovascular events. We analyzed the determinants of RI in the general adult population.
Methods: We randomly selected families from the general population in Bern, Geneva and Lausanne. We measured anthropometric parameters, cardiovascular risk factors, blood pressure, 24 hours urine and performed a renal Doppler ultrasound. RI was assessed in the segmental arteries of superior, middle and inferior poles of each kidney according to a standardized protocol. Generalized estimating equations were used to identify determinants of RI adjusting for pulse rate, center and other covariates, taking familial correlations into account.
Results: We analyzed 282 men and 307 women aged 46.8 ± 17.4 and 48.9 ± 16.6 years respectively. Mean RI value of both kidneys was 0.63 ± 0.06 for men and 0.65 ± 0.05 for women (p <0.001). In multivariable regression analysis adjusted for confounders, age, diabetes, female sex, hypertension and SBP were significantly associated with higher RI. Urinary sodium excretion was also significantly associated with higher RI (coefficient per 100 mmol 0.01, SE 0.003; p <0.01) while urinary potassium (coefficient per 50 mmol –0.01, SE 0.005; p = 0.02) and urea excretion (coefficient per 100 mmol –0.006, SE 0.002; p = 0.01) were associated with lower RI.
Conclusion: The associations of RI with urinary sodium, potassium and urea excretion suggest that diet plays a role intra-renal arterial compliance and extrarenal resistance. These results are in line with previously described vasoconstrictive effects of salt intake and vasodilatatory effects of potassium and protein intake on renal arteries.

P19

No Correlation of Initial Antipla2r Positivity With Long Term Outcome Of Renal Function in Idiopathic Membranous Nephropathy

F. Burkhalter[1], H. Hopfer[1], E. Hoxha[2], M. J. Mihatsch[1]
[1]Basel, [2]Hamburg/DE

Purpose: Background: Recent findings in membranous nephropathy (MN) suggest that in most patients with idiopathic MN the underlying cause is a anti-phospholipase A2 receptor (antiPLA2R) antibody. In addition it was shown that antiPLA2R titer correlates with disease activity. Whether there is a correlation with initial antiPLA2R positivity and longterm outcome of renal function is unclear.
Objectives: Evaluation of the longterm outcome of patients with iMN in correspondance to their initial antiPLA2R status.
Methods: This is a single centre retrospective observational study in patients with biopsy proven membranous glomerulonephritis from 1992 until 2007. Patient were selected by avaiability of serum sample from the day of renal biopsy (n = 38). In 19 patients with iMN, follow up data were available. In 13 patients follow-up measurement of antiPLA2R were performed.

Results: biopsy proven MN		n = 38	
antiPLA2R	positiv	negativ	
	n = 18	n = 20	
secondary cause of MN		n = 12	
idiopathic MN	*n = 18*	*n = 8*	
(time of biopsy)			
median GFR MDRD(ml/min)	88.3 (23–153)	99.1 (34–134)	p = 0.91
median proteinuria (g/d)	6.5 (3.5–17)	7.7 (2.2–10)	p = 0.82
Last follow-up	*n = 14*	*n = 5*	
Immunsuppressive therapy	n = 6	n = 2	
median follow up (years)	9.5 (5.2–19.4)	12.8 (6.7–18.2)	p = 0.92
median GFR MDRD(ml/min)	91.7 (25.6–174)	111.5 (87–126.7)	p = 0.31
median proteinuria (g/d)	1.2 (0.01–2.9)	0.01	p = 0.038
ESRD	n = 1	n = 2	p = 0.155
anti-PLA2R follow up	*n = 10*	*n = 3*	
negativ	70%	100%	
positiv	30%	0	

Conclusion: There is no correlation of initial PLA2R positivity with the longterm renal function in this sample of patients with iMN with an overall median follow up of 10.9 years.

P20

Determinants and Heritability Of Kidney Length in a Family-Based Population Study

M. Pruijm[1], B. Ponte[2], D. Ackermann[3], P. Vuistiner[1], G. Ehret[2], I. Guessous[2], U. Eisenberger[3], F. Paccaud[1], P.-Y. Martin[2], M. Mohaupt[3], F. J. Frey[3], M. Burnier[1], M. Bochud[1]
[1]Lausanne, [2]Geneva, [3]Berne

Purpose: Kidney length is an important parameter in renal clinical decision making, yet data from population-based studies are sparse, and the heritability of kidney size is unknown. We assessed the heritability and the determinants of kidney length in a family-based population study.
Methods: The SKIPOGH study (Swiss Kidney Project on Genes in Hypertension) is a cross-sectional survey exploring the role of kidney hemodynamics and genes in blood pressure regulation and hypertension. Anthropometric parameters and renal ultrasound measurements were obtained in subjects chosen at random from the general population and at least one first-degree relative of each selected subject. The ASSOC program in SAGE (Statistical Analysis in Genetic Epidemiology) was used to estimate the age, sex, weight and height, eGFR (CKD-EPI), and center-adjusted narrow sense heritability.
Results: In total, 793 participants from 205 nuclear families were included. Mean (±SD) kidney length was 11.4 ± 0.8 cm in men (n =

374, age 47 ± 18 years, BMI 26.2 ± 4 kg/m², eGFR 98 ± 18 ml/min/1.73 m²), and 10.7 ± 0.8 cm in women (age 48 ± 17 y, BMI 24.5 ± 5, eGFR 95 ± 17). There was no significant difference in length between the right and left kidney. In multilevel adjusted linear regression analysis, body height, weight, and eGFR were positively associated with kidney length, whereas gender, diabetes, and hypertension were not. There was a quadratic association between age and kidney length. The heritability (h^2) of kidney length, adjusted for confounders, was 52 ± 8%, p <0.001.
Conclusion: This study suggests that kidney length is an inherited trait, independently of other important determinants such as age, estimated kidney function, body height and weight.

P21

Reference Values of Kidney Length According to Body Height in the Swiss Population

M. Pruijm[1], B. Ponte[2], D. Ackermann[3], P. Vuistiner[1], G. Ehret[2], I. Guessous[2], U. Eisenberger[3], F. J. Frey[3], M. Mohaupt[3], F. Paccaud[1], P.-Y. Martin[2], M. Burnier[1], M. Bochud[1]
[1]Lausanne, [2]Geneva, [3]Berne

Purpose: In clinical practice nephrologists are often confronted with the question which kidney size is normal for a given body height, yet reference values from the general population are sparse. In this study, we assessed kidney length in healthy Swiss adults.
Methods: In the ongoing SKIPOGH study (Swiss Kidney Project on Genes in Hypertension), nuclear families were randomly selected from the general population in Lausanne, Geneva and Bern, and renal gray-scale ultrasounds were performed according to a standardized protocol. For the purpose of this analysis, participants with renal structural abnormalities, obesity (BMI ≥30 kg/m²), diabetes, chronic kidney disease (eGFRckd-epi <60 ml/min/1.73 m²) or insufficient ultrasound quality were excluded.
Results: Of the 854 participants, 570 were included in this analysis. In the 269 men and 301 women, mean(SD) age was 44(17) and 47(16) years, BMI 24.7(3) and 22.9(3) kg/m², eGFR 102 (16) and 96 (15) mL/min/1.73 m², respectively. Median kidney length (25–75 percentile) according to tertile of body height was 109 (106–116), 112 (108–118) and 118 (112–123) mm in men with median(range) body height of respectively 171 (157–174), 177 (174–180) and 183 (180–201) cm, versus 103 (100–109), 107 (102–110), 110 (106–115) mm in women with body height 159 (148–162),165 (162–168), 171 (168–182) cm. In multilevel adjusted linear regression, body height was the strongest determinant of kidney length (adjusted regression coefficient β per cm body height [95% CI]: 0.32 [0.23–0.41], p <0.001).
Conclusion: Kidney length correlates strongly with body height. The reference values reported in this study are intended to help the physician determine whether kidneys are small, normal, or large for a given body height.

P22

Methadone and Glomerulonephritis

C. Jaeger, H. Heule
1Altstätten

Purpose: Methadone is mainly used in substitution programs for intravenous heroin abusers. We remarked in our district an increase of glomerulonephritis (GN) in persons who use methadone.
Methods: We studied in a retrospective way 6 patients on methadone, who were referred to our renal centre. All of them had a marked increase of serumcreatinine, 3/6 with nephrotic syndrome.
None of them had an active hepatitis, whereas 4/6 were seropositive for anti-HCV and 2/6 for anti-HBc. We have done renal biopsy in all of these patients.
Results: 5/6 were practising intravenous methadone abuse. One of them had a right ventricular endocarditis resulting in a necrotizing endocapillary GN and is now on dialysis. The other 4/6 were showing all a membranoproliferative GN. Two of them were treated immunosuppressive. One patient on prednisone monotherapy had a remission in proteinuria but GFR worsened from 82 to 41 ml/min. The other is effectively treated with mycophenolate and corticoids. His GFR is stable, proteinuria fall from 11 to 0.3 g/d. One young lady, who achieved stopping intravenous use improved GFR from 46 to 106 ml/min, proteinuria dropped from 0.5 to 0.06 g/d. One patient was lost to follow up. 1/6 without intravenous drug abuse had mesangioproliferative GN with crescents and was treated with cyclophosphamide, corticoids and azathioprine in remission with actually normal serum creatinine.
Conclusion: The cause of relationship between methadone and glomerulonephritis is not known. 5/6 of our patients admitted a contaminated intravenous abuse of methadone which is designed for orally use. Think of intravenous methadone use when finding an "idiopathic" membranoproliferative GN.

P23

Renal Tissue Oxygenation in Patients With Chronic Kidney Disease and Healthy Controls as Measured With BOLD-MRI: An Interim Analysis

M. Pruijm[1], L. Hofmann[1], C. Zweiacker[1], E. Zürcher[1], M. Piskunowicz[2], M.-E. Muller[1], M. Stuber[1], B. Vogt[1], M. Burnier[1]
[1]Lausanne, [2]Gdansk/PL

Purpose: Animal studies have suggested that renal tissue hypoxia plays an important role in chronic kidney disease, yet data in humans are sparse. We are actually assessing cortical and medullary oxygenation in patients with chronic kidney disease (CKD) and healthy normotensive controls using blood oxygenation level dependent magnetic resonance imaging (BOLD-MRI).
Methods: Patients with CKD stage I-V (all causes except polycystic kidney disease) undergo BOLD-MRI under standardized hydration conditions. Four coronal slices are selected in both kidneys, and a multi gradient echo sequence is used to acquire T2* weighted images. The mean R2* values (= 1/T2*) are calculated, a low R2* indicating a high tissue oxygenation. Kidney function is estimated using the MDRD formula and measured by 24h urinary creatinine clearance (ucreat).
Results: So far, 62 CKD patients and 32 controls have been recruited; mean (SD) age is 55 (15) and 43 (14)y, eGFR 59 (29) and 95 (16) ml/min/1.73 m², ucreat 79 (54) and 121 (24) ml/min, and 33% vs 58% are female. Mean cortical R2* (18.6 ± 4.7 vs 17.2 ± 2.2 sec-1, p = 0.2) and medullary R2*(29.8 ± 1.6 vs 29.9 ± 2.4 sec-1, p = 0.8) are not significantly different between CKD and healthy participants. In multivariate logistic regression, adjusted for age, sex, hemoglobin, sodium intake and BMI, CKD is not associated with cortical or medullary R2*. In adjusted multivariable linear regression analysis, no association is present between eGFR or ucreat and cortical R2* (β –0.001, p = 0.56 vs -0.03, p = 0.06) or medullary R2* levels (β 0.001, p = 0.93 vs –0.001, p = 0.92).
Conclusion: In this interim analysis, R2* as a measure of kidney oxygenation is not altered in CKD patients, nor associated with kidney function, suggesting that kidney oxygenation is tightly maintained over a broad range of kidney damage.

P24

Natriuretic Peptides for Early Prediction of Acute Kidney Injury in Community Acquired Pneumonia

A. Nowak[1], T. Breidthardt[2], S. Dejung[1], M. Christ-Crain[2], R. Bingisser[2], B. Drexler[2], C. Meune[2], D. Marono[2], T. Mosimann[2], B. Müller[3], C. Mueller[2]
[1]Zurich, [2]Basel, [3]Aarau

Purpose: Background: Community-acquired pneumonia (CAP) is common and associated with a considerable risk of acute kidney injury (AKI).
Methods: We prospectively enrolled 341 patients presenting to the emergency department with CAP (mean age 72, male 61%). Blinded measurements of three natriuretic peptides (NT-proBNP, MR-proANP and BNP) were performed upon presentation. The primary endpoint was the accuracy of the natriuretic peptides to predict AKI within 48 hours, the median follow-up 942 days.
Results: AKI occurred in 21 patients (7.6%) within the first 48 hours. NPs and creatinine were significantly higher in AKI compared with patients without AKI (NT-proBNP 9517 [2042–26792] vs 1177 [280–4167] pg/ml; MR-proANP 641 [196–1075] vs 182 [99–352]pmol/l; BNP 592 [230–1630] vs 160 [64–463] pg/ml; creatinine 166 [131–289] versus 100 [78–134] µmol/L, P <0.001 for each). Predictive accuracy, as quantified by the area under the receiver operating characteristics curve, was moderate to high: NT-proBNP 0.79 (95%CI 0.70–0.88), MR-proANP 0.78 (95%CI 0.67–0.88), BNP 0.74 (95%CI 0.63–0.85), creatinine 0.77 (95%CI 0.66–0.88). In multivariate logistic regression analysis, NPs remained the only independent AKI predictors: table 1. NPs and the Pneumonia Severity Index were more closely associated with short- and long-term mortality than traditional AKI predictors (serum creatinine, preexisting chronic kidney disease), as assessed in multivariate cox regression analysis
Conclusion: NP levels at presentation can be useful predictors for early AKI in patients with community acquired pneumonia and seem to be closely associated with mortality.

Table 1
Prediction of acute kidney injury development.

Multivariate logistic regression	OR	p-value
NT-proBNP	1.01 (1.00–1.01)	0.009
Creatinine	1.00 (1.00–1.01)	0.07
CKD	1.92 (0.65–5.66)	0.24
PSI	1.00 (0.99–1.02)	0.72

P25

Evaluation of a Novel Diagnostic and Treatment Algorithm for Hyponatremia

A. Bock[1], A. Huber[1], C. Blum[1], C. Nickel[2], P. Schuetz[1], M. Bally[1], B. Arici[2], I. Suter-Widmer[2], B. Winzeler[2], N. Nigro[2], B. Müller[1], M. Christ-Crain[2]
[1]Aarau, [2]Basel

Purpose: Correctly diagnosing of the cause of hyponatremia is a frequent challenge in the emergency room setting, because appropriate therapy (water restriction; NaCl infusion) depends on the cause. Existing algorithms rely heavily on a correct assessment of extracellular fluid volume. The present new algorithm was designed with the idea to use the kidney as gauge for volemia and ADH state. The present prospective oberservational study had the goal to evaluate the real-life performance of the algorithm and to calibrate it against measurements of the ADH precursor peptide copeptin.
Methods: Patients presenting with severe hyponatremia (<125 mmol/l) to the emergency room of the two participating hospitals were prospectively attributed to diagnostic groups based on urine osmolarity, the fractional excretions of urea and uric acid as well as some evident signs of volume overload (Edema/Ascites/Orthopnea) as follows:
Algorithm based diagnoses were contrasted with final clinical diagnoses. The present data represent an interim analysis.
Results: Out of 37 evaluable patients of one hospital, the algorithm correctly identified 5/5 patients with polydipsia, 13/16 patients with general hypovolemia (3/16 erroneously as "SIADH"), 5/6 patients with intravascular hypovolemia and 8/8 patients with SIADH. The final diagnosis was unclear in 2.
Conclusion: The new algorithm performed well. "SIADH" was the most common misclassification.

U-Osm	FE-Urea/ Uric Acid	Overload signs	Creatinine	Diagnostic Group	Example	Therapy
<200	–	–	–	Exogenous Water Excess	1° polydipsia	H_2O restriction
>200	"prerenal"	no	–	General Hypovolemia	Diarrhea, vomiting, diuretics	0.9% NaCl
>200	"prerenal"	yes	–	Intravasc Hypovolemia	Congestive heart failure, liver cirrhosis	H_2O restriction
>200	"renal"	–	increased	Renal failure		H_2O restriction
>200	"renal"	–	normal	SIADH		H_2O restriction

P26

Renal Phosphate Handling in Gitelman Syndrome – The Results of a Case-Control Study

M. G. Bianchetti[1], C. Viganò[2], C. Amoruso[2], S. A. G. Lava[1], A. Bettinelli[2]
[1]Bellinzona, [2]Merate-Lecco/IT

Purpose: Patients with Gitelman syndrome, a hereditary salt-wasting tubulopathy, have loss-of-function mutations in the SLC12A3 gene coding for the thiazide-sensitive sodium chloride co-transporter in the distal convoluted tubule. Since the bulk of filtered phosphate is reabsorbed in the proximal tubule, renal phosphate wasting is considered exceptional in Gitelman syndrome. We noticed a tendency towards low inorganic phosphate levels in some of our Gitelman patients which led us to investigate the renal handling of this ion in the context of a study.
Methods: We investigated the renal handling of inorganic phosphate in 12 unselected patients affected with Gitelman syndrome (5 females and 7 males, aged 6.0–18 years, median age 12 years) and in 12 healthy subjects matched for gender and age. The diagnosis of Gitelman syndrome among the patients had been made clinically and confirmed by molecular biology studies.
Results: The biochemical hallmarks of Gitelman syndrome, namely hypochloremia, hypokalemia, hypomagnesemia, increased urinary excretion of sodium, chloride, potassium and magnesium and reduced urinary excretion of calcium, were noted in the 12 patients. In addition, both the plasma inorganic phosphate concentration (1.28 [1.12–1.36] versus 1.61 [1.51–1.66)] mmol/L; median and interquartile range) and the maximal tubular reabsorption of inorganic phosphate (1.08 [0.99–1.22] versus 1.41 [1.38–1.47] mmol/L) were significantly lower (P <0.001) in Gitelman patients than in control subjects. Circulating levels of 25-hydroxyvitamin D, intact parathyroid hormone and osteocalcin were similar in patients and controls.
Conclusion: The present case-control study discloses a hitherto unrecognized tendency towards renal phosphate wasting with mild to moderate hypophosphatemia in Gitelman syndrome.

P27

Assessment of Adult Formulas for Glomerular Filtration Rate Estimation in Children

H. Chehade[1], E. Girardin[2], K. Iglesias[1], P. Ramseyer[1], P. Frey[1], D. Bardy[1], D. Mosig[1], F. Cachat[1]
[1]Lausanne, [2]Geneva

Purpose: Estimated glomerular filtration rate (eGFR) is animportant diagnostic instrument in clinical practice. TheNKF–KDOQI guidelines do not recommend using formulasdeveloped for adults to estimate GFR in children; however, studiesconfirming these recommendations are scarce. The aim of ourstudy was to evaluate the accuracy of the new Chronic KidneyDisease Epidemiology Collaboration (CKD-EPI) formula, theModification of Diet in Renal Disease (MDRD) formula, and the Cockcroft–Gault formula in children with various stages of chronic kidney disease (CKD).
Methods: A total of 550 inulin clearance (iGFR) measurements for 391 children were analyzed. The cohort was divided into three groups: group 1, with iGFR >90 ml/min/1.73 m^2; group 2, with iGFR between 60 and 90 ml/min/1.73 m^2; group 3, with iGFR of <60 ml/min/1.73 m^2.
Results: All formulas overestimate iGFR with a significant bias (p <0.001), present poor accuracies, and have poor Spearman correlations. For an accuracy of 10%, only 11, 6, and 27% of the eGFRs are accurate when using the MDRD, CKD-EPI, and Cockcroft–Gault formulas, respectively. For an accuracy of 30%, these formulas do not reach the NKF–KDOQI guidelines for validation, with only 25, 20, and 70% of the eGFRs, respectively, being accurate.
Conclusion: Based on our results, the performances of all of these formulas are unreliable for eGFR in children across all CKD stages and are associated with accuracies far below the KDOQI targets of ≥90% needed for validation. These formulas cannot be applied in children, therefore confirming the NKF–KDOQI recommendations.

P28

Renal Cysts and Diabetes Syndrome due to Mutation of the Hepatocyte Nuclear Factor-1-Beta Gene

K. König, S. Kalbermatter, I. Grendelmeier, D. Kiss
Liestal

Purpose: A 53-year old caucasian was referred to our outpatient clinic for the evaluation of chronic kidney failure. At the age of 18, the patient was diagnosed with diabetes and years later the diabetes became insulin dependent. Over the following years, kidney function impaired, without relevant proteinuria.
Methods: n/a
Results: On presentation the patient complaint about fatigue and weakness. Family history showed a brother with diabetes. Blood pressure was 142/73 mm Hg, pulse 114/min regular, BMI 19.4 kg/m^2. Labararoty studies showed a creatinine of 443 mmol/l with an estimated glomerular filtration rate of 13 ml/min/1.73 m^2 and urea of 27.1 mmol/l. Urinanalysis was significant for 2+ protein, 1+ glucose and a urine protein/creatinine ratio of 131 mg/mmol. Hemoglobin was 106 g/l. C-peptid <0.02 nmol/l with an HbA1c of 9.1%. i-PTH 251 pmol/l, calcium 2.55 mmol/l and phosphate 1.88 mmol/l. MRI was performed which revealed multiple cystic lesions in both kidneys with normal kidney size.
Conclusion: The combination of cystic kidney disease with early-onset diabetes best fits to the renal cyst and diabetes syndrome (RCAD) that was formerly called MODY type 5. The results of the genetic analysis confirmed mutation of the hepatocyte nuclear factor-1-beta gene (HNF1-beta). Affected patients can develop a variety of manifestations. These includes pancreatic atrophy which leads to diabetes and abnormal renal development with slowly progressive renal failure. Mutations in the HNF1-beta gene inhibit the expression of Pkhd1 which leads to cyst formation. Mutations of Pkhd1 gene are responsible for the autosomal recessive form of polycystic kidney disease.
We thank Prof. T. Fehr for the helpful comments.

P29

Varicella-Zoster-Virus Vasculopathy Presenting as a Stroke After Kidney Transplantation

I. Koneth, G. Kaegi, K. Boggian, I. Binet
St. Gallen

Purpose: VZV cerebral vasculopathy is a rare disorder.
Methods: We report a case in a kidney transplant recipient presenting as a stroke
Results: A 41-year old man received a preemptive deceased kidney transplantation. Induction consisted of antithymoglobulins, steroids, mycophenolate, tacrolimus and iv Ig. 3 weeks post-transplant the patient complained of fatigue and demonstrated a strange behaviour. Lab results were normal; ABPM showed no blood pressure dip, 24-hours-ECG and cerebral CT-scan were normal. 8 weeks after transplantation the patient was hospitalised because of headache, hemiparesis, dysarthria and complex oculomotor disorder. No fever,

no meningism, normal blood analysis. MRI showed acute ischemic infarction in the pons and subacute ischemias in the pedunculus cerebelli and corpus callosum. Because of the subacute aspect of most lesions, lysis was not performed. CSF showed mononuclear pleocytosis, high proteins and normal glucose. Intrathecal VZV PCR and anti-VZV – Ab were positive. The patient was seropositive for VZV pretranslant but did not present herpes zoster at any time. Prednisone was shortly increased, mycophenolate mofetil paused for 2 weeks and restarted at halfdose, iv acyclovir administered at a dose of 30 mg/kg/d then switched to oral valacyclovir 1 g tid. After 4 months neither VZV PCR in liquor nor new ischemias in MRI were detectable, prophylaxis with valacyclovir 500 bid was installed. 6 months after the acute illness a mild weakness of the left upper extremity and deficits in memory and attention persist.
Conclusion: VZV vasculopathy should be suspected in immunocompromised patients with multiple acute and subacute cerebral ischemias even in the absence of a characteristic zoster rash. Diagnosis requires a liquor analysis for viral DNA and VZV antibodies.

P30

PRES – A Diagnosis not Only for Neurologists: Two Case Reports in Renal Patients

K. Günther, C. Bucher, G. Kaegi, I. Binet
St. Gallen

Purpose: Posterior reversible encephalopathy syndrome (PRES) is characterized by acute onset of headache, nausea, seizures, altered consciousness and visual disturbance along with radiological findings mainly of symmetric white matter defects in the parietal and occipital lobes. Nephrotic syndrome, chronic and acute kidney disease, HUS, organ transplantation, immunosuppressive therapy, autoimmune diseases and electrolytes disturbances are predisposing factors. We report two cases of PRES in young patients with ESRD.
Methods: First patient is a 34-year old man on peritoneal dialysis due to diabetic nephropathy. He was hospitalized for chronic intermittant vomitting and chronically high blood pressure. After a blood pressure spike he developped an acute headache and sudden loss of vision with cortical blindness. The MRI showed bilateral symmetric cortical occipital defects. Hypokalemia (2.7 mmol/l), hypocalcemia (iCa 1.05 mmol/l) and metabolic alkalosis were present. The second patient is a 21 year old man on chronic hemodialysis due to IgA-nephropathy and under immunosuppression with steroids and cyclophosphamide. He presented with a first dyscognitive seizures followed by a bilateral clonic-tonic seizure. The MRI showed multiple bilateral T2-weighted hyperintense signal cortical and subcortical, temporoparietal and frontal. EEG demonstrated later on no seizure activity. CSF was unremarkable.
Results: After a closely monitored blood pressure lowering therapy and magnesium infusion in the ICU both patients experienced a complete recovery of their neurological symptoms.
Conclusion: Think about PRES in front of a renal patient with badly controlled hypertension and/or immunsuppressive therapy who presents with neurological findings such as severe headache, seizure or vision disturbances.

P31

A Wolf In Sheep's Clothing; The Course of Thin Basement Membrane Nephropathy (TBMN) on Basis of a Family Pedigree

I. Grendelmeier[1], H. Hopfer[2], D. Kiss[1]
[1]Liestal, 2Basel

Purpose: Thin basement membrane nephropathy (TBMN) is a common (up to 5–6% in gen. pop.) in general familial disorder (30–50% of cases with family history) with usual benign course and is therefore also called "benign familial hematuria". The only finding on renal biopsy is thinning of the glomerular basement membrane. The prognosis is excellent in most patients with true TBMN. However, slowly progressive renal impairment can occur and is often manifested on renal biopsy by focal segmental glomerulosclerosis (FSGS). New data suggest that this may be more common than previously suggested.
Methods: We report of a family with hematuria and autosomal dominant inheritance.
Results: 3 younger individuals had biopsy proven TBMN and different degrees of FSGS all of them with significant proteinuria (>2 g/l), microhematuria and normal kidney function at time of presentation. 6 first, second and third degree relatives had microhematuria in whom 4 of them developed renal insufficiency/renal failure at older age (>50). Alport syndrome was excluded clinically.
Conclusion: In conclusion TBMN is a common cause of familial hematuria and is caused by diffuse thinning of the glomerular basement membrane. Heterozygous defects in COL4A3 or COL4A4, the genes that encode for the alpha-3 and alpha-4-chains of collagen type IV, are responsible for this disorder in 40–50% of affected families. Those can be considered as "carriers" of autosomal recessive Alport syndrome. In most patients prognosis is excellent, however, some patients develop proteinuria. In these cases we recommend renal biopsy, close follow-up and treatment of those individuals who obviously exhibit progressive renal disease course.

P32

Flank Pain, Hypertension and Renal Infarcts in a Young Man: A Rare Manifestation of Sarcoidosis

L.-Y. Mani, B. Vogt, D. Golshayan
Lausanne

Purpose: A 34-year-old male was admitted to hospital with a few days history of abdominal and flank pain, headaches, malaise and visual blurring, without fever or urinary symptoms. He had suffered weight loss, asthenia and exertional dyspnea over the last 2 months. Physical examination was normal except for severe hypertension. Laboratory tests revealed elevation of serum creatinine at 127 umol/l and moderate systemic inflammation, urinalysis was normal.
Methods: The CT scan showed multiple thoraco-abdominal adenopathies, lung micronodules and peripheral hypodense cortical lesions in both kidneys. A mediastinoscopy with node biopsies was performed, with the histological diagnosis of granulomas.
Results: Our patient thus presented with a systemic granulomatous disease. He underwent further laboratory investigations including infectious and immune serologies,Tb-spot and B2-microglobuline. Following on the CT aspect of the kidneys, a selective abdominal angiography was performed, confirming the presence of multiple renal infarcts as well as revealing microanevrisms on renal as well as splenic arteries. Based on these findings, the diagnosis of sarcoidosis was made. High dose steroids were started followed by azathioprine together with anti-hypertensive drugs.
Conclusion: Sarcoidosis is an inflammatory multisystem disorder characterized by the presence of noncaseating granulomas. Virtually every organ can be affected, but about 90% of cases have lung involvement. Up to half of the patients are asymptomatic, identified upon incidental radiological findings. Clinically relevant renal disease seems to represent only an occasional problem in sarcoidosis, the main finding being granulomatous interstitial nephritis. Granulomatous angiitis and in particular renal vasculitis is a very rare manifestation of sarcoidosis.

P33

Caplan's Syndrome: Rarely Presenting as "Pulmo-Renal" Syndrome

A. Nowak, K. Göhner, C. D. Cohen
Zurich

Purpose: Presentation of a rare differential diagnosis of pulmo-renal syndrome.
Methods: History and admission findings. A 59-year-old man complied having dry cough for months and a recent sudden onset of asymmetric arthritis, myalgia as well as lack of appetite. He presented an occupational history of 12-year exposure to anorganic dust as uranium miner in German Democratic Republic followed by 21 years as heavy construction worker in Germany and in Switzerland. Laboratory work-up tested positive for microhematuria and anti-neutrophilic cytoplasmic antibodies (ANCA). Chest X-rays and CT scan showed bilaterally scattered nodules. Thoracoscopic wedge resection was performed, histopathological analysis revealed granuloma with central necrotic area containing black coal dust and silica depositions. The pulmonary opacities on X-ray and typical histology in the light of significant dust exposure allow the diagnosis of a Caplan's syndrome.
Results: Treatment and course. Symptoms improved rapidly under steroid therapy. A clear renal cell carcinoma was diagnosed as a cause for the persistent microhematuria.
Conclusion: Rheumatoid arthritis, pulmonary nodules and history of prolonged dust exposure are classical findings that define Caplan's syndrome. These patients present with different immunological phenomena – in our case ANCA positivity without vasculitis. Interestingly, renal cell carcinoma, causing microhematuria and leading to the "pulmo-renal" syndrome, is another health problem overrepresented in uranium mine workers.

P34

A Trial of Complement Inhibition in a Patient With Cryoglobulin-Induced Glomerulonephritis

P. Hirt-Minkowski[1], M. Trendelenburg[1], I. J. Gröschl[1], A. Fischer[2], I. Heijnen[1], J. A. Schifferli[1]
[1]Basel, [2]Lucerne

Purpose: Cryoglobulinemia induces an immune complex-mediated glomerulonephritis that is characterized by the presence of large immune deposits, including complement C3 and C5b-9, marked macrophage influx and mesangial cell proliferation. The precise role of complement in cryoglobulin-induced glomerulonephritis in humans

remains unclear, whereas in mice there has been evidence that complement activation might be a central factor favoring glomerular inflammation, particularly by the recruitment of neutrophils.
Methods: We report on an exceptional case of cryoglobulin-induced glomerulonephritis in a patient with mixed essential cryoglobulinemia type II. The clinical features included relapsing proteinuria and renal function impairment that were controlled by plasmapheresis. Complement was low in plasma and two renal biopsies at one-year interval showed prominent immunoglobulin and complement deposits, with unusual high numbers of neutrophils. In a one-patient clinical trial we tested whether the monoclonal anti-C5 antibody eculizumab would be sufficient to control renal function at the time of a relapse.
Results: Although during the initial weeks renal function was stabilized, slow increase in creatinine could not be controlled by this treatment, so that plasmapheresis was reinstituted.
Conclusion: This result suggests that despite evidence for a role of complement in enhancing renal damage in this patient, other inflammatory processes dominated.

P35

Neonatal Hemolytic Uremic Syndrome Due to Shiga-Toxin-Producing Escherichia Coli

B. Bucher[1], L. Kottanattu[1], S. Tschumi[1], A. Stritt[1], M. Steinmann[1], N. von Steiger[1], R. Stephan[2], H. Hächler[2], G. D. Simonetti[1]
[1]Berne, [2]Zurich

Purpose: Hemolytic uremic syndrome (HUS) is a leading cause of acute renal failure in childhood. The majority of cases are preceded by an episode of diarrhea mostly due to Shiga-toxin-producing Escherichia coli (STEC). Cobalamin C disorder, defective regulation of the alternative complement pathway and congenital ADAMTS13 deficiency are possible causes for atypical HUS in the neonatal period. STEC can also rarely lead to neonatal HUS.
Methods: Description of a case.
Results: A newborn male, presenting with biliary vomiting two days after birth without diarrhea, showed on day six of life a sudden increase of total bilirubin. Laboratory findings showed hemolytic anemia with fragmentocytes, thrombocytopenia and acute renal failure. 24 hours later he developed epileptic seizures with good response to antiepileptic therapy. Family history was negative for renal diseases and none of the parents had shown gastrointestinal symptoms. Since the newborn recovered quickly with normalization of all parameters within 48 hours and normal neurological condition, plasma exchange or the monoclonal antibody against terminal complement protein C5 were not considered. Testing for causes of atypical HUS remained negative. Fecal analysis of both the newborn and his mother disclosed STEC, indistinguishable by microarray analysis, and pulsed-field gel electrophoresis, and harboring stx2B. Shiga-toxin Stx2B is of low virulence, not normally causing HUS. We postulate that the mother is a healthy carrier, who transmitted the bacteria by fecal-oral route to the newborn during delivery. In a newborn's steril bowel this microorganism can exceedingly proliferate thus leading to HUS.
Conclusion: HUS due to STEC expressing a toxin type of even low virulence can occur immediately after birth by mother-to-child fecal-oral transmission.

P36

Pseudorenal Failure of Hospitalized Patient With Isopropyl Alcohol Ingestion

M. T. Tufail Hanel, T. Klima, A. Bock
Aarau

Purpose: Isopropyl is a common source of clinical intoxication. Acute renal failure due to Isopropyl ingestion is rare. We describe a patient, who ingested Isopropyl and presented with renal failure.
Methods: A 39 year old female with a history of alcohol abuse was hospitalized for acute alcohol intoxication. At admission her physical and neurological examination were unremarkable. When she was shifted to the normal ward, she drank 300 ml disinfectant containing 60% Isopropyl. The patient complained about headache and abdominal pain. The blood tests showed a rapid rise in creatinine from 48 to 208 µmol/l, eGFR 24 ml/min (MDRD). However, urea was 3.1 mmol/l and Cystatin C GFR >150 ml/min. The diuresis was preserved and blood pressure remained normal. The serum osmolality was 336 mosmol/Kg with a calculated osmolar gap of 40 mosmol/kg, without metabolic acidosis (HCO3: 24 mmol/l) and normal anion gap. There was slight ketonuria. After 36 hours the creatinine started to decline. The electrolytes and daily urinary output remained normal. One month after this case, we observed another isopropyl intoxication where a massive transent impairment of MDRD GFR <15 ml/min contrasted with only a minor impairment of Cystatin C GFR 35 ml/min. Isopropyl is a secondary alcohol and is completely absorbed following oral ingestion. It is metabolized by alcohol dehydrogenase to acetone, which interferes with colorimetric assay to determine creatinine. The half-life of Isopropyl is 2.5–8 hours.
Results: Isopropyl Intoxication can elevate creatinin by:
pt1:
– interference of acetone with the colorimetric assay
– Enzymatic assays and Cystatin C assays are not interfered by acetone
pt 1+2:
– Is there a creatinine secretion inhibition by acetone or isopropyl?
Conclusion: In Isopropyl-poisoning, Cystatin C should be used to verify renal function

---

P37

A Woman With an Unusual Composition of her Stone

M. Zobrist[1], H.-R. Räz[2]
[1]Wetzikon, [2]Baden-Dättwil

Purpose: A 28-year-old healthy caucasian women suffered from flank pain with macrohematuria. A computed tomography scan (CT) of the abdomen showed bilateral urolithiasis. ESWL followed by left-side ureterorenoscopy was undertaken. Analyses of the extracted stone revealed 2,8-dihydroxyadenine (2,8-DHA), what to our knowledge in Switzerland only was reported once [1]. Measurement of enzyme-activity in the erythrocyte lysate of our patient confirmed complete deficiency of the enzyme adenine-phosphoribosyl-transferase (APRT). Genetic analysis showed a homozygous single thyrosin insertion in intron 4 (IVS4+2 insT mutation) of the APRT-gene (chromosome 16q24). Our patient increased her fluid intake to dilute her urine to a density ≤1.010 g/cm^3, reduced her dietary purine intake and is taking allopurinol regularly. Under this treatment, she stayed stone-free. For follow up, the number of 2,8-DHA crystals/ul under this therapy will be determinded. The cause for hyperexcretion of 2,8-DHA into the urine is complete deficiency of APRT, a rare autosomal recessive disorder. Adenin, originating from purine metabolism, normally is transformed to 5'-AMP. In APRT deficiency, adenine is oxidized by xanthin oxydase to 2,8-DHA, which is highly insoluble and crystallizes in the urine. The disease manifests not only as recurrent urolithiasis at any age but also as crystalline nephropathy, that sometimes causes ESRD. Allopurinol, by inhibiting xanthin-oxydase, effectively reduces production and excretion of 2,8-DHA and prevents the formation of new stones. This case demonstrates the importance of stone analysis to prevent ESRD by a simple medication.
Reference
1. H. Simmonds: 2,8-Dihydroxyadenine lithiasis. Clin Chim Acta 1986;160:103–8.

P38

Chicken Pox in a Varicella Vaccinated Hemodialysis Patient

A. Kneubühl, T. Bregenzer
Lachen

Purpose: Varicella zoster Virus (VZV) primary infection may cause complications such as pneumonitis or encephalitis mainly in immunocompromized patients e.g. kidney transplant recipients. Varicella vaccination is recommended before kidney transplantation in seronegative patients.
Methods: We report the case of a 27 year old man vaccinated with two doses of VZV vaccine fifteen years ago. He had endstage kidney disease because of cystinosis and became kidney transplanted fourteen years ago. Immunosuppression consisted of Tacrolimus, Mycophenolate and Steroids. Tacrolimus was substituted by Ciclosporin when B-Cell Non Hodgkin Lymphoma was successfully treated 2003. Kidney transplant failure and start of dialysis followed 2010. In February 2012 his unvaccinated brother with no history of VZV infection before, fell ill with chicken pox. Two weeks later our patient presented with primary VZV infection – Vesiculo papular rash, fever, head- and musclepain. VZV-IgG-level was <0.60. (IndexImmun >0.89). While on therapy with Valaciclovir 500 mg twice daily for one week and because of side effects 500 mg once daily for an other week he fully recovered within three weeks.
Results: Chicken pox after kidney transplantation is rare because most of these patients are seropositive after primary infection in childhood or they are vaccinated. More often reactivation of VZV persistent latent infection in sensory ganglia may result in shingles. The seroconversionrate after vaccination measured by immunofluoreszence assay is 100% but results in only 88% protection rate.
Conclusion: Despite Varicella vaccination primary infection of VZV may occur. Immunosuppressive treatment and/or immunocompromized status due to chronic renal failure may reduce vaccination efficacy. Monitoring of VZV-IgG levels and revaccination should be discussed.

P39

Resetting of Kidney Renin-Angiotensin, Kallikrein-Kinin, and Catecholamine Systems After Unilateral Renal Denervation

J. Bohlender[1], F. Birkhäuser[2], J. Nussberger[3], E. Grouzman[3], G. Thalmann[2], H. Imboden[2]
[1]Freiburg, [2]Berne, [3]Lausanne

Purpose: Catheter-based renal denervation is an effective treatment for resistant hypertension. The resetting of renal hormone systems following denervation is still unclear and may include compensatory mechanisms.
Methods: Left kidneys of 8 WKY rats were surgically denervated and 8 rats sham-operated. After 6 days, renal concentrations of bradykinin (BK), kallikrein (K), renin (R), angiotensin (A) I and II, A(2-8), A(3-8), A(4-8), norepinephrine (NE), epinephrine (E), dopamine (D), and plasma R concentration (PRC) were determined by HPLC or biochemically. The renal innervation was studied immunohistologically, mRNA expression by PCR.
Results: PRC gave no group difference (p = NS). Catecholaminergic or sensory nerve fibers were absent in denervated kidneys. Left denervated kidneys showed lower AI (39.5%), AII (35.1%, p = NS), R (30.2%), NE (99.2%), D (-30.4%) and BK (30.4%) concentrations (p <0.05) vs. sham left kidneys; K, A fragments, and E were comparable (p = NS). Right kidneys had lower BK (26,6%, p = NS), AI (41.2%, p = NS), AII (22.5%, p <0.05) and higher E (46.4%, p <0.05) levels vs. sham right kidneys without differences for K, R, A fragments, NE or DA. Intraindividually, left denervated kidneys had lower R (23.5%), A (2-8) (45.9%), D (28.6%), and NE (99.2%) levels vs. right kidneys (p <0.05). mRNA levels of D-decarboxylase (DDC), D-β-hydroxylase, eNO-synthase (eNOS) and transforming growth-factor β (TGFβ) were higher (22–59%) in denervated vs. right (p = NS) or sham kidneys (eNOS, p <0.05; DDC; TGFβ); A-substrate and converting enzyme were unchanged.
Conclusion: Unilateral denervation suppresses ipsilateral kidney NE and bilateral AII and BK levels by side-dependent mechanisms targeting the renal pressure-natriuresis relationship. This may include neuropeptide-mediated pathways.

P40

Pregnancy Outcome Following Exposure to Angiotensin-Converting Enzyme Inhibitors or Angiotensin Receptor Antagonists: A Systematic Review

M. Bullo[1], S. Tschumi[1], B. S. Bucher[1], M. G. Bianchetti[2], G. D. Simonetti[1]
[1]Berne, [2]Bellinzona

Purpose: The objective was to analyze the outcome following prenatal exposure to angiotensin-converting enzyme inhibitors (ACE-Is) or angiotensin receptor antagonists (ARBs).
Methods: For this purpose, a systematic review of published cases reports and case series dealing with intrauterine exposure to ACE-Is or to ARBs using Medline as source of data was performed. The publications retained for analysis included patients who were described individually, revealing at minimum the gestational age, substance used, period of medication intake and the outcome.
Results: In total, 72 reports were included; 37 articles (118 well-documented cases) described the prenatal exposure to ACE-Is and 35 articles (68 cases) described the prenatal exposure to ARBs. Overall, 52% of the newborns exposed to ACE-Is and 13% of the newborns exposed to ARBs did not exhibit any complications (p <0.0001). Neonatal complications were more frequent following exposure to ARBs and included renal failure, oligohydramnios, death, arterial hypotension, intrauterine growth retardation, respiratory distress syndrome, pulmonary hypoplasia, hypocalvaria, limb defects, persistent patent ductus arteriosus or cerebral complications. The long-term outcome is described as positive in only 50% of the exposed children.
Conclusion: Fetopathy caused by exposure to ACE-Is or ARBs has relevant neonatal and long-term complications. The outcome is poorer following exposure to ARBs. We propose the term “fetal renin-angiotensin system blockade syndrome” to describe the related clinical findings. Thirty years after the first description of ACE-I fetopathy, relevant complications are at present regularly described, indicating that the awareness of the deleterious effect of prenatal exposure to drugs inhibiting the renin-angiotensin system should be improved.

P41

Nedd4-2 Ablation in Mice Leads to Upregulation of NCC Compensated by Decreased αENaC and Increased ROMK

C. Ronzaud[1], D. Loffing-Cueni[2], P. Hausel[1], A. Debonneville[1], S. Ram Malsure[1], N. Fowler-Jaeger[1], B. Yang[3], R. Koesters[4], M. Maillard[1], E. Hummler[1], J. Loffing[1], O. Staub[1]
[1]Lausanne, [2]Zurich, [3]Iowa City, US/US, [4]Heidelberg/DE

Purpose: Control of renal Na and K transport by aldosterone is crucial for maintaining Na/K balance and blood pressure (BP). Aldosterone acts in part by preventing ENaC degradation by the ubiquitin ligase Nedd4-2 (N4-2). To determine the role of renal N4-2 in mediating salt-sensitive hypertension observed in N4-2 total knockout (KO) mice (Shi et al., 2008), inducible renal tubule-specific N4-2 KO mice were generated using the TetOn/CreLoxP systems to delete exons 5-6 of the N4-2 gene.
Methods: Pax8/LC1 mice, allowing tetracycline-inducible Cre-mediated recombination in renal tubules, were bred with N4-2fl/fl mice to obtain mutants (N4-2fl/fl/Pax8/LC1) and controls (N4-2fl/fl/Pax8 or N4-2fl/fl/LC1), all treated with doxycyclin.
Results: N4-2 was completely lost in all renal tubular segments in dox-treated mutants. Under both standard and high-Na diets, mutants were able to maintain normal Na/K balance, whereas plasma aldosterone and urine volume were increased. Interestingly, mutants displayed hypertension and elevated urine Ca excretion under high-Na diet that could be treated with thiazides, suggesting increased NCC activity. Consistently, mutants showed increased NCC protein abundance and phosphorylation. βENaC, γENaC, and ROMK protein expression was increased as well. Unexpectedly, αENaC protein and mRNA were decreased, likely related to the lowered plasma aldosterone levels and compensating the increased NCC activity.
Conclusion: These *in vivo* data show: 1) N4-2 effects on β/γENaC, but not αENaC; 2) importance of N4-2 for controlling BP and Ca absorption via regulating NCC; 3) αENaC downregulation and ROMK upregulation that may help preventing hypertension and hyperkalemia, respectively.

P42

Inhibitory Effects of Geldanamycin Analogues on Adipocyte Differentiation and Fat Mass Accumulation

S. Desarzens, A. Debonneville, O. Staub, N. Faresse
Lausanne

Purpose: The mineralocorticoid and the glucocorticoid receptors (MR and GR) are two steroid receptors expressed in adipocyte. Their activation either by aldosterone or glucocorticoids promotes the adipogenic transcriptional program that leads to the conversion of preadipocyte to mature adipocyte. We and others have demonstrated that geldanamycin analogues have a potent inhibitory effect on MR and GR signaling. Geldanamycin analogues are anti-tumor antibiotics used in phase II clinical trials in cancer treatment. They bind to the chaperone protein Hsp90 and, by altering its function, render steroid receptors insensitive to their ligands.
Methods: Given that the expression and activity of MR and GR are crucial for adipogenesis, we investigated whether geldanamycin analogues have an effect on adipocyte differentiationin vitrousing 3T3-L1 cells. In parallel, we evaluated the effects of geldanamycin analogues on mice fed with high fat diet.
Results: We found that early exposure of preadipocyte cells to geldanamycin analogues inhibits their adipocyte conversion, by inhibiting the adipogenic transcriptional program and lipid droplets accumulation into the cell.In vivo, the weight gain of mice treated with geldanamycin analogue is significantly lower than control mice. This difference in body weight is explained by a reduction of fat mass and not in lean mass or water, as evidenced by evaluation of the body mass composition. Biochemical analysis revealed that anti-adipogenic effects of geldanamycin analogues are related to an inhibition of MR activity and a reduction of GR expression.
Conclusion: Taken together, our observations lead us to propose geldanamycin analogues as potent therapeutics not only in cancer treatment but also in the control of obesity and all its related metabolic complications.

P43

Renal Alterations in the Post Partum After a Preeclampsia

A. Péchère-Bertschi[1], G. Wuerzner[2], B. Ponte, P. Montillier[1], M. Boulvain[1], M. Burnier[2], O. P. Irion[1]
[1]Geneva, [2]Lausanne

Purpose: Preeclampsia is a hypertensive disorder of the pregnancy defined by BP ≥140/90 mm Hg and proteinuria >0.3 g/24 h, or 2++ on dipstick, occuring after 20 weeks gestation. Epidemiologic data underscore that preeclampsia is not just a disease of pregnancy that resolves with delivery, but may be considered a risk marker for later-life diseases, including cardiovascular and renal diseases and the metabolic syndrome.

Methods: Longitudinal prospective observational cohort study to analyze the renal abnormalities in the post-partum in 182 women having suffered from preeclampsia.Office and 24h ambulatory BP, renal function, ultrasensitive CRP, urine spot and 24h collect and genetics were obtained at 6we and 6mo post partum, and then followed yearly.
Results: Mean [SD] age was 32.0 [5.8] years and BMI 29.5 [4.2] kg/m^2. Seventy percent were Caucasian and 21% were blacks. At 6 we, urine albumin/creatinine ratio was 10.8 [4.7] mg/mmol. The 24h urine excretion of albumin was 216.7 [97.1] mg/d. GFR estimated with the Cockroft formula was 149 ml/min [11.9]. Ultrasensitive C reactive protein was 12.7 [7.1]. The prevalence of hypertension or ongoing antihypertensive treatment was 36%. Ambulatory blood pressure daytime was (systolic/diastolic) 120.9 [15.6]/83.5 [10.8] and 112.3 [19.5]/75.1[11.6] mm Hg. Thirty-nine % of women had an ambulatory blood pressure daytime ≥135/85 mm Hg, and 10% were non-dippers.
Conclusion: Preeclamptic women do not normalize their blood pressure and renal function in the post-partum. The prevalence of both office and ambulatory hypertension seems to be very high together with an elevated excretion of microalbuminuria and some degree of renal hyperfiltration. These women have to be identified and targeted in order to establish an efficient prevention and to impact throughout the postpartum course.

P44

Relationship of Aldosterone to Urinary Sodium and Potassium Excretion in the Swiss Kidney Project on Genes in Hypertension

D. Ackermann[1], B. Ponte[2], M. Pruijm[3], M. Mohaupt[1], F. J. Frey[1], M. Burnier[3], P.-Y. Martin[2], I. Guessous[2], B. Dick[1], M. Bochud[3]
[1]Berne, [2]Geneva, [3]Lausanne

Purpose: Aldosterone plays a major role in the control of sodium and potassium balance by promoting renal sodium reabsorption and potassium secretion. Urinary excretion is mainly determined by diet which in turn should influence aldosterone production. We hypothesized this relationship to be maintained in a circadian pattern within an adult population.
Methods: Nuclear families were randomly selected from the adult population of Lausanne, Geneva and Bern. Sodium and potassium excretions were measured in discrete day and night urines according to a standardized protocol. Tetrahydroaldosterone, the major urinary aldosterone metabolite, was measured by gas chromatography-mass spectrometry and standardized for urinary creatinine.
Results: So far, 282 participants (150 women and 132 men) had data available for this analysis. Mean (SD) age and BMI were 47 (17) years and 25.3 (5) kg/m², respectively. Mean (SD) day and night excretions were 95.6 (49.2) and 43.1 (27.3) mmoL for sodium, and 47.2 (18.4) and 18.4 (8.5) mmoL for potassium. Daytime tetrahydroaldosterone excretion was higher than night time, even after considering duration (17.8 [24.9] versus 7.9 [13.6] µg/period, P <0.001). In multivariable mixed linear models adjusted for age, sex, center, BMI and renal function, higher tetrahydroaldosterone excretion was associated with lower sodium excretion (coefficient [SE] = –0.067 [0.012] and –0.076 [0.022] per 10 mmoL, P ≤0.001, for day and night, respectively) and higher potassium excretion (0.014 [0.003] and 0.048 [0.007] per mmoL, P <0.001, respectively).
Conclusion: Circadian urinary sodium and potassium excretion is strongly associated to urinary tetrahydroaldosterone excretion in a population-based sample suggesting a timely adaptation to environmental demands.

P45

The Role of Renal SGK1 in the Control of Potassium Homeostasis in Inducible Nephron-Specific Sgk1-KO Mice

L. Al-Qusairi[1], A. Debonneville[1], N. Faresse[1], J. Loffing[2], O. Staub[1]
[1]Lausanne, [2]Zurich

Purpose: Dietary K+load results in increased kalemia, leading to aldosterone (aldo) release in order to stimulate K+secretion in the ASDN. However, the regulatory mechanisms of this regulation are unclear. Here, we aim to identify new pathways involved in the regulation of K+secretion. More specifically, we have investigated the role of the aldosterone inducible Sgk1 kinase.
Methods: To avoid the compensatory mechanisms which may mask the role of SGK1 during development, we employed the inducible nephron specific SGK1 KO mice which take advantage of the previously described TetOn/CreLoxP system, in which rtTA is under the control of the Pax8 promotor, allowing inducible inactivation of the floxed Sgk1 allele in the renal tubules (Sgk1fl/fl/Pax8/LC1 mice) (Faresse et al., AJP, 2012). Normal and mutant animals were challenged by different K+diets. The metabolic parameters were analyzed, together with the protein and the mRNA levels of the key players in K+regulation.
Results: Our results indicate that K+secretion is altered in nephron-specific SGK1-KO mice. Indeed, mutant animals exhibit 35% decrease in urine K+level after 2 days under high K+diet (5%). Interestingly, urine Na+level are also decreased in these conditions.
Conclusion: Our results indicate that SGK1 plays an important role in the long-term regulation of K+secretion. We are currently investigating accompanied changes in aldosterone, electrolyte levels and blood pressure. Moreover, blood pressure measurements will be undertaken.

Poster Presentations – Dialysis

P46

Troponin T for the Detection of Dialysis-Induced Myocardial Stunning in Hemodialysis Patients

T. Breidthardt[1], J. O. Burton[2], A. Odudu[2], M. T. Eldehni[2], H. J. Jefferies[2], C. W. McIntyre[2]
[1]Basel, [2]Derby/UK

Purpose: Recently, recurrent episodes of myocardial ischemia and transient segmental left ventricular wall-motion abnormalities have been established to occur commonly during standard thrice-weekly HD. These repeated episodes of myocardial stunning can eventually lead to myocardial hibernation, myocardial remodeling, scarring, and irreversible loss of contractile function and are becoming increasingly appreciated as a principal pathophysiologic foundation of excess cardiovascular mortality in HD patients. Cardiac troponins, structural proteins unique to the heart, are sensitive and specific biochemical markers of myocardial damage. In addition, cardiac troponin levels, as measured by fully automated standard assays, are superior to all other clinically available biomarkers for the diagnosis of acute myocardial ischemia. However, circulating levels of cardiac troponins are frequently elevated in long-term dialysis patients even in the absence of acute coronary syndromes. We therefore aimed to assess the association between the presence and extent of HD-induced myocardial ischemia and troponin T levels in unselected patients undergoing maintenance HD.
Methods: In 70 prevalent hemodialysis (HD) patients, HD-induced myocardial stunning was assessed echocardiographically at baseline and after 12 months. Nineteen patients were not available for the follow-up analysis. Dialysis was performed using Hospal Integra monitors (Hospal, Mirandola, Italy). Dialysate composition was sodium, 138 mmol/L; potassium, 1 mmol/L; calcium, 1.25 mmol/L; magnesium, 0.5 mmol/L; bicarbonate, 32 mmol/L; glucose, 5.6 mmol/L; and acetate, 3 mmol/L. All treatments were of 4 hours' duration. The extent to which predialysis troponin T was associated with the occurrence of HD-induced myocardial stunning was assessed as the primary endpoint.
Results: The median troponin T level in this hemodialysis cohort was 0.06 ng/ml (interquartile range, 0.02–0.10). At baseline, 64% of patients experienced myocardial stunning. These patients showed significantly higher troponin T levels than patients without stunning (0.08 ng/ml [0.05–0.12] versus 0.02 ng/ml [0.01–0.05]). Troponin T levels were significantly correlated to measures of myocardial stunning severity (number of affected segments: r = 0.42; change in ejection fraction from beginning of dialysis to end of dialysis: r = 20.45). In receiver operating characteristic analyses, predialytic troponin T achieved an area under the curve of 0.82 for the detection of myocardial stunning. In multivariable analysis, only ultrafiltration volume (odds ratio, 4.38 for every additional liter) and troponin T (odds ratio, 9.33 for every additional 0.1 ng/ml) were independently associated with myocardial stunning. After 12 months, nine patients had newly developed myocardial stunning. Patients developing myocardial stunning showed a significant increase in troponin T values over their baseline value (0.03 ng/ml [IQR, 0.01–0.05 ng/ml] at baseline versus 0.05 ng/ml [IQR, 0.03–0.11 ng/ml] at 1 year; P = 0.02). The median troponin T increase in the patients developing stunning was 0.03 ng/ml. Troponin T values did not significantly change in patients stunning at neither or both timepoints. Again, troponin T levels were significantly higher in patients who developed stunning after 12 months compared with patients not stunning at 12 months (stunning, 0.06 ng/ml [IQR, 0.03–0.11 ng/ml] versus 0.05 ng/ml [IQR, 0.01–0.07 ng/ml]; P = 0.04) regardless of baseline stunning.

Conclusion: In conclusion, troponin T levels in patients undergoing long-term HD are associated with the presence and severity of HD-induced myocardial stunning. This mechanism provides a robust pathophysiologic basis for the prognostic importance of elevated troponin levels in the HD population.

P47

Prevalence of Tunneled Catheter Colonization in Longterm Hemodialysis Patients Using Different Catheter Lock Strategies

P. Grosse, S. Erb, S. Tschudin, A. Widmer, U. Neff, M. Fischer, M. Dickenmann
Basel

Purpose: Catheter related bloodstream infections (CRBSI) in longterm hemodialysis patients with permanent venous catheters have been attributed to adverse outcomes in terms of mortality, morbidity and excess costs. To minimize the danger of CRBSI by manipulation of dialysis catheters the needleless luer-lock device TEGO®connector has been FDA-approved in 2006 as an alternative to standard catheter caps (SCC).
Methods: Our prospective, interventional study investigated the prevalence of asymptomatic catheter colonization as a risk factor for CRBSI in three different catheter lock techniques in 39 patients with permanent venous dialysis catheters in the University Hospital of Basel, Switzerland: (i) TEGO®system with saline locking solution, (ii) SCC with 46.7% citrate locking solution and (iii) SCC with 30% citrate locking solution plus intensive training of dialysis staff in aseptic catheter manipulation technique.
Results: We could demonstrate significantly higher rates of catheter colonization using the TEGO®connector with saline locking solution as compared to SCC with 46.7% citrate solution (OR 0.22, 95% CI 0.07–0.71, p = 0.011) or 30% citrate solution (OR 0.07, 95% CI 0.01–0.35, p = 0.001).
Conclusion: We recommend cautious use of needle-free connectors for dialysis venous accesses since they might bear the danger of increased rates of CRBSI.

P48

Efficacy and Safety of Citrate-Based Anticoagulation in Patients With AKI in the Intensive Care Unit

F. Stucker, J. Pugin, J. Tataw, B. Ponte, L. Brochard, P.-Y. Martin, P. Saudan
Geneva

Purpose: A systemic anticoagulation is often required to prevent clotting of filter and extracorporeal circulation in ICU patients undergoing continuous renal replacement therapy (CRRT). A regional citrate-based anticoagulation (RCA) does not induce a systemic anticoagulation and prolongs the filter lifespan. Nevertheless metabolic side-effects have been associated with this therapy. We are conducting a randomized controlled trial with patients requiring CRRT to determine whether a RCA is more effective than heparin in terms of renal replacement delivered dose.
Methods: Patients: included if: >18 yrs old with AKI requiring CRRT. excluded if: active hemorrhagic disorder, pregnancy, history of heparin-induced thrombopenia, consent form not obtained. Methods: patients randomized to either CRRT with RCA or heparin anticoagulation. Treatment with Prismaflex (HDF mode, overall RRT dose 30 ml/kg/h and filter change every 72 hours). Primary endpoints: effective daily RRT dose and filter lifespan. Secondary endpoints: survival at 28 days, number of hemorrhagical events requiring blood transfusions, severe metabolic complications.
Results: From 10.2010 until 04.2012, 52 out of 120 patients treated with CRRT were randomized (exclusion criteria were active hemorrhagic disorders or severe thrombocytopenia [21%], terminal liver failure [9%], chronic maintenance dialysis [25%] or others [45 %]). Mean age was 60 ± 9 years. Etiology of AKIs were medical (77%), surgical (23%). Mean CRRT duration was 5 ± 5 days. Effective daily RRT dose was 96 ± 12% in the RCA group and 85 ± 15% in the heparin group (p = 0.057). 28-days mortality was 29% in the RCA group and 27% in the heparin group.
Conclusion: These preliminary results seem to show an advantage of RCA over heparin-based anticoagulation in terms of effective daily delivered RRT dose.

P49

Outcome of Patients on Renal Replacement Therapy (RRT) for Amyloid Nephropathy of Familial Mediterranean Fever (FMF)

M. Voskanyan, H. Nazaryan, S. Babloyan, A. Sarkissian
Yerevan/AM

Purpose: Renal amyloidosis (RA) of FMF – although largely preventable – still is a major health problem in Armenia and an important cause of death.
Methods: From January 2002 till September 2012 279 patients were admitted to our centre for RRT, of whom 40 (14.3%) had RA of FMF. Their mean age was 31.4 ± 12.7 (range 12.6–52.9), 60% were males. Mean duration of hemodialysis (HD) was 1.6 ± 1.7 years (range 0.1–6.0).
Results: Hemodialysis: Of the 28 patients not undergoing renal transplantation (Tx) in Armenia, 9 died (systemic amyloidosis – 5, heart attack – 2, stroke –2); 15 moved to another country and 4 remained on dialysis. Complications were severe erythropoietin resistance (7), intradialytic hypotension (4) and nephrotic syndrome (1). Living related donor Tx was done in our institution in 12 patients aged 38 ± 11.6 years, i.e. 12.5% of all Tx (96) done at the same period. In addition to standard immunosuppression all received low dose colchicine (0.6–1.2 mg/day). Main complications were rejection (8), delayed graft function for tubular necrosis (2), lymphocele (2), CMV disease (2) and tuberculosis (1). Additional problems included diarrhea (colchicine, MMF, generalized amyloidosis; 9) and severe neuropathy due to interaction of cyclosporine with colchicine (1). One patient died of generalized amyloidosis and 1 kidney was lost after reduction of immunosuppression due to tuberculosis. Ten patients have good renal function.
Conclusion: The number of patients with amyloidosis of FMF requiring RRT is alarming, and preventive measures must be strengthened. Outcome of HD was poor in contrast to renal Tx.

P50

What Determines Beta 2 Microglobulin Levels in HD Patients in the 2010's?

D. Bischof, D. Tsinalis, E. Matheis, I. Binet
St. Gallen

Purpose: Beta2-microglobulin (B2M) is used as a marker for middle molecules uremic toxins and is associated with outcome on haemodialysis (HD). Our goal was to evaluate the determinants of B2M in a chronic haemodialysis population treated according to the current guidelines.
Methods: We retrospectively analysed B2M levels in all HD patients treated for at least 6 months at our center between 01.01.2010 and 01.08.2012. The B2M value from the last measurement was analysed. At the timepoint of B2M measurement we analysed dialysis modality, Kt/V, vascular access, urine output and residual renal function (RRF), diabetes, active neoplasia or autoimmune disease, current immuno-suppression, CRP and severe infection within 3 months.
Results: 136 patients (m = 86, f = 50) fulfilled the criteria, 62 on HDF, 37 on HD and 37 on Genius dialysis. B2M levels were 20.68 ± 7.6 mg/L after a mean dialysis time of 42.4 ± 49.6 months. Mean eKt/V was 1.5 ± 0.39, mean RRF 3.1 ± 4.1 mL/min. In an univariate analysis HD- modality, vascular access, dialysis vintage, weekly HD hours, HD frequency, Urine output and RRF were identified as possible predictors for B2M. 12/136 (9%) patients who had a B2M >30 mg/L showed lesser RRF (1.2 vs 3.2 ml/min, p <0.005) and slightly shorter HD hours (12.0 vs 12.2 hrs/wk, p <0.005) but no difference regarding HD vintage. However, in a multivariate analysis RRF emerged as the only significant predictor for B2M levels (p = 0.0003), which was also true for patients with B2M >30 mg/L.
Conclusion: In the era of high-flux membranes and for patients dialysed according to the current guidelines neither HD-modality nor vascular access were relevant for the B2M levels. In this series the multivariate analysis revealed that residual renal function was the only determinant of B2M levels.

P51

Accuracy of Five 25-Hydroxyvitamin D [25(Oh)D] Assays as Compared to the Reference Method Lc-Ms/Ms in Chronic Hemodialysis (Hd) Patients

B. Fellay[1], Y. Guillod[2], N. Jorand[1], G. Allemann[1], J.-L. Magnin[1], E. Descombes[1]
[1]Freiburg, [2]Niederwangen

Purpose: There are few data on the accuracy of 25(OH)D assays in HD patients. Thus, we wanted to assess the reliability of some currently used assays by comparing them to the reference method LC-MS/MS.
Methods: The 25(OH)D serum levels determined by liquid chromatography-tandem mass spectrometry (Thermofisher, Quantum LC-MS/MS) were compared to those of the following assays:Roche

D3(Cobas 6000),Roche D2-D3(Cobas 6000),Abbott(Architect i1000SR),IDS(iSYS),Thermo scientific(Dionex HPLC Recipe). Blood samples were taken before HD. The results were evaluated by the Passing-Bablok test.
Results: The25(OH)D measurements with the different assays showed significant deviations compared to those obtained with LC-MS/MS. Compared to the reference method, the Passing-Bablok regression equations were:Roche D3(y = –2.53+0.71x, n = 49);Roche D2-D3 (y = –1.1 + 0.82x, n = 142); Abbott (y = 16.03+0.47x, n = 142); IDS (y = 7.86 + 0.61x, n = 49); Thermo HPLC(y = –2.18+1.18x, n = 49). These results show a significant and, for the majority of the assays excepted the Roche D2-D3 and the Thermo HPLC, a very important deviation from the ideal intercept and slope of the curve (ideally close to 0.00 and 1.00, resp.). These deviations are much more important compared to the assays' accuracy data of the manufacturers (derived from 25[OH]D measurements in healthy individuals mostly with low levels).
Conclusion: 1) These results show that in HD patients significant different serum values of vitamin D could be obtained depending on the assay used. 2) These discrepancies seem to be much more important in HD patients than those reported in healthy subjects and vary depending on the considered assay. These points should be considered for the diagnosis and therapy of vitamin D deficiency, or when comparing the results of the studies dealing with vitamin 25(OH) D, in HD patients.

P52

Oral Post-Dialysis Cholecalciferol (Vitd3) Supplementation in Patients on Maintenance Hemodialysis (HD): One Size Does Not Fit All

E. Descombes[1], B. Fellay[1], O. M. Hemett[1], G. Fellay[2], J.-L. Magnin[1]
[1]Freiburg, [2]Villars-sur-Glâne

Purpose: We studied in HD patients (pts) the post-HD dose of VitD3 (given as Dialvit with added 2000 IU of VitD3) needed to maintain the vitD levels in the optimal range of 75–150 nmol/l.
Methods: Were included 26 pts (age 68 ± 9.8yr) with low vitD who gave their informed consent. 25(OH)D2-D3 was measured bimonthly (Jan 2011 to 2012) with the Roche vitD total assay. The first 2 months, 2000 IU of VitD3 were given after each HD by replacing 1 of the 2 tablets of Dialvit with 1 of *DialvitD* (Bichsel AG). After month 2, the pts with vitD <75 nmol/l received 2 tablets/HD (= 12000 IU/wk). After month 4, the dose was adapted every 2 mo (by giving 1 to 6 of *DialvitD*/wk) to maintain vitD levels in the target. Twelve pts not receiving vitD served as controls.
Results: In controls there was a significant seasonal increase of vitD from 29.2 ± 15.6 nmol/l to 65.6 ± 17.0 nmol/l in July (p <0.01).
Results:

	WINTER	SUMMER	WINTER
Month	**0**	**6**	**12**
n	26	26	24
25(OH)VitD (nmol/l)	27.50 ± 14.90	140.10 ± 28.30*	95.60 ± 20.9*
25(OH)VitD: range (nmol/l)	5 to 57	89 to 199	53 to 138
VitD <50 nmol/l	96%	0%	0%
VitD >150 nmol/l	0%	35%	0%
Weekly dose of VitD3 (IU)	0	7000 ± 3212	5917 ± 4106
Range of weekly dose (IU)	0	4000 to 12000	0 to 12000
1-25(OH)vitD (pmol/l)	32.9 ± 15.1	32.5 ± 23.1	30.6 ± 11.0
* p <0.01 compared to baseline and controls			

At month six, 35% of the pts had vitD >150 nmol/l due to a higher mean dose of vitD associated to the seasonal increase of vitD. At month 12, 86% of the pts had vitD levels within the target but the amount needed to maintain these levels varied from 0 (n = 2) to 12000 IU/wk (n = 5).
Conclusion: a) The prevalence of vitD deficiency in Swiss HD pts varies according to the season, as in the general Swiss population; b) the amount of oral post-HD VitD3 needed to maintain the vitD levels within the optimal range varies widely among pts and may be influenced by seasonal changes.

P53

Comparison of Hb Events (Decreases >1 g/Dl) in a Hemodialysis Patient Population – an Analysis of the Swiss MOTION Survey

L. Gabutti[1], P. Meier[2], A.-K. Schwarzkopf[3], J. Záruba[4], P. Ambühl[5]
[1]Locarno, [2]Sion, [3]Berne, [4]Urdorf, [5]Zurich

Purpose: Controversy exists regarding optimal Hb level and the upper limit of the desired range by ESA treatment. Current recommendations suggest an optimal target for Hb concentration between 10–12 g/dL. The survey's objective is to analyze significant Hb events.
Methods: Multicenter, retrospective/prospective, observational, non-interventional survey in HD patients documenting Hb events defined by a drop <10 g/dl and by >1 g/dl from baseline along with associated factors. We present an analysis of 378 patients with a mean follow-up (SD) of 12 (0.2) months from 25 sites. Of all patients 118 (31%) had 143 Hb events (group 1, n = 143). In order to analyze the impact of ESA dosing and Hb values prior to a drop below 10 g/dl the data was compared to decreases of >1 g/dl in the overall population remaining in the target range of 10–12 g/dL (group 2, n = 309).
Results: 378 patients (40% female), mean age and weight (SD) was 66 (14) years and 73 (16) kg, respectively.
Group 1: Reduction in Hb from 11.4 g/dL to 9.2 g/dL (–19%), 5% dose reduction. Group 2: Hb reduction of 15% (12.4 g/dL to 10.5 g/dL), 20% dose reduction. Most frequent reasons for Hb drops <10 g/dl: illness 36%, bleeding 21%.
Conclusion: For Hb events remaining within target range (group 2) an intentional ESA dose reduction was documented prior to the observed Hb decrease. For Hb events below target range (group 1) this was less pronounced, suggesting interfering factors other than ESA dose adjustments as a trigger.
Survey supported by Amgen

	Months	–6	–5	–4	–3	–2	–1	Hb event	+1	+2	+3	+4	+5	+6
1	Hb	11.2	11.3	11.4	11.4	11.6	11.4	9.2	10.0	10.7	11.3	11.5	11.4	11.3
	Dose/ week	44	46	47	47	44	42	51	67	65	62	54	54	54
2	Hb	11.5	11.6	11.7	12.0	12.3	12.4	10.5	10.8	11.3	11.5	11.6	11.6	11.6
	Dose/ week	45	45	47	47	40	32	36	46	47	45	44	43	43

P54

New Generation of High-Flux Dialyzers: In-Vivo Quantification of Small and Large Size Solute Transport

B. von Albertini, C. Mathieu, A. Bösch, D. Huber, A. Cherpillod
Lausanne

Purpose: A ≈10% smaller inner diameter of hollow-fibers in newly available high-flux dialyzers results in greater resistance to blood flow and thereby increases hydrostatic pressure gradients across the membrane. This results in increased internal filtration with convection of solutes from blood, with backfiltration of fresh dialysate to blood occuring simultaneously under volumetric control of the equipment. Aim was to quantify small and large solute transport with such dialyzers in-vivo.
Methods:

Dialyzer studied	Manufact.	Membrane	m²	Inner Diameter	Wall Thickn.	UF-Coeff.
FX CorDiax 80	Fresenius	PS Helixone® plus	1.8	185 µm	35 µm	64 ml/h*mm Hg
RevaclearMax	Gambro	Polyarylethersulfone	1.8	190 µm	35 µm	60 ml/h*mm Hg

Results: (ml/min)	at Qb 400, Qd 650:				at Qb 500, Qd 800:			
Means ± S.D.	Kurea	Kcreat	Kphos	Kβ2M	Kurea	Kcreat	Kphos	Kβ2M
CorDiax 80 (n = 3)	319 ±2	232 ± 11	250 ± 6	83 ± 2	381 ± 15	267 ± 20	285 ± 12	87 ± 12
RevaclearMax (n = 4)	320 ± 19	222 ± 24	250 ± 20	91 ± 7	360 ± 17	249 ± 22	287 ± 22	95 ± 6
n.s.								

In-vivo clearance (K) measurements were performed in 4 stable ESRD patients during routine hemodialysis. Solute mass removal for calculation of K was derived from blood and dialysate, means were used for analysis. For urea, creatinine and phosphates, whole blood K was calculated, based on blood flow rate, hematocrit, solute plasma concentrations and specific red cell diffusion coefficients. For β2microglobulin, straight plasma clearance was calculated from plasma concentration and flow rate. Further studies are planned.

Conclusion: High-flux HD, done with one of these highly permeable dialyzers, yields sizable diffusive small solute transport, which increases with higher Qb. The unprecedented high β2m clearance found suggests primarily convective transport by important internal filtration and simultaneous backfiltration under volumetric control and approximates in magnitude that of HDF.

P55

Prevalence and Characteristics of Diabetic Subjects on Maintenance Dialysis in the Canton de Vaud in 2009

C. M. M. Stamm[1], O. Phan[2], T. Gauthier[3], G. Halabi[4], F. Barbey[5], C. Mathieu[6], A. Cherpillod[6], B. von Albertini[6], D. Teta[6], M. Burnier[6], A. Zanchi[6]
[1]Montreux, [2]Payerne&Lausanne, [3]Vevey, [4]Yverdon-Les-bains, [5]Nyon, [6]Lausanne

Purpose: The prevalence of ESRD in type1 and type2 diabetes is increasing around the world but data in Switzerland are lacking. Aims were to establish the prevalence and characteristics of diabetic subjects on maintenance dialysis in the Canton de Vaud as of December 31, 2009.
Methods: Diabetic subjects were identified in all 8 dialysis centres and data was collected from medical records.
Results: A total of 101 diabetic subjects (DM) on dialysis were identified with a sex ratio M/F of 2.15 and a DM2/DM1 ratio of 7.3. Mean age was 69.6 ± 10.6y (38–88 y) and mean BMI 27.2 ± 4.3 kg/m^2. Mean Hb1Ac was 6.9 ± 1.8%. Diabetes duration to dialysis was 16.2 ± 11.4 y. Dialysis duration was 3.6 ± 3.2 y. In only 18% of cases, ESRD had clearly another etiology than diabetes. 54% of patients experienced at least one macrovascular complication. 64% had diabetic retinopathy, 20% underwent amputation and 19% had chronic lower extremity wounds. Mean BP was 146/70 mm Hg at pre-dialysis. The mean Hb was 117.9 ± 11.0 g/l. DM1 subjects were younger, lighter, had a higher Hb1Ac and a longer DM duration at start of dialysis than DM2 subjects. The prevalence of diabetes among subjects on maintenance dialysis in the Canton de Vaud was 35.6%. Compared with a study conducted by Sandoz and al. in 2001, the prevalence of DM2 in the dialysis population has increased from 18% to 31% in 8 years while the total number of subjects on dialysis increased from 182 to 284.
Conclusion: The number of diabetic subjects on dialysis has greatly increased in 8 years in the Canton de Vaud. This increase is mainly due to an increase in DM2 cases. Considering the costs, poor quality of life and mortality linked to dialysis therapy, diabetic subjects with diabetic nephropathy and/or a decline in renal function urgently need multidisciplinary and intensified care to delay ESRD.

P56

Prognostic Value of Circulating Klotho and FGF23 in Dialysis Patients

A. Nowak[1], B. Friedrich[2], F. Artunc[2], T. Breidthardt[3], R. Twerenbold[3], M. Potocki[3], A. Serra[1], C. Mueller[3]
[1]Zurich, [2]Tübingen/DE, [3]Basel

Purpose: Klotho is known to activate the phosphatonin fibroblast growth factor (FGF23) which stimulates urinary phosphate excretion in an attempt to overcome the reduced phosphate excretion capacity in kidney disease. Thus, Klotho is assumed to have cardiovasculo-protective and antiaging properties, but this hypothesis has not been proven. The present study therefore explored the effects of circulating Klotho levels on all-cause mortality in a large hemodialysis patient cohort.
Methods: We prospectively measured the baseline circulating Klotho and FGF23 levels of 239 prevalent hemodialysis patients from three dialysis facilities. The primary hypothesis of the study was that low circulating Klotho levels may correlate with mortality in a long-term observation period. The exploratary hypothesis was that high circulating Klotho levels may protect from atrial fibrillation (AF).
Results: Thirty-seven patients (15%) died within the median follow-up time of 682 [657–761] days. Klotho levels were not significantly different in non-survivors and survivors (347 [260–456] vs. 339 [260–425] RU/ml, P = 0.59). Increasing FGF23 (HR 1.2; 95%CI 0.98–1.45, P = 0.04 per 1000 RU/ml increase) but not Klotho levels (HR 1.0; 95%CI 1.00–1.00, P = 0.46 per 100 pg/ml) were associated with mortality, as assessed by the multivariate adjusted analysis. Klotho (350 [262–447] vs. 307 [238–383], P = 0.03) and FGF23 (474 [209–1413] vs. 177 [73–618], P = 0.003) levels were significantly higher in patients with versus without AF. In an adjusted analysis for age, gender, dialysis center, cardiovascular comorbidities and anuria, the relationship between Klotho and AF remained significant.
Conclusion: Low circulating Klotho levels are not associated with mortality in hemodialysis patients. However, higher circulating Klotho levels seem to be protective against AF.

P57

Too Many Late Nephrology Referral in Pre-Esrd Patients in Switzerland

H. Elsässer, D. Kiss
Liestal

Purpose: Multiple observational studies reported increased morbidity and mortality in pre-ESRD patients referred late to specialized renal services. Early detection and intervention to retard progression as well as prevention and treatment of uremic complications are the goals of pre-ESRD management. In addition patient information, choice and preparation for the individually adapted renal replacement modality needs an early referral to the specialist. Obtaining incident data of ESRD patients starting renal replacement therapie (RRT) in Switzerland was the aim of this survey.
Methods: For the timeframe of January 1st to June 30th 2012 all haemodialysis centers in Switzerland were asked for the number of incidents for ESRD leading to implementation of RRT. Patient age, serumcreatinine and eGFR (MDRD), time after the first contact to the nephrologist, civil status and independence in terms of daily living at inition of RRT were of further interest.
Results: Preliminary results considering 36 swiss nephrological centers (nearly 50% of all) showed: 244 individuals with the mean age of 67.1y (31–90), started renal replacement therapy in the first half of 2012. 79/244 (32%) patients contacted a renal specialist for the first time less than one month before inition of RRT. Mean serumcreatinine at that moment was 656umol/l, eGFR (MDRD) 9 ml/min. 22/79 (27.8%) were older than 75y. However 165/244 (68%) presenting with ESRD were met their nephrologist substantially earlier (33.5 [6–98] months). 34/165 (20.1%) were older than 75y.
Conclusion: Around one third of the new swiss ESRD patients were unexpected, e.g. have seen the nephrologist less than one month before inition of renal replacement therapy – too late for optimal pre-ESRD management.

P58

Results from the Multicenter Observational READY Survey: Effective Treatment With Mircera in Patients on Peritoneal Dialysis

A. Komarek[1], A. Fischer[2], N. Marangon[3], S. Segerer[4]
[1]Reinach, [2]Lucerne, [3]Geneva, [4]Zurich

Purpose: C.E.R.A. (methoxy-polyethylene glycol-epoetin beta), with its once monthly dosing interval, allows outpatient anemia management in patients (pts) with chronic kidney disease and may be useful in peritoneal dialysis (PD).
Methods: The READY postmarketing survey (03.2010–12.2011) was performed in 12 Swiss nephrology centers on 54 PD pts treated with erythropoiesis stimulating agents (ESA) at least once monthly over 12 months. Demographics, hemoglobin (Hb), previous ESA dose and iron parameters were evaluated in 3 questionnaires at baseline (BL), months 6 and 12.
Results: Here we present interim data after 6 months of observation. 42 pts were treated with C.E.R.A. and had complete data sets. 11 pts did not complete the 6 months due to switch to hemodialysis (5), transplantation (3), change of medication (2) and death (1). Pts were on PD for 3.5 years (range 1–9 years). During the first 6 months mean Hb stayed stable, with 11.2 ± 1.1 g/dl at BL and 11.1 ± 1.0g/dl at month 6 (p = 1.0). Mean dose of C.E.R.A. remained stable (111 ± 65 µg/month at BL vs. 127 ± 65 µg/month at month 6, p = 0.38).
Mean iron parameters were sufficient at BL (ferritin 311 ± 204 µg/l, TSAT 29 ± 11%) and month 6 (ferritin 372 ± 207 µg/l, TSAT 34 ± 11%). After 6 months, of the 46% of PD pts below guideline levels (ferritin <200 µg/l and/or TSAT <20%), 1/4 remained below. In 7 pts C.E.R.A. application intervals were extended up to 6 weeks, achieving stable Hb levels without need for increased doses.
Conclusion: Swiss PD pts receiving C.E.R.A. maintain stable Hb levels within the range recommended by guidelines at the start of the survey in 2010. Application intervals extended up to 6 weeks provided stable Hb values. Thus, physicians may combine C.E.R.A. administration with regular hospital visits, potentially improving drug adherence and patient convenience.

P60

Inhibition Of Sodium-Glucose Cotransporters Prevents Disease Progression in Han:SPRD Rats With Polycystic Kidney Disease

X. Wang, S. Zhang, L. Yang, D. Spichtig, S. Kapoor, S. Segerer, A. Serra, O. Devuyst, R. P. Wüthrich
Zurich

Purpose: Transepithelial cyst fluid secretion is one of the key features involved in the progression of polycystic kidney disease (PKD). The role of the apical renal Na+-glucose co-transporters (SGLT) in that process is not known.
Methods: We tested the hypothesis that induction of glycosuria and osmotic diuresis with the SGLT inhibitor phlorizin could inhibit cyst growth and delay renal disease progression in a rat model of PKD. To that end we induced glycosuria by subcutaneous injection of phlorizin (400 mg/kg/d) in male heterozygous (Cy/+) and wild-type (+/+) Han:SPRD rats. As expected, phlorizin induced immediate and sustained glycosuria and osmotic diuresis in these rats.
Results: Cy/+ rats treated with phlorizin for 5 weeks showed a 56% increase in creatinine clearance, with a 12.6% lower 2 kidneys/body weight (2K/BW) ratio and a 28.4% lower renal cyst index, as well as a 63% reduction in urinary albumin excretion as compared with vehicle-treated Cy/+ rats. Ki67 staining revealed a significantly lower number of positive nuclei in dilated tubules and cysts of Cy/+ rats treated with phlorizin, as well as a marked inhibition of the activated MAP kinase pathway. In contrast, the mTOR pathway remained unaltered.
Conclusion: These data demonstrate that long-term treatment with phlorizin has a significant inhibitory effect on cystic disease progression in a rat model of PKD, supporting the hypothesis that induction of glycosuria and osmotic diuresis (gycuresis) by renal SGLT inhibition could have a therapeutic effect in polycystic kidney disease.

P61

Efficacy of PA21, a New Iron-based Phosphate Binder, as Compared to Lanthanum Carbonate and Sevelamer Carbonate on Mineral Metabolism Disorders and Vascular Calcifications in Uremic Rats

O. Phan[1], M. Maillard[1], F. Funk[2], M. Burnier[1]
[1]Lausanne, [2]St. Gallen

Purpose: The present study compared the efficacy of PA21 with lanthanum carbonate (La) and sevelamer carbonate (Se) on hyperphosphatemia, secondary hyperparathyroidism and vascular calcification in rats with chronic renal failure (CRF).
Methods: CRF was induced by feeding a 0.75% adenine-enriched high phosphorus (P 1.3%) diet for 4 weeks. Rats were randomized to one of 3 binder treatment groups (PA21, La and Se) or to CRF and non-CRF controls for another 4 week period. The concentration of each binder (% of binder added to the diet) was chosen to deliver approximately the same amount of active pharmaceutical ingredient to each rat: PA21 5% (corresponding to 1% iron), La 2% (1% lanthanum), Se 1.5% (1% sevelamer). A computer-assisted automated quantitative measurement evaluated the calcification from von Kossa stained vessel sections. Data were expressed as the relative proportion (%) of calcified to total surface area of each vascular ring.
Conclusion: PA21 is as effective as La and Se to control hyperphosphatemia and secondary hyperparathyroidism. The extent of vascular calcifications in all phosphate binder treated animals was significantly lower than in the CRF control animals. In the upper part of the thoracic aorta, PA21 was even more efficient than La to prevent calcifications.

Results:	N	Creat µmol/l	P mmol/l	iPTHpg/ml	Abdominal Aorta %	Inferior Thoracic Aorta %	Superior Thoracic Aorta %
Non CRF control	4	48 ± 3	2.09 ± 0.05	275 ± 38**	0	0	0
CRF control	20	144 ± 11*	3.30 ± 0.29	3567 ± 593	1.25 ± 0.66	3.22 ± 1.31	8.05 ± 2.01
CRF PA21	19	141 ± 10	2.06 ± 0.06**	1459 ± 242**	0.10 ± 0.06**	0.34 ± 0.21**	0.14 ± 0.13**,***
CRF Se	20	147 ± 11	2.51 ± 0.12**	1569 ± 238**	0.53 ± 0.40	0.55 ± 0.38**	1.59 ± 0.91**
CRF La	18	140 ± 8	2.24 ± 0.07**	1360 ± 170**	0.20 ± 0.15	0.39 ± 0.27**	3.93 ± 1.13**

Values are shown as mean ± SD, *p <0.05 vs non-CRF control, ** p <0.05 vs CRF control, ***p <0.05 vs CRF La

P62

Susceptibility of Podocytes to Palmitic Acid Is Regulated by LXR-Dependent Stearoyl-CoA Desaturases 1 and 2

J. Sieber[1], K. Kampe[1], M. Lindenmmeyer[2], C. D. Cohen[2], P. Mundel[3], A. W. Jehle[1]
[1]Basel, [2]Zurich, [3]Boston, MA/US

Purpose: Type 2 diabetes mellitus is associated with elevated free fatty acid (FFA) levels. We reported effects of saturated FFAs (SFAs) and monounsaturated FFAs (MUFAs) for podocyte survival, with SFAs being deleterious and MUFAs protective. Here, we elucidate expression of stearoyl-CoA desaturases (SCDs) in glomeruli of diabetic nephropathy (DN), and whether induction of SCDs prevents palmitic acid-induced podocyte death.
Methods: Immunohistochemistry of SCD-1 in glomeruli of patients with type 2 diabetes and controls. Human glomerular gene expression of SCD-1 was studied by microarray analysis. Apoptosis and necrosis was assessed by staining with annexin V and PI in murine podocytes. Knockdown and overexpression studies of SCDs were performed in podocytes using a lentiviral system. Incorporation studies were performed using [3H]palmitic acid and TLC.
Results: By immunohistochemistry, SCD-1 in glomeruli was prominent in podocytes only. Gene expression analysis revealed a significant induction of SCD-1 in diabetic glomeruli. The effect of SCDs was investigated by LXR-agonists TO901317 (TO) and GW3965 (GW), known SCD-inducers. TO induced Scd-1 (2.5x) and Scd-2 (5.0x) mRNA and reduced palmitic acid-induced apoptosis and necrosis by 40% and 30%. Results with GW were similar. Only combined gene silencing of Scd-1 and Scd-2 reverted the TO-effect, indicating a causative role for both SCD-isoforms. In addition, Scd-1-overexpression ameliorated survival of palmitic acid-treated podocytes. Finally, TO shifted palmitic acid-derived FFAs into biologically inactive triglycerides.
Conclusion: These results indicate a protective effect of SCDs on palmitic acid-induced podocyte death. SCD-1 in podocytes of patients with DN may be part of a protective mechanism against SFAs.

P63

Spleen Tyrosine Kinase Is Important in the Production of Proinflammatory Cytokines and Cell Proliferation in Human Mesangial Cells following Stimulation with IgA1 Isolated from IgA Nephropathy Patients

M. J. Kim[1], J. Barratt[2], K. Molyneux[2], C. D. Pusey[3], F. W. K. Tam[3]
[1]Basel, [2]Leicester/UK, [3]London/UK

Purpose: We previously reported that the inhibition of spleen tyrosine kinase (SYK) by a SYK inhibitor, R406 (fostamatinib), or SYK siRNA reduces the synthesis of various cytokines by human mesangial cells (HMC) following stimulation with IgA1 isolated from the serum of IgA nephropathy (IgAN) patients (pIgA1). We now examine whether SYK is involved in mesangial cell proliferation and production of extracellular matrix (fibronectin) following stimulation with pIgA1.
Methods: IgA1 was purified from the serum of IgAN patients and aggregated at 63 °C for 150 min (aIgA1). HMC were incubated with aIgA1 for 24h and cell proliferation assay with BrdU was performed. We then incubated HMC with R406, 1h before stimulation with aIgA1 (200 µg/mL). HMC were then transfected with either Syk siRNA or negative control siRNA, 72h before stimulation with aIgA1. In the next experiment, human fibronectin produced by HMC following stimulation with pIgA1 for 24h was examined in culture supernatants by ELISA. HMC were then incubated with R406, 1h before stimulation with pIgA1 (50 µg/mL).
Results: The proliferation of HMC was increased upon stimulation with aIgA1 and inhibited by R406 in a dose dependent manner. HMC transfected with SYK siRNA proliferated significantly less than the cells transfected with negative control siRNA. The concentration of human fibronectin in culture supernatants increased significantly following stimulation with pIgA1. The preincubation with R406 did not reduce the concentration of fibronectin.
Conclusion: Our previous and current data suggest the involvement of SYK in HMC in the production of various cytokines and mesangial cell proliferation, but not in the synthesis of fibronectin, upon stimulation with pIgA1. SYK may be considered as a potential target in the treatment of IgAN.

P64

Functional Study of SLC2A9 (Glut9) Variants Associated With Plasma Uric Acid Level

A. Ruiz, O. Bonny
Lausanne

Purpose: Uric acid is the end product of purine metabolism in humans. GLUT9 has been identified as a new voltage-sensitive urate transporter expressed in the liver, the kidney and other tissues. In humans, GLUT9 loss-of-function mutations (L75R, T125R, R198C, R380W) have been associated with familial renal hypouricemia, a

condition associated with low plasma uric acid levels, uric acid kidney stones and exercise-induced acute renal failure. Moreover, a non-synonymous single-nucleotide polymorphism (SNP) V253I, present with a frequency of 22.3% in the population, has been associated with low plasma uric acid level. By contrast, a loss-of-function mutation identified in the Dalmatian dog strain (C210F) leads to hyperuricemia, kidney stone formation and renal insufficiency. Ongoing debates about functional studies of some GLUT9 variants and the absence of functional data of others have prompted us to study all of them.
Methods: Xenopus oocytes were injected with various GLUT9 variants cRNAs and were subjected to 14C-urate uptake (n = 30 oocytes, 3 batches) and surface expression was assessed by immuno-staining.
Results: The uptake assay revealed a decrease of 14C-urate flux for all the mutants compared to wildtype GLUT9 (69.8 ± 8.4% L75R; 85.6 ± 8.0% T125R, 58.4 ± 21.1% R198C, 72.3 ± 13.8% R380W for human mutants and 80.1 ± 5.1 C210F, p <0.01 for all compared to WT) with conserved expression at the cell surface. The SNP V253I showed a decreased14C-urate uptake by 41.2 ± 22.2% (p <0.05) related to a decreased surface expression.
Conclusion: These results represent the first complete characterization of known GLUT9 variants and pave the route toward a better understanding of the structure-function relationship for the urate transporter GLUT9.

P65

Role of the Glucocorticoid Receptor in Podocyte Function?

I. Auberger, M. Lindenmeyer, M. Sorensen, Y. Sugano, U. Ziegler, J. Loffing, C. D. Cohen
Zurich

Purpose: In proteinuric diseases glucocorticoids (GC) show a prompt anti-proteinuric effect in steroid sensitive patients. However, central aspects of the underlying mechanisms of GC's action are still incompletely understood and both local effects on podocytes as well as systemic immune-modulating properties are discussed. Podocytes, which play an important role in nephrotic syndrome, express the glucocorticoid receptor (GR). Hence we hypothesize that GC control via the GR a transcriptional program that is critical for glomerular development, structure and function.
Methods: Datasets of the European Renal cDNA Bank allowed us to study the overall expression of GR-dependent gene transcripts in glomeruli of patients with minimal change disease, focal segmental glomerulosclerosis or membranous glomerulonephritis. Candidate genes were selected and further studied in an immortalized murine podocyte cell-line treated with dexamethasone. For further investigations a mouse model with a target deletion of the GR in podocytes was created.
Results: Glomeruli of patients with nephrotic syndrome showed lower expression of several reported GR-dependent gene transcripts than control tissue. A dose- and time dependent up-regulation of FKBP5 and DUSP1, two of these GR-dependent glomerular transcripts, by GC in podocytes could be documented. Podocyte-specific-GR-knockout mice exhibit protrusions of the glomerular basement membrane, collagen bundles, podocyte foot process effacements as well as changes of endothelial cells.
Conclusion: Collectively, these findings underline a link between the GR and glomerular function. Further studies are underway to understand the molecular function and the detailed phenotype of the podocyte-specific GR-knockout.

P66

Quantification of Multiple Bile Acids Using Ultra-Performance Liquid Chromatography-Tandem Mass Spectrometry: Impact of Uninephrectomy on Circulating Bile Acids in Rats

C. A. Penno[1], D. Arsenijevic[2], T. Da Cunha[1], G. Kullak-Ublick[2], J.-P. Montani[2], A. Odermatt[1]
[1]Basel, [2]Freiburg, [3]Zurich

Purpose: Bile acids (BAs) are end products of cholesterol catabolism and act as emulsifiers of lipophilic compounds. Besides, they were recently recognized as important signaling molecules. To understand the roles of individual BAs and due to limited blood sample volumes available from experimental animals, improved methods for the simultaneous quantification of multiple BAs are needed.
Methods: We developed and validated an ultra-performance liquid chromatography tandem mass spectrometry (UPLC-MS/MS) method for the quantification of 24 BAs, including 11 unconjugated, 6 glycine-conjugated and 7 taurine-conjugated BAs, in 50 µL of rat serum or plasma. The application of UPLC-MS/MS allows a highly specific detection of BAs by using multiple-reaction monitoring (MRM) with specific fragmentation. The method showed acceptable intra- and inter-day accuracy, precision, extraction recovery and enhanced sensitivity compared with earlier approaches. We applied the established method to assess time-dependent changes of BAs in plasma from sham-operated and uninephrectomized male Sprague-Dawley rats.
Results: The levels of several primary and secondary BAs were transiently elevated one week after uninephrectomy, followed by normalization after the second week. In contrast, several conjugated BAs showed increased levels at the second week post-surgery, followed by normalization thereafter.
Conclusion: The established UPLC-MS/MS method allows the simultaneous and specific quantification of multiple BAs in 50 µL serum or plasma samples. Application of the method revealed a transient increase of several primary and secondary BAs in uninephrectomized rats that was followed by a transient increase in conjugated BAs. The presented method can be used to assess BA profiles in various patho-physiological situations.

P67

Roles of Claudins and Zonula Occludens in Epithelial Collecting Duct Cell Senescence

X. Qiao, E. Dizin, I. Roth, E. Feraille, U. Hasler
Geneva

Purpose: Repair of kidney epithelia following acute injury depends on rapid cell proliferation, which is low under normal conditions. Since hypertonicity reduces cell division and promotes cell senescence, cell proliferation in the renal medulla is likely even lower. Recent evidence indicates that cell repopulation is achieved by differentiated tubular cells that retain an intrinsic ability to divide. This implies that loss of contact between neighboring cells might promote surviving cells to dedifferentiate and reenter the cell cycle. This led us to examine the roles of claudins and zonula occludens (ZO) as modulators of cell proliferation under both isotonic and hypertonic conditions.
Methods: Proliferation of principal collecting duct (CD) mCCDcl1 cells grown under isotonic and hypertonic (500 mOsoml/kg) conditions was examined by microscopy analysis and FSC cell sorting. Claudin and ZO expression was examined by Real-Time PCR, Western blot and confocal analysis.
Results: mCCDcl1 cell proliferation under isotonic conditions decreased with cell confluency. This was accompanied by a reduced S phase and increased G0/G1 phase. While ZO-1 and ZO-2 abundance was unaffected by cell confluency, both ZO-3 and claudin-8 abundance increased while claudin-4 abundance decreased with cell confluency. All observed changes were accelerated in cells grown in hypertonic medium. Microscopy analysis further revealed increased ZO-1 abundance by chronic hypertonic challenge and increased ZO-1 expression in medullary CD.
Conclusion: Together, these results indicate that CD cell senescence is associated with increased claudin-8 and ZO expression. Modulated expression of TJ components will help establish their roles in cell proliferation and paracellular transport.

P68

The Calcium Channel TRPC6, Known to Cause FSGS When Chronically Hyperactive, Protects Podocytes From Acute Complement-Mediated Injury

A. Kistler[1], G. Singh[2], J. W. Pippin[3], S. J. Shankland[3], J. Reiser[4]
[1]Zurich, [2]New Delhi/IN, [3]Seattle/US, [4]Miami/US

Purpose: Gain-of-function mutations in the calcium channel TRPC6 lead to genetic FSGS. Podocyte expression of normal TRPC6 is increased in acquired human glomerular diseases, particularly in membranous nephropathy (MN). We therefore speculated that overexpression of TRPC6 in cultured podocytes leads to cell damage.
Methods: We used standard methods, including podocyte culture, lentiviral gene transfer, Ca-imaging and cell surface biotinylation.
Results: Overexpression of TRPC6 in differentiated podocytes did not affect podocyte integrity despite correct membrane localization and activity of the channel. Unexpectedly, overexpression of TRPC6 protected podocytes from complement-induced injury, an in vitro model of MN. In contrast, overexpression of dominant-negative TRPC6, knock down of TRPC6 and the administration of a TRPC antagonist increased podocyte sensitivity to complement. This effect was mediated by CaMKII: complement attack activated CaMKII in podocytes and the degree of activation correlated with TRPC6 levels. Pretreatment of podocytes with a CaMKII inhibitor phenocopied the effect of TRPC6 inhibition. Human MN biopsy samples, where induced TRPC6 expression has been previously shown, displayed increased activity of CaMKII. In the nephrotoxic serum nephritis model, where complement contributes to glomerular injury, podocyte-specific TRPC6 transgenic mice showed stronger CaMKII activation, reduced podocyte FP effacement and reduced levels of proteinuria, whereas TRPC6 knock out mice exhibited reduced CaMKII activation and higher levels of proteinuria compared to wt controls.
Conclusion: These data suggest an unexpected dual role of TRPC6 in podocytes: whereas chronic hyperactivity leads to FSGS, acute activation protects from complement-mediated damage in the short term.

P69

Solid Organ Fibrosis: Evidence for a Common Pathway Across Species!

A. Anagnostopoulou[1], A. Scherer[2], A. Jeffs[3], J. Bedford[3], J. Leader[3], R. Walker[3], H.-P. Marti[1]
[1]Berne, [2]Kontiolahti/FI, [3]Dunedin/NZ

Purpose: We described a transcriptomic classifier of metzincins and related genes (MARGS) discriminating renal allografts and other solid-organs with or without fibrosis *(AJT, 2009; Virchows Arch, 2011)*. Rats exposed to lithium are known to develop fibrosis *(Nephrology, 2010)*. In this study, we wanted to demonstrate, if our MARGS-based algorithm has diagnostic value in rat renal fibrosis.
Methods: Male Wistar rats (n = 12) were divided into a control group (n = 6), and an experimental group (n = 6) that received 40 to 60 mmol lithium carbonate/kg dry food up to 24 weeks. After six months, animals were sacrificed to dissect cortex and medulla. We used 24 Affymetrix Rat Exon 1.0 ST arrays: healthy cortex (n = 6), healthy medulla (n = 6), lithium exposed cortex (n = 6) and lithium exposed medulla (n = 6). Three MARGS were examined by immunofluorescence.
Results: There were more differentially expressed genes in medulla-dataset than in cortex-dataset (ANOVA). MMP-2, CD44 and TGFβ2 were up-regulated in both lithium-treated cortex and medulla samples. In gene set analyses (GSEA), lithium-treated cortex and medulla samples showed enrichment of MARGS, TGFb, ECM and fibrosis genes; lithium-treated medulla samples were also enriched in immune response pathways. The MARGS based IF/TA classifier was able to classify all samples correctly. Ingenuity pathway analysis of diffierentially regulated genes in medulla depicts relationship within MARGS and with respective mi-RNA. Immunofluorescence confirmed up-regulation of MMP-2, CD44 and TGFβ2.
Conclusion: Our MARGS classifier represents a cross-organ and cross-species classifier of fibrotic conditions irrespective of etiology and may help to design a low density array (LDA) to diagnose and to monitor fibrosis. These results provide evidence for a common pathway in the pathogenesis fibrosis.

P70

Epha2 Receptors Contribute to the Renal Tubular Response to Hypoxic Injury

K. Franziska Koenig[1], S. Rodriguez[2], S. Ravikumar[2], C. Rosenberger[3], U. Huynh-Do[2]
[1]Basel, [2]Berne, [3]Berlin/DE

Purpose: Acute Kidney Injury (AKI) is a major challenge to the nephrologist, and regional hypoxia is believed to play an important role irrespective of the underlying conditions. To identify novel mechanisms involved in the kidney response to hypoxic injury,
Methods: we performed segmental renal artery branch ligation in rats, a model which has been shown to induce an oxygen gradient vertical to the corticomedullary axis. Three distinct zones can be distinguished: (1) tubular necrosis, (2) infarction border zone, (3) preserved normal tissue.
Results: In previous work we showed that in the mouse skin, local oxygen deprivation triggered upregulation of Eph receptors, a family of receptor tyrosine kinases required for somitogenesis, vasculogenesis and axonal guidance in the embryo, and playing a central role for the homeostasis of many organs in the adult. In control kidneys, EphA2 receptor was expressed in tubular cells of Henle's loop, and its ligand ephrinA1 in endothelial cells of the glomeruli and vessels. Hypoxia induced HIF-1α stabilization in the infarction border zone mainly. In this area, EphA2 receptor was upregulated in tubular cells, while ephrinA1 expression increased in neighboring interstitial cells. This coordinated upregulation in adjacent cells highly suggested that these processes would trigger juxtacrine signalling. We showed that in MDCK cells, endogenous EphA2 expression significantly increased following hypoxia. Stimulation of MDCK with ephrinA1/Fc enhanced cell adhesion and deposition of laminin, an important component of the tubular basement membrane.
Conclusion: Our findings present evidence that EphA2 receptors may contribute to the tubular response to hypoxic damage by influencing the extracellular matrix composition and increasing cell-matrix interactions at the sites of injury.

Poster Presentations – NCCR Kidney.CH

P71

Coordinated Control Of Basolateral Na,K-ATPase and Tight-Junctions in Response to Apical Sodium Entry in Collecting Duct Cells

Y.-B. Wang[1], V. Leroy[2], T. Ernandez[1], A. Maunsbach[3], P.-Y. Martin[1], E. Feraille[1]
[1]Geneva, [2]Lille/FR, [3]Aarhus/DK

Purpose: CD principal cells are exposed to large variations of Na transport. Na crosses the apical membrane via epithelial Na channels (ENaC) and is extruded into the interstitium by the Na,K-ATPase. Backflux of reabsorbed Na is prevented by tight-junctions. This study was designed to decipher the cross-talk between ENaC, Na,K-ATPase and intercellular junctions in CD cells.
Methods: Doxicyclin-inducible overexpression of γ-ENaC was performed in mCCD cells. Total and biotinylated cell surface proteins were detected by Western-blot. mRNA levels were measured by RT PCR.
Results: γ-ENaC overexpression increased transepithelial Na current and expression of both total and cell surface Na,K-ATPase. Pulse-chase experiments with 35S-Methionine revealed that Na,K-ATPase synthesis was unchanged while its degradation was decreased. Pulse-chase labelling of cell surface proteins demonstrated that Na,K-ATPase endocytosis was decreased. γ-ENaC overexpression inhibited p38 kinase activity and endocytosis of Na,K-ATPase was decreased in response to p38 kinase inhibitors. Overexpression of γ-ENaC also increased transepithelial resistance in relation with increased expression of claudin-8 mRNA and protein but did not alter expression of E-cadherin.
Conclusion: We found that increasing apical Na entry via ENaC increased activity and cell surface expression of Na,K-ATPase through inhibition of p38 kinase. This cross-talk between apical and basolateral Na transport may prevent variations of intracellular Na. Transepithelial resistance is increased via incorporation of newly synthesized claudin-8 in tight-junctions that is independent of p38 kinase. This remodeling of tight junctions may prevent backflux of reabsorbed Na. In contrast E-cadherin-dependent intercellular junctions are not altered in response to transepithelial flux.

P72

Effects of Renal Dysfunction on Bile Acid Homeostasis

G. Zhibo[1], G. Kullak-Ublick[1], J.-P. Montani[2], C. Lei[3]
[1]Zurich, [2]Freiburg, [3]Jinan/CN

Purpose: Although the kidney is believed to play a minor role in bile acid (BA) excretion, chronic renal failure (CRF) has been reported to be associated with increased serum bile acid levels and alterations in the BA balance. This study was designed to examine the effects of naturally progressing CRF of longer duration on gene expressions of the key factors involved in hepatic bile acid synthesis and transport, i.e., Cyp7A1, Ntcp, Bsep.
Methods: Wistar rats were randomized to the CRF group (5/6 nephrectomy) and sham-operated, placebo-treated normal controls. They were allowed free access to regular rat chow and studied 8 weeks after surgery. Uninephrectomied (UNX) rats were also used to examine the impact of renal functions on bile acid metabolism. Liver mRNAs and protein mass or activities of the above factors were studied.
Results: The CRF group exhibited significantly increased plasma cholesterol concentration and bile acid levels. Hepatic Cyp7a1 mRNA, and Cyp7a1 protein mass measurements were virtually identical in the two groups. Examine on bile acid transporters showed elevated Mrp3, Ost-α and Ost-β expressions at both mRNA and protein levels, indicating a shift of bile acid transport from apical canaliculi to basolateral blood. Similar changes of plasma bile acid level and bile acid transporters were found in UNX rats.
Conclusion: In summary, chronic renal failure is associated with a strong increase in plasma bile acid levels, which is shown to be an early event before the time when kidney function is affected. Maintenance of bile acid synthesis and elevated basolateral Mrp3 and Ost-α/β expressions may either be a desired response during chronic renal disease to raise serum bile acid concentration or it may be a failing feedback regulation on bile acid formation and disposition.

P73

Flow-Mediated Regulation of Sodium Transport in the Collecting Duct

T. Ernandez, A. Chassot, P.-Y. Martin, E. Feraille
Geneva

Purpose: Na transport in renal tubules is tightly controlled and plays a central role in of body fluid homeostasis. In addition to the classical neuro-endocrine regulatory inputs, local factors such as luminal flow may participate to Na homeostasis.
Methods: We designed an in vitro experimental setting to explore the effect of apical flow on a cellular model of collecting duct (CD) using the well-described mouse mCCDcl1 principal cells grown on filters. Directional flow was generated using an orbital shaker delivering a shear-stress of 2 dyne/cm^2 mimicking physiological luminal flow.
Results: We observed a delayed and sustained decrease of the amiloride-sensitive Na current in cells subjected to flow reaching a plateau at 8h (40% decreased). This was correlated with a significantly decreased mRNA expression of ENaC subunits and SGK1. The flow-mediated Na transport decrease was not prevented by PKD1 or KIF3A genes silencing, excluding a role of the primary cilium in this response. This unique organelle protruding on the apical side of CD cells is indeed described as a putative mechanosensor. To obtain more insights on factors involved in ENaC flow-mediated regulation, we performed a whole-genome transcriptional analysis in mCCDcl1cells subjected or not to flow. Significant down-regulation of genes involved in PKA and Rho GTPases pathways were identified. We speculate therefore that shear-stress alters cAMP pathway and cytoskeleton dynamics that are involved in Na transport regulation. Preliminary data using PKA inhibitor suggest a central role of PKA in the flow-mediated regulation of Na transport. We are currently investigating this avenue.
Conclusion: Flow-mediated regulation of Na transport might be of particular relevance in increased glomerular blood flow conditions such as in living kidney donors.

P74

Hypoxia-Associated Gene Transcripts are Altered in Acquired Nephropathies

M. Lindenmeyer, D. Hoogewijs, D. Stiehl, R. Wenger, H. Moch, P. Wild, C. D. Cohen
Zurich

Purpose: Most chronic kidney diseases (CKD) are initiated as glomerular damage with loss of glomerular capillaries. The pathogenesis of the glomerular insult can be manifold. The best morphologic indicator of disease progression and development of end-stage renal disease, however, is interstitial fibrosis accompanied by a capillary rarefaction. As hypoxia – a potential consequence of the capillary rarefaction – has been associated with fibrosis the question arises whether renal cells indeed face hypoxia in CKD and respond with a transcriptional program which could lead to disease progression.
Methods: Expression of hypoxia-associated genes was assessed in genome-wide expression profiles from more than 160 renal biopsies from patients with different CKD stages. Proximal tubular cells and podocytes with stable HIF1 and/or HIF2 suppression were generated.
Results: From a total of 84 established HIF-target genes 27 correlated with renal function (eGFR) in the cortical tubulointerstitium and 22 in glomerular samples. Importantly, these correlations were both positive and negative and in part compartment-specific. The celltype-specific response to hypoxia was tested by qPCR in the knock-down derivates and revealed specific HIF1/HIF2-dependencies in the different cell lines. To validate the results on protein level we are currently establishing immunohistochemistry of HIF-target genes in human biopsies from patients with a wide range of renal function.
Conclusion: Our gene expression studies do not indicate an over-all hypoxic milieu in acquired kidney diseases. However, the data clearly point to compartment- and celltype-specific dysregulation of hypoxia-associated genes in CKD. Elucidation of the mechanisms involved may help to understand the pathogenesis of anemia in CKD, interstitial fibrosis, and renal failure.

P75

Impact of Chronic in Utero Hypoxia on Renal Glomerulogenesis and Tubulogenesis

M. Janot, P. Boissier, S. Rodriguez, U. Huynh-Do
Berne

Purpose: Chronic kidney diseases (CKD) represent a growing public health problem, due to the aging population and higher prevalence of the metabolic syndrome. Recent studies have also suggested the role of early events in life. Intrauterine Growth Restriction (IUGR), resulting from an adaptation to inadequate supply of oxygen and/or nutrients during pregnancy, is thus thought to be responsible for adult hypertension, insulin resistance, cardiovascular and renal diseases. Studies of IUGR are still scarce and the molecular actors responsible for a deficient nephrogenesis remain to be better characterized. Our goal is thus to study the impact of chronic exposure to hypoxia on kidney development, using a mouse model of chronic hypoxia in utero.
Methods: Pregnant mice were exposed to hypoxia (9.5% vs. 21% O_2) during renal development (E11.5 to D7) with quantification of food intake for caloric adjustment (control group). Kidneys from pups were collected at E18.5 and analyzed.
Results: First experiments (E14.5 to E18.5) showed a decreased food intake by hypoxic dams with no reduction in litter sizes. Pups from hypoxic dams showed a significantly lower birth weight compared to pups from normoxic dams (with or without adjusted caloric intake). Microarray and qPCR analyses of E18.5 kidneys showed a modified expression of genes mostly implicated in coagulation, lipid metabolism and vascular calcification. Morphometric and immunohistochemistry analyses are ongoing.
Conclusion: This study gives new insights into the mechanisms linking IUGR and abnormal kidney development and identifies potential molecular actors implicated in this process.

P76

The Furosemide-Induced Increase of Plasma Parathyroid Hormone is Mediated by the Calcium-Sensing Receptor in Humans

M.-E. Muller, V. L. Forni, C. Zweiacker, M. Maillard, O. Bonny, M. Burnier
Lausanne

Purpose: Furosemide has been reported to increase intact plasma parathormone (iPTH) levels in humans, but the mechanisms of this interaction are still unknown. Experiments on rats suggested that acute administration of a calcimimetic blunts this effect. We designed a prospective randomized placebo-controlled crossover study addressing the role of the calcium sensing receptor in the iPTH response to furosemide.
Methods: 12 Caucasian, non-smoker healthy males were enrolled. After 3 days of a fixed salt diet, they received either a single dose of 60 mg cinacalcet or placebo with at least one week interval. Three hours after cinacalcet, 20 mg furosemide were given iv. Plasma levels of iPTH and plasma and urinary levels of calcium, sodium and potassium were measured at baseline (before cinacalcet), before and at regular time intervals after the furosemide injection .
Results: Plasma iPTH levels were suppressed (38.0 ± 12.0 ng/l vs 2.4 ± 1.7 ng/l, $p < 0.05$), and calciuria was increased 3 h after administration of cinacalcet. Under placebo, a sharp increase in plasma iPTH levels was seen as soon as 15 min after furosemide injection (from 20.9 ± 6.6ng/l before to 33.2 ± 10.7 ng/l, mean ± SD), whereas under cinacalcet, iPTH response was blunted (from 2.4 ± 1.7 ng/l to 3.2 ± 2.9 ng/l, mean ± SD). Furosemide induced a significant decrease in plasma ionized calcium in cinacalcet-treated subjects, an effect which was absent under placebo. The changes in plasma Na and K after furosemide were comparable in both cinacalcet and placebo groups.
Conclusion: These data show in humans that furosemide acutely stimulates iPTH an effect which is blunted by the administration of a calcimimetic despite a decrease in plasma ionized calcium. Changes in sodium and potassium levels do probably not play any role in the iPTH response to furosemide.

P77

Oxygen-Regulated Expression of Erythropoietin in Cellular Models

F. Storti[1], I. Abreu-Rodriguez[1], S. Frede[2], J. Fandrey[2], R. Wenger[1], D. Hoogewijs[1]
[1]Zurich, [2]Essen/DE

Purpose: Erythropoietin (Epo), the key hormone regulating red blood cells homeostasis, is mainly produced in the adult kidney in response to hypoxia and anemia. Epo is regulated by the prolyl-hydroxylases (PHDs)/von Hippel-Lindau protein (VHL)/hypoxia-inducible factors (HIFs) pathway, but its tissue-specific induction remains largely unknown, mainly due to the lack of a kidney-derived cellular model capable of expressing Epo in a hypoxia-inducible manner. Recently, a new renal cell model (called Renal Epo Producing Cells, REPCs) became available and we started the characterization of this cell line. Moreover, Epo overexpression can be the cause of erythrocytosis, a disease in which the number of red blood cells is increased. Mutations in PHD2, VHL or HIF2A have been reported in patients with secondary congenital erythrocytosis. We aim to functionally characterize recently identified mutations found in the PHD2 gene of patients with erythrocytosis.
Methods: We are currently using REPCs, and the hepatoma cell lines HepG2 and Hep3B to explore the role of different players of the PHDs/VHL/HIFs pathway in Epo transcriptional regulation, by a shRNA-mediated knockdown strategy. HIF transcriptional and stability assays, as well as mutant PHD2 overexpression in PHD2-silenced cells, are used to assess the functional effect of the novel PHD2 mutations.
Results: Besides HIF-2α, a novel transcription factor belonging to the

ETS family was found to have a strong effect on Epo transcriptional regulation in REPCs. Mutations in PHD2 lead to functional differences regarding HIF regulation.
Conclusion: The presented in vitro approach enabled us to identify a novel factor regulating oxygen-dependent Epo expression and functionally investigate erythrocytosis-associated PHD2 mutations.

P78

Rapid Homeostatic Effects of Oral Potassium Loading on the Kidney

S. Grossmann[1], M. Sorensen[1], M. Rösinger[1], D. Loffing-Cueni[1], G. Barmettler[1], U. Ziegler[1], A. Odermatt[2], O. Staub[3], J. Loffing[1]
[1]Zurich, [2]Basel, [3]Lausanne

Purpose: A large dietary potassium (K+) load is a homeostatic challenge for mammals. It is known to induce a rapid kaliuretic and natriuretic response. These renal effects are reported to occur even before plasma K+ and aldosterone levels increase. Here we elucidate the underlying molecular mechanisms of K+ induced kaliuretic and natriuretic response.
Methods: We analyzed in mice the time course (15', 30', 2h, and 6h) of the effect of a gastric K+ load on plasma ion concentrations, aldo levels, urinary ion excretion, and expression and/or phosphorylation of renal ion transport proteins.
Results: Following a gastric gavage of 2% KCl, plasma K+ concentrations rose rapidly (at 15'), followed by a significant rise of plasma aldo (at 30'). Enhanced urinary K+ and Na+ excretion was detectable as early as spot urines could be collected (~30'). The functional changes were accompanied by a rapid and sustained dephosphorylation of the NaCl cotransporter (NCC) (15'-6h) and a later up-regulation of proteolytic activated epithelial sodium channels (ENaC) (6h). The rapid effect on NCC and the late effects on ENaC were independent from the co-administered anion (same effect with KHCO3; no effect with NaCl). In contrast to the proteolytic ENaC regulation, NCC dephosphorylation was independent of plasma aldo as indicated by experiments in aldo-deficient mice. The observed urinary Na+ loss was likely related to NCC, as it was not seen in NCC-deficient mice.
Conclusion: Rapid down-regulation of NCC contributes to the early kaliuresis and explains the natriuresis in response to an oral K+ load. Enhanced activation of ENaC occurs quite late and might be more important for the long-term control of K+ homeostasis.

P79

V-Atpase B1 Subunit Polymorphism P.E161K Affects Urinary Acidification in Vivo

N. Dhayat, A. Pasch, D. Fuster
Berne

Purpose: The V-ATPase proton pump on the luminal membrane of α-intercalated cells is critical for urinary acidification. The V-ATPase consists of two multi-subunit domains, the V0 and V1 domain. The soluble cytosolic 640 kDa V1 domain is composed of subunits A-H in a A3B3C1D1E1F1G2H1 stoichiometry. In humans, there are two different isoforms of the B subunit in the V1 domain, of which B2 is ubiquitous whereas B1 is restricted to specialized epithelia of the inner ear, epididymidis and the distal renal tubule. Mutations in the B1 subunit gene ATP6V1B1 cause autosomal-recessive distal renal tubular acidosis. We recently identified a polymorphism in the human V-ATPase B1 subunit (p.E161K) that greatly diminished pump function in vitro (Fuster, Moe et al., Kidney Int 2008).
Methods: To study the impact of the p.E161K polymorphism on acidification in humans in vivo, we conducted a retrospective analysis in our renal stone patient registry. Exon 6 of the ATP6V1B1 gene was sequenced in all patients bi-directionally.
Results: Patients heterozygous for the p.E161K polymorphism (n = 19) had higher urinary pH and lower urinary citrate excretion in 24 hr urines than patients carrying two wild-type alleles (n = 547). Blood pH, pCO_2 and HCO3- and 24 hr sulphate excretion (measure of protein intake) were not different between the two groups of patients. Furthermore, acid challenging experiments (ammonium chloride loading tests) revealed a pathological urinary acidification response in a so far limited number of p.E161K carriers (n = 3).
Conclusion: Thus, our preliminary data are compatible with the phenotype of incomplete distal tubular acidosis in p.E161K polymorphism carriers. Clearly, however, much more work is needed to clarify the role of this polymorphism in human physiology and pathophysiology.

P80

Role of Oxygen-Inducible PAG1 in Chronic Kidney Disease

S. Santambrogio[1], M. Lindenmeyer[1], A. Schorg[1], F. Storti[1], J. Schodel[2], D. R. Mole[2], C. D. Cohen[1], R. Wenger[1], D. Hoogewijs[1]
[1]Zurich, [2]Oxford/UK

Purpose: Accumulating evidence exists that hypoxia is an important modulator of chronic kidney disease (CKD) and the identification of novel hypoxically regulated genes will improve our understanding of the transcriptional mechanisms involved in CKD. Recently, we discovered PAG1(Phosphoprotein Associated with Glycosphingolipid enriched microdomains) as a novel hypoxia-inducible gene. PAG is exclusively localized in lipid rafts, plays a crucial role in the regulation of Src-kinase family and is involved in several signaling pathways. The aim of our work is to understand the role of PAG in kidney pathophysiology.
Methods: RT-qPCR, immunoblotting and ChIP experiments were conducted to study hypoxia-dependent PAG regulation.
Results: PAG protein levels were robustly induced by hypoxia in non-malignant cells whereas 786-O showed high expression levels in normoxia and reconstitution of VHL as well as shRNA-mediated HIF-2α knock-down resulted in decreased PAG expression levels. Moreover, in *vivo* experiments confirmed hypoxically induced PAG mRNA levels in kidneys of mice exposed to 9% O_2. Interestingly, ChIP-qPCR experiments provided evidence for HIF-2α/HIFβ binding in 786-O and this putative enhancer site is localized 85 kb upstream of the PAG1 promoter, suggesting a novel mode of hypoxic gene regulation. A comprehensive screen of PAG expression in gene array data from glomerular and tubulointerstitial compartments of patients with progressive and non-progressive nephropathies revealed robust PAG induction in several glomerulopathies. The array data were confirmed by RT-qPCR in independent nephropathy samples.
Conclusion: Unraveling the role of hypoxic PAG regulation in a pathological context will add new insight in understanding CKD and will help to clarify its physiological role.

P81

Establishment of a Novel Genetically Modified Mouse Model Targeting the Renal Oxygen Sensing and EPO-Producing Cells

I. Abreu Rodríguez[1], P. Spielmann[1], M.-L. Dénéréaz[2], E. Hummler[2], A. Hesse[3], D. M. Katschinski[3], D. Hoogewijs[1], R. H. Wenger[1]
[1]Zurich, [2]Lausanne, [3]Goettingen/DE

Purpose: In CKD, renal oxygen consumption is decreased and the oxygen gradient disrupted, leading to a drop in erythropoietin (EPO) synthesis and anemia. Although the mechanisms underlying inducible expression of Epo are generally understood, the mechanisms of constitutive tissue-specific and inducible anemic/hypoxic Epo gene regulation are largely unknown. EPO is synthesized by insufficiently characterized peritubular interstitital cells. These cells cannot be cultured and novel models are hence urgently required to study the complexity of the renal oxygen signaling cascade and Epo regulation.
Methods: Transgenic Cre strains will be used to study the expression of EPO in a temporal and spatial manner, indirectly by reporter genes (eGFP and LacZ) and directly (by Cre-mediated VHL deletion); and to generate specific knock-outs of recently discovered novel members of the oxygen signaling cascade to investigate their role in (patho) physiological EPO regulation. Renal hypoxia imaging will be complemented by using transgenic ODD-luc mice.
Results: Generation of novel BAC transgenic Cre vectors expressing the Cre recombinase under the control of the mouse Epo gene locus. Following pronuclear microinjections, two potential founder lines were obtained and they are currently being analyzed.
Conclusion: These mice model will be used for detailed characterisation of the Epo-producing renal cells, analysing the physiologycal relevance of novel factors involved in oxygen signaling for EPO expression regulation and imaging the number and distribution of EPO producing cells during development, hypoxic insults and CKD.

P82

The Impact of Reduced Kidney Mass on Adipose Tissue Metabolism and Whole-Body Glucose Homeostasis in Mice

S. H. Chin, F. Item, S. Wueest, M. Wiedemann, E. J. Schoenle, D. Konrad
Zurich

Purpose: Reduced kidney function deteriorates insulin sensitivity in children and adults. However, the underlying mechanisms are poorly understood. Activation of the RAAS/angiotensin receptors (ATR) in adipose tissue impairs insulin signalling in adipose tissue, skeletal muscle and liver and its prevention by ATR blockade (pharmaceutically or genetically) improves glucose homeostasis. We therefore hypothesise that reduced kidney mass impairs glucose metabolism via activation of the RAAS.

Methods: Seven-week-old C57Bl6/J mice underwent uninephrectomy (UniNx) or sham operation. After operation, animals were fed either a chow (standard) or a high fat diet (HFD) and glucose homeostasis was assessed 2, 8, and 20 weeks after surgical intervention.
Results: No significant differences were observed in glucose tolerance in chow-fed animals. However, in HFD-fed animals, glucose tolerance was further impaired in UniNx mice after 8 and 20 weeks when compared to sham-operated mice. Moreover, skeletal muscle insulin resistance was significantly deteriorated and adiposity was increased in UniNx mice after 20 weeks of HFD. In contrast, hepatic steatosis was decreased and hepatic insulin sensitivity was improved in UniNx mice. Plasma angiotensin I concentration was elevated in UniNx compared to sham-operated mice under both chow and HFD 2, 8 and 20 weeks after surgical intervention. Moreover, expression of proinflammatory cytokines was decreased in both mesenteric and epididymal fat of UniNx compared to sham-operated mice after 20 weeks of HFD.
Conclusion: Uninephrectomy further impairs obesity-induced skeletal muscle insulin sensitivity but protects from obesity-induced adipose tissue inflammation as well as hepatic insulin resistance and steatosis.

P83

Uninephrectomy May Alter Immune And Metabolic Regulation: A Role for the Brain?

D. Arsenijevic[1], J. Plamondon[2], I. Scerri[1], J.-F. Cajot[1], D. Richard[2], J.-P. Montani[1]
[1]Freiburg, [2]Quebec/CA

Purpose: Uninephrectomy (UniNX) induced a small decrease in fat pads and a chronic elevation in markers of lipolysis (plasma glycerol, hormone-sensitive lipase, adipocyte triglyceride lipase). Increase in lipolysis was associated with increased levels of circulating cytokines known to be involved in lipolysis and body fat regulation (interferon-gamma, IFNg; granulocyte macrophage colony stimulating factor, GM-CSF) and acylation stimulating protein, ASP, rather than with changes in hormones such as T3, leptin, insulin or ghrelin. Does the brain play a role in lipolysis via these immune peptides?
Methods: To study the metabolic consequences of UniNX young male Sprague Dawley rats, fed on an isocaloric standard diet, were subjected to either sham-operation or UniNX, and sacrificed at selected time intervals (1, 2, 4 and 6 weeks) after surgery.
Results: In UniNX rats, receptors for the three immune peptides (IFNg, GM-CSF, ASP) were upregulated in the brain stem and hypothalamus at the mRNA level as early as 1 week after surgery and remained elevated throughout the study. Proteins for the IFNg and GM-CSF receptors were also present. mRNA levels of melanocortin 4 receptor (MC4R) were upregulated in the same brain areas. Other studies have shown that central MC4R activation can induce lipolysis in peripheral fat pads via an increase in fat pad phosphorylated hormone sensitive lipase (p-HSL), an increase we also observe in our model.
Conclusion: In summary, experimental UniNX in rats promotes lipolysis by mechanisms that seem to implicate central pathways involving cytokine receptors along with MC4R, signalling to fat pads to increase lipolysis. UniNX resulted in chronic activation of brain regulatory pathways associated with lipolysis. What other central pathways are modified by UniNX remains to be determined.

P84

Role of the Renal Mineralocorticoid Receptor for Potassium Homeostasis

M. Roesinger[1], D. Loffing-Cueni[1], C. Ronzaud[3], J. Canonica[3], O. Staub[3], E. Hummler[3], J. Loffing[1]
[1]Zurich, [2]Lausanne

Purpose: The mineralocorticoid hormone aldosterone (aldo) is released from the adrenal glands upon a dietary K+ load. Aldo enhances renal K+ excretion via ROMK channels and stimulates apical epithelial Na+ channels (ENaC) to provide the electrochemical gradient for K+ secretion. Here we studied knockout mice (MRflox/AQP2cre) with targeted inactivation of the mineralocorticoid receptor (MR) in the renal collecting system (CS).
Methods: KO as well as WT mice were loaded for 2 days with 5% K+. Blood pressure, food and water intake- and excretion was monitored. Kidneys were perfused and prepared for Immunoblotting (IB) or Immunohistochemistry (IHC), respectively. IB and IHC was performed using Antibodies against ENaC, ROMK, and the Sodium Chloride Co-transporter (NCC).
Results: The mice tolerate K+ loading without major illness, but show slightly reduced food intake indicating K+ avoidance. IB and IHC reveal a K+ diet-induced increase in ROMK protein abundance and apical localization that is similar for the CS of control and KO mice. However, KO mice show less K+ diet-induced α- and γENaC proteolytic activation and the apical translocation of ENaC is restricted to the remaining MR-positive late DCT. ENaC apical translocation is not seen in the MR-negative CS. Interestingly, KO mice have a reduced abundance and phosphorylation of NCC in the DCT. This NCC down-regulation is observed already under basal conditions, but becomes even more pronounced on 5% K+ diet.
Conclusion: K+ diet-induced activation of ENaC, but not of ROMK, appears to depend on MR. In KO mice, loss of the MR-dependent activation of ENaC in the CS might be compensated, at least in part, by down-regulation of NCC, which increases Na+ delivery to cells with remaining ENaC activity, and hence improves the electrochemical gradient for K+ secretion.

P85

Dissection of the Aldosterone- and Glucocorticoid-Dependent Pathway Implicated in Sodium Retention in the Rat by Zinc Finger Nucleases (ZFN) and Transcription Activator- Like Effector Nucleases (TALEN)

V. Ponce de Leon, J. Canonica, A.-M. Mérillat, E. Hummler
Lausanne

Purpose: Our research is concentrated on oedema formation and its evolution into nephrotic syndrome, liver cirrhosis and cardiac failure, where adrenal steroid-dependent and -independent mechanisms of sodium retention may be at work. Rats harboring a mutation of the glucocorticoid receptor that inhibits dimerization will bring insights to the role of monomer and dimer forms of the receptor in sodium retention mechanisms.
Methods: The techniques used for the generation of these rats are Zinc Finger Nucleases (ZFNs) and Transcription Activator-Like Effectors (TALENs). Both molecules bind to target DNA and cut it; generating double stranded breaks (DSB). DSBs increase the chances of homologous recombination of up to 1000 fold. A donor plasmid containing the mutated sequence and homology arms allows the insertion of the mutated sequence into the rat genome.
Results: A pair of Zinc Finger Nucleases and five pairs of TALENs were designed. Donor plasmids were constructed either by cloning or synthetization. Vectors expressing the ZFNs or TALENs along with the corresponding donor plasmids were transfected into rat C6 cells. Three TALEN pairs were transfected into C6 rat cells and TALENs showed cutting activity of 20%, which was confirmed by clone screening. First injections of TALEN mRNA into rat oocytes along with the donor plasmid will give F0 pups carrying the mutation. Due to low expression of the ZFN pair, no cutting activity was detected in the rat GR sequence.
Conclusion: TALENs designed and cloned using the REAL method allowed the engineering of TALENs that bind and cut the targeted sequence with up to 20% efficiency as observed by Surveyor assay. ZFNs and TALENs are both powerful and complementary techniques to generate genetically engineered mice and rats for the study of salt homeostasis.

P86

Impact of Uninephrectomy on Body L-Arginine Homeostasis, Enos Function and Blood Pressure Control in Mice

S. M. Pillai, F. Verrey
Zurich

Purpose: The proximal tubule of the kidney is the primary site for L-arginine metabolism. In cases of uninephrectomy (UNX), the remnant kidney increases in size and compensates GFR to some degree. The aim of this study is to test the hypothesis that UNX impacts on the metabolism of L-arginine and its metabolite ADMA and thereby also on endothelial NO production and blood pressure control.
Methods: C57B/6 female and male mice were submitted to left UNX or sham operated. Blood pressure was measured using a tail-cuff system, plasma amino acids and other parameters analyzed and kidney mRNA levels of transporters and enzymes involved in L-arginine metabolism determined.
Results: In the first series of UNX conducted for 3 weeks on female mice, the remnant kidney displayed a weight increase of 30%. A higher blood pressure was observed 7 days after UNX compared to sham operated mice (120 ± 2.14 vs. 112 ± 1.97 mm Hg). In terms of transporter expression, no significant changes of transcript levels were observed whereas that of the metabolizing enzyme arginase II was decreased by 48 % in remnant kidneys. The changes however appeared to be sex dependent, since UNX males displayed a tendency to stronger increase in remnant kidney weight (36%) and no difference in terms of blood pressure and relative mRNA levels. Determinations of amino acid and ADMA levels and of blood pressure by telemetry are currently under way.
Conclusion: Our observations suggest that UNX affects blood pressure and remnant kidney transcript expression in females, whereas these effects are less pronounced in males. It is suggested that this gender difference is related to the more important remnant kidney compensatory growth observed in males.

P87

Minimal Role of Sodium-Calcium Exchanger 1 in Thiazide-Induced Hypocalciuria

W. Li, O. Bonny
Lausanne

Purpose: Thiazide-type diuretics are commonly used in the treatment of calcium-containing kidney stones for their abilities to decrease renal Ca2+ excretion. However, the mechanisms of thiazide-induced hypocalciuria remain debated over whether the enhanced Ca2+ reabsorption occurs in the proximal tubule, the thick ascending limb or the distal nephron. Here, we investigated the role of sodium-calcium exchanger 1 (NCX1), an antiporter for Ca2+ reabsorption in the distal tubule, in thiazide-induced hypocalciuria by using kidney-specific NCX1 knockout mice (NCX1fl/fl, Ksp:Cre+).
Methods: A single dose of 25 mg/kg hydrochlorothiazide (HCTZ) was injected intraperitoneally to NCX1-KO mice and their control littermates. Time-dependent responses to HCTZ were studied on spot urines collected 0, 2, 4, 6 and 12 h after injection.
Results: NCX1-KO and control mice exhibited similar diuretic responses to HCTZ as shown by similar increases in urinary Na+/creatinine ratio and more diluted urines by 2 hours of HCTZ administration compared to vehicle. Concomitantly, Ca2+/creatinine ratio of KO and control mice were respectively reduced to 0.26 ± 0.06 (mean ± S.D., n = 5) and 0.45 ± 0.15 (n = 5) by 4 hours of HCTZ treatment, compared to 0.72 ± 0.25 (n = 6) and 0.57 ± 0.18 (n = 5), respectively, by vehicle treatment. The time-dependent changes in urinary Ca2+/Na+ ratio were identical between KO and control mice after HCTZ treatment, suggesting similar mechanisms of Ca2+excretion.
Conclusion: Thiazide-induced hypocalciuric effect was maintained in the kidney-specific NCX1-KO mice. The data suggest minimal role of NCX1-dependent pathway in thiazide-induced increase in Ca2+reabsorption.

Poster Presentations – Miscellaneous

P88

Decline of Pediatric Urolithiasis in Armenia: 20 Years Observation

A. Sarkissian[1], E. Leumann[2], A. Hesse[3], A. Babloyan[1], N. Arikyants[1]
[1]Yerevan/AM, [2]Zurich, [3]Bonn/DE

Purpose: Urolithiasis is an important health problem in Armenia, also in children. The aim of the study is to determine whether the changes of the social and economic conditions in Armenia – which were catastrophic in the 90-ies and have since much improved – have had an effect on paediatric urolithiasis.
Methods: We studied prospectively all 391 patients aged 1 month to 16 years admitted with urolithiasis from 1992 to 2011 at the Arabkir hospital (national referral center for pediatric nephrology). All calculi were examined by infrared spectroscopy. We compared the data obtained in two decades, 1992–2001 (period I; 254 patients) and 2002–2011 (period II; 137 patients), characterized by major changes in economy and social life in Armenia.
Results: The overall number of children with urolithiasis decreased by 46%. The age distribution has remained similar with mean age 7.8 ± 4.1 years in period I versus 7.04 ± 4.9 in period II. The proportion of male patients has decreased from 69% to 59%. Period II is characterized by a higher proportion of (mostly metabolic) CaOx stones (73% vs. 64% in period I) and by lower proportions of infectious stones (primarily struvite, 11.7% vs. 16%) and of endemic stones (5.8% vs. 7%). Most striking is the strong decrease of the number of endemic primary bladder stones from 20 (8%) to 3 (2%, p <0.001).
Conclusion: In connection with the improved economic situation and better living conditions the incidence of pediatric urolithiasis has nearly halved, mainly as the result of a strong decrease of infectious stones and endemic renal stones and of the near disappearance of primary bladder stones. These changes are presumably the effect of a more balanced nutrition and of better management of pyelonephritis.

P90

Prevalence of Kidney Disease in an Outpatient Diabetic Clinic and Prevalence of Diabetes in an Outpatient Nephrology Clinic in the Canton De Vaud.

J. Parafita, M. Pruijm, D. Kraus, M. Bochud, J. Ruiz, N. Pitteloud, M. Burnier, A. Zanchi
Lausanne

Purpose: The prevalence of diabetic kidney disease (DKD) is rising due to several factors as the expansion of type 2 diabetes and the aging population. We recently identified that the number of diabetic subjects on maintenance hemodialysis almost tripled in only 8 years in the Canton de Vaud reaching 35.6% of patients in 2009. Additional epidemiological data on DKD are lacking in Switzerland.
Methods: We conducted a cross-sectional study on the prevalence of chronic kidney disease (CKD) in the outpatient diabetic clinic and of diabetes in the outpatient nephrology clinic at the University hospital of Lausanne (CHUV).
Results: For the diabetes clinic, data were extracted from the electronic database entries from 2010-2012. The presence of CKD was based on the KDIGO definition. Staging was defined according to the highest value for ACR and lowest value for eGFR (CKD-EPI). Among 670 subjects identified (mean age [y] ± SD: 53.4 ± 14.3,) 49.5%, 13%, 15.1%, 19.1%, 2.7% and 0.6% were respectively in stage 0; 1, 2, 3, 4 and 5. For the outpatient nephrology clinic, electronic entries did not allow to differentiate accurately between diabetic and non diabetic subjects. For this reason, all patient files followed actively at the clinic were manually screened for diagnosis and kidney staging according to last values of eGFR (CKD-EPI). Among 530 subjects identified, 20% were diabetic (mean age: 67 ± 12 y). Among diabetic subjects followed (n = 107), 7%, 12%, 46%, 26% and 9% were in stage 1, 2, 3, 4 and 5.
Conclusion: Prevalence of stage 3-5 kidney disease is high (22.4%) in this university diabetic outpatient clinic. In addition, diabetes is present in 20% of patients followed at the nephrology outpatient clinic. These data underline that a tight collaboration between the diabetic and kidney outpatient clinics is warranted.

P91

Sex Differences in Dietary Patterns Associated With Salt Intake at The Population Level

A. Chappuis[1], N. Glatz[1], P. Suter[2], D. Conen[3], P. Erne[4], I. Binet[5], V. Forni[6], L. Gabutti[6], A. Gallino[7], F. Muggli[8], I. Guessous[9], P. Meier[10], D. Hayoz[11], A. Péchère-Bertschi[9], F. Paccaud[1], M. Burnier[1], M. Bochud[1]
[1]Lausanne, [2]Zurich, [3]Basel, [4]Lucerne, [5]St. Gallen, [6]Locarno, [7]Bellinzona, [8]Vezia, [9]Geneva, [10]Sion, [11]Freiburg

Purpose: Current recommendations to lower population salt intake are not sex-specific, despite known sex differences in dietary patterns and behaviors. We explored the associations of body mass index (BMI), dietary protein and potassium intakes with dietary salt intake specifically in men and women.
Methods: Cross-sectional population-based survey in 1310 people (51% women) aged 15 years and over from three linguistic regions of Switzerland. Twenty-four hour urine collection was used to estimate dietary salt, potassium and protein intakes. We used multiple linear and logistic regressions to take potential confounders and statistical interactions into account.
Results: Men had higher mean [SD] age (49.4 [18.2] vs 47.1 [18.1] years) and BMI (26.1 [4.2] vs 24.4 [4.8] kg/m^2) than women. Men had higher urinary Na, K and urea excretions than women (18 [71] vs 133 [56], 74.5 [25.7] vs 59.0 [21.3] and 434 [143] vs 305 [101] mmoL/24h , respectively). Obesity was positively associated with high dietary salt intake (>10 g/24 h) in men (OR [95%CI] = 4.1 [1.92; 8.76], P <0.001), but less so in women (1.81 [0.75; 4.66], P = 0.18, P interaction<0.05). BMI was associated positively with salt intake in post-, but not in pre-menopausal women. Estimated protein intake was more strongly associated with high salt intake in women than in men (1.84 [1.53; 2.21] vs 1.28 [1.13; 1.46] per 10 g/24h, P int <0.05), whereas the opposite was true for potassium intake.
Conclusion: High protein intake and overweight were associated with high dietary salt intake in Swiss adults, with sex differences. Overall, the results are compatible with a sex-related difference in salt intake, partly related to the role of sex hormones. This might have to be considered when developing a salt reduction strategy at the population level.

P92

Physical Performance and Activity in Hemodialysis Patients Analyzed in a Large Prospective Swiss Dialysis Cohort

R. Winzeler[1], L. Walther[2], F. Barnert[3], M. Vonwiller[1], M. Stücheli-Morssinkhov[4], B. Sam Aka[5], D. Kiss[6], P. Ambühl[1]
[1]Zurich, [2]Baden, [3]Lachen, [4]Schaffhausen, [5]Winterthur, [6]Liestal

Purpose: Impaired physical performance and poor physical activity are common problems among hemodialysis (HD) patients. The aim of the present study was to quantify physical capacity and bodily activity in a Swiss HD population.
Methods: 375 patients were evaluated from the *monitor!* project, a prospective dynamic hemodialysis cohort assessing a wide range of clinical, laboratory and anthropometrical data. Submaximal levels of functional capacity were determined by three-minute walk test (3MWT) and upper body strength (UBS) by a handgrip dynamometer. 24-hour step count and calorie consumption were measured by an armband motion detector (sensewear®, Bodymedia).
Results:

	Age, yr	CCI*	3MWT, m	UBS, kg	Steps/day, n	Calories/day, kcal
Mean±SDV	67.5 ± 14	3.9 ± 2	158 ± 63	23.4 ± 10	3630 ± 3265	2005 ± 488
Median	71.0	3	155	23	3630	1940
25thpercentile	60.0	2.0	111	15.0	962	1693
75thpercentile	78.0	5.0	205	30.2	5370	2261
*) CCI: Charlson comorbidity index						

In the study cohort, walking distance (3MWT), but not handgrip (UBS), was reduced by 40 and 50% in male and female patients, respectively, compared to an age matched non-dialysis population. By multivariate analysis 3MWT and 24-hour step count, but not UBS, were inversely and independently correlated with age and comorbidity, but not with time on dialysis.
Conclusion: Age and comorbidity, but not dialysis vintage, are major determinants of impaired physical performance and activity – as determined by walking distance and step count – in a Swiss HD population. In contrast, upper body strength is comparable to healthy individuals. Implications of these findings on functional dependence, quality of life and survival in HD patients need to be studied.

P93

Experience in Recruiting Participants to the Swiss Kidney Project on Genes in Hypertension

S. Tremblay[1], G. Gok-Sogut[1], D. Siminski[1], M.-O. Levy[2], U. Schupbach[3], S. Estoppey-Younes[1], M. Bochud[1], The Skipogh Study[1]
[1]Lausanne, [2]Geneva, [3]Berne

Purpose: Family-based studies are costly and difficult to conduct, in particular when they are population-based. We here report our experience in recruiting families from the population for a study including renal ultrasound, ambulatory blood pressure monitoring and 24-hour urine collection.
Methods and results: Nuclear families were randomly selected from the general population in Lausanne, Geneva and Berne. In Lausanne and Geneva, around 400 participants from 100 families were recruited in three years. Recruitment is still ongoing in Berne. The first phone call to explain the purpose of the study lasted from 10 to 40 minutes. Important aspects at first contact were to mention the public source of funding, to listen to participants, to be flexible with appointment visits and to give sufficient time for participants to make their decision. Main reasons for refusal were refusal from family members, familial conflicts, chronic diseases, plan to move away, having done health studies before. People were often difficult to reach during working hours and easier to reach after 6 pm. In Lausanne and Berne, the study included a home visit, which was sometimes difficult to organize for non-retired adults, in particular those with small children. Phenotypes most appreciated by participants were renal ultrasound, ECG and bioimpedance. The clinic visit lasted between 2 and 3 hours. The self-reported prevalence of hypertension and diabetes were 26% and 4.1%. The vast majority of participants (90.1%) considered being in good health.
Conclusions: It is feasible, but challenging and time-consuming, to conduct a family- and population-based study within the Swiss context. Contact strategies play a key role in convincing people to participate to a study with extensive phenotyping.

The numbers refer to the pages of this supplement.