PP6.1-1

Effect of thrombin and Factor Xa on cardiomyocytes in a three-dimensional cell culture model

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Background and Objectives: The occurrence of ventricular arrhythmia during acute thrombotic coronary occlusion or reperfusion may be related to direct action of thrombin on cardiomyocytes since concentrations of thrombin are high in thrombus formation. The aim of the present study was to investigate the potential pro-arrhythmic effects of thrombin and factor Xa on cardiomyocytes in a three-dimensional (3D) cell culture model.

Methods: Cardiomyocyte microspheres (spheroids) were prepared from chicken embryo hearts. The contraction frequency of cells was monitored in a three-dimensional (3D) cell culture model. Platelet aggregations (1.25µM ADP: 26.5% vs 23%; 5µM ADP: 53% vs 47%; collagen: 17% vs 11%, p<0.001) were measured. Potentiation by low-dose epinephrine and inhibition by selective alpha-2 receptor blocker atipamezole were determined. To assess the alpha-2 adrenergic activity, potentiation by low-dose epinephrine and inhibition by selective alpha-2 receptor blocker atipamezole were determined.

Results: Thrombin causes a concentration-dependent and time-dependent increase in contraction frequency of cardiomyocyte 3D spheroids (p<0.0001, n=18). High concentrations of thrombin lead to loss of contraction after 1 hour or longer incubation. Similar observations were made after addition of factor Xa (p<0.0001, n=18). Control experiments with flecainide showed a concentration-dependent reduction in contraction frequency (p<0.0001, n=18), figure A and B.

Conclusions: In a 3D-biohybrid system, cardiomyocyte contraction frequency is stimulated both by thrombin and factor Xa, but decreased by flecainide. These data demonstrate a functional link between thrombus-associated molecules and cardiomyocyte contraction, suggesting a potential pro-arrhythmic effect of thrombus formation in the setting of myocardial infarction.
PP6.1-4  
Thrombophilia in young patients with acute myocardial infarction  
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Thrombophilia is a prominent risk factor for venous thromboembolism, its role in arterial events is less defined. We screened for MTHFR C677T, Factor V Leiden, Factor II G20210A, antithrombin, protein C and D deficiencies, lupus anticoagulant, anti-cardiolipin antibodies, hypofibrinogenemia and conventional risk factors (hyperlipidemia, hypertension, smoking, impaired fasting glucose (IFG), diabetes mellitus, and overweight). 137 consecutive patients (99m and 38 F; mean age 44.2 ± 10.5yrs) with a first episode of acute coronary syndrome in young age (<50 years), and 203 age- and sex-matched controls (131m and 72 F; mean age 42.6 ± 6.8yrs). MTHFR homozygosity was found in 22.1% of patients and in 16.4% of controls. Factor V Leiden and Factor II G20210A were found respectively in 7.4% cases vs. 8.5% controls and 8.1% vs. 9.5%; these differences were not significant. There was no difference between patients and controls as to the protein C, S and antithrombin deficiency. We found hypertension in 44.1% of patients and in 19.5% of controls (p<0.0001; OR 3.26; 95% CI 2.00–5.3), cigarette smoking (80.9% vs. 41.0%; p<0.0001; OR 6.0, CI 3.65–10.16) and diabetes (9.7% vs. 0.8%; p<0.002; OR 13.1, CI 7.12–111.0). There was no statistical difference in prevalence of hyperlipidemia (46.4% vs. 40.2%), IFG (12.9% vs. 11.6%), overweight (55.6% vs. 53.1%) and hyperhomocysteinemia (38.8% vs. 39.1%). Cardiovascular risk factors, except for hyperlipidemia, increase the risk of coronary artery disease in our population, whereas prothrombin G20210A mutation, FV Leiden, MTHFR C677T mutation, protein C and S deficiencies and hyperhomocysteinemia did not increase the risk.

PP6.1-5  
A case of venous thrombosis of the popliteal vein through compression by exostosis in a seventeen-year old male patient  
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We report the case of a seventeen-year old male patient who developed first symptoms of exostoses at his right femur and humerus. His mother and brother also presented with exostoses, whereas his father had a history of recurrent venous thromboses and a heterozygous Factor V-Leiden mutation. The patient presented with pain and swelling in his right calf after two 4-hour trainrides and sitting in a lecture with angled legs for several hours. Deep venous thrombosis of the right popliteal vein, the fibular veins and the posterior tibial veins was diagnosed by contrast-mediated venography and colour-coded duplex ultrasound.(Fig 1 and 2) The ultrasound showed compression of the veins by exostoses of the proximal tibia and fibula in sitting position. The exostoses were examined by X-ray. The patient was tested positive for heterozygous Factor V-Leiden mutation. The patient was anticoagulated with body weight-adjusted LMWH (nadroparin) following by phenprocoumon (INR 2–3) for six months. The exostoses of the femur and the tibia were excised one month before ending the anticoagulation. The ultrasonic examination after excision did not show compression of the veins any longer, but advancing recanalization of the posterior tibial veins and the fibular veins, with complete recanalization of the popliteal vein.

PP6.1-6  
Baseline and stress-related levels of coagulation and fibrinolysis factors in patients with anxiety disorders: A case-control study  
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Objectives: The purpose of this study was to investigate the prevalence of different genetic and acquired abnormalities associated with VTE in ArAbs. living in Kuwait. Design and Methods: 640 VTE cases and 222 healthy controls were tested, all of whom were ArAbs living in Kuwait. Coagulometric testing was performed for presence of lupus anticoagulants (LA) and activated protein C resistance (APC-R), and to determine levels of protein C (PC), protein S (PS) and antithrombin (AT). Results and Conclusions: Abnormalities were found in 275 patients (43%) and 31 controls (14%). In details, abnormalities in patients and controls, respectively, were as follows: LA 4.7% & 1.4%; APC 16.1% & 1.8%; PS deficiency 12.8 & 4.5%; PS deficiency 18.4% & 7.7%; AT 5.3% & 1.8%; FVL heterozygous 14.1% & 1.8%; FVL homozgyous 2% & 0%; prothrombin G20210A mutation and HR 2 haplotype. Results: A high proportion of patients and controls showed abnormal levels of procoagulant markers. Conclusion: A high proportion of patients and controls showed abnormal levels of procoagulant markers.

PP6.1-7  
Thrombophilia in ArAbs. in Kuwait  
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Objectives: Venous thromboembolic diseases (VTE) occur due to different genetic and acquired abnormalities in the coagulation factors and their regulatory proteins. The prevalences of such abnormalities were found to be different in different ethnic groups. VTE in Kuwait was reported to be as common as in Western countries. This study report the prevalence of different genetic and acquired abnormalities associated with VTE in ArAbs. living in Kuwait. Design and Methods: 640 VTE cases and 222 healthy controls were tested, all of whom were ArAbs living in Kuwait. Coagulometric testing was performed for presence of lupus anticoagulants (LA) and activated protein C resistance (APC-R), and to determine levels of protein C (PC), protein S (PS) and antithrombin (AT). Results and Conclusions: Abnormalities were found in 275 patients (43%) and 31 controls (14%). In details, abnormalities in patients and controls, respectively, were as follows: LA 4.7% & 1.4%; APC 16.1% & 1.8%; PS deficiency 12.8 & 4.5%; PS deficiency 18.4% & 7.7%; AT 5.3% & 1.8%; FVL heterozygous 14.1% & 1.8%; FVL homozygous 2% & 0%; prothrombin G20210A mutation and HR 2 haplotype.

PP6.1-8  
Evaluation of primary hemostasis in patients undergoing cardiac surgery  
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Objectives: The purpose of this study was to investigate the influence of different cardiac procedures on primary hemostasis. Design and Methods: Prospectively 79 elective patients (age: 71±10 years) without antiplatelet medication or with acetylsalicylic acid monotherapy were enrolled. Three groups were under investigation: coronary artery bypass grafting (CABG; n=29), single valve replacement (SVR; n=25), and double valve replacement or valve repair (VR; n=25). Design and Methods: Prospectively 79 elective patients (age: 71±10 years) without antiplatelet medication or with acetylsalicylic acid monotherapy were enrolled. Three groups were under investigation: coronary artery bypass grafting (CABG; n=29), single valve replacement (SVR; n=25), and double valve replacement or valve repair (VR; n=25). Results: Hemoglobin concentration and platelet count were significantly reduced after surgery. Postoperatively leukocyte counts showed a significant increase with the highest values for SVR and COMPLEX compared to CABG. The values of aggregometry parameters were significantly decreased postoperatively with no significant differences among the groups. PFA-100® CT were preoperatively prolonged for SVR (208±27s) and COMPLEX (186±25s) but not for CABG (96±11s). Postoperatively, a significant reduction of CT values was observed reaching the normal range in all groups. The VWF:Ag and VWF:RCo were higher after surgery, reaching statistically significant nivoue for SVR and COMPLEX. Conclusions: Cardiac procedures led to a decreased platelet aggregation in W and B which can be explained at least in part by the procedure-associated diminished platelet counts. The postoperative normalisation of PFA-100™ CT are most likely due to the increase of VWF:Ag levels as a compensatory mechanism for primary hemostasis.
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Endocrine follow-up of patients experiencing ischemic stroke should be performed 6-12 months after an episode of ischemic stroke. Thus, endocrine evaluation and neu-roendocrine follow-up of patients experiencing ischemic stroke should be performed on a regular basis, in order to monitor pituitary function and, eventually, providing appropriate replacement treatment. Whether this finding can influence the clinical outcome of the ischemic disease remain to be clarified.

Coagulation factor V g allele, HR2 haplotype, 6533T>C and the risk of myocardial infarction

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In a previous study we found associations of the non Leiden g allele (291A>G; 2864A>G; 2868A>G; 2863A>G) coding for amino acids Ser729, Lys830Arg, His837Arg and Lys897Glu with high FV levels and with a protective effect on deep vein thrombosis (DVT). The HR2 haplotype and 6533T>C (Met2120Thr) have been reported to be associated with reduced FV levels. Because high FV levels have been found to be associated with an increased risk of myocardial infarction (M1), we examined how the presence of the g allele, the HR2 haplotype and 6533T>C affected the risk of M1 in men older than 50 years. The above mentioned polymorphisms were examined in 50 men with M1 and in 63 healthy men without M1 and without DVT. A mong the men with M1 30 (53%) were heterozygous and 5 (9.5%) homozygous carriers of the g allele. In the group of the healthy men were 23 (36%) heterozygous and two (3.2%) homozygous carriers of the g allele. 11 (12%) men with M1 and 8 (13%) men of the controls were heterozygous carriers of the HR2 haplotype. Three (4.8%) heterozygous, one (1.6%) homozygous carriers of the 6533T>C polymorphism were found in men with M1 and three (3.3%) heterozygous carriers in controls. The frequencies of the g allele, the HR2 haplotype and 6533T>C did not differ in the two groups.

Conclusion: The risk of myocardial infarction for men older than 50 years is not associated with the g allele, the HR2 haplotype or the polymorphism 6533T>C in the FV gene.

Association between plasma lipoprotein(a) concentration and restenosis after stent implantation in the superficial femoral artery

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Background: Elevated levels of Lp(a) (Lp(a)) are associated with athe-rosclerotic and thrombotic vascular disease. In recent literature plasma Lp(a) is described as an independent predictor of stent restenosis after percutaneous coro-nary intervention. The aim of our study was to evaluate the association between plasma Lp(a) concentration and in-stent restenosis in peripheral artery occlusive disease (PAOD). We decided to include only patients with stenting of the superficial femoral artery because restenosis after stent implantation in this area is a clini-cal problem and the reason therefore unsolved so far.

Methods and Materials: 66 patients (34 male/32 female) with a mean age of 63.7 years for male and 76.4 for female patients with stenting of the superficial femoral artery were included in our study. Plasma Lp(a) concentration was measured 3 months after stent implantation. In-stent restenosis was assessed with duplex scan of the femoral superficial artery 3.6 and 12 months after stent implantation. A stenosis was considered as relevant when stenosis grade was >50%.

Results: 17 of the 66 patients developed restenosis >50% after stent implantation in the superficial femoral artery, 3 of these 17 patients developed a stent-occlusion.

In these patients the plasma concentration of Lp(a) (median 21mg/dl) was not statistically significant higher (p=0.533) compared with the patients who developed no restenosis (median 18mg/dl).

Conclusion: We conclude that patients with higher plasma Lp(a) concentration are not on a higher risk for stent-restenosis in the superficial femoral artery than patients with lower Lp(a) levels.

The association of circulating Factor Seven Activating Protease with clinical outcome in patients with atrial fibrillation

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Objectives: The atrial fibrillation (A F) is accompanied by a hypercoagulable state that may contribute to the development of atrial thromboembolism. FSA P regulates hemostasis and may influence the progression of atherothrombotic cardiovascular disease. It is not known if FSA P is related in any way to the clinical outcome in patients with different forms of arrhythmias. The present study was performed to examine the relation between plasma FSA P and hypercoagulable state in patients with AF.

Design and Methods: 80 patients with AF comprised the study group. FSA P concentrations was assessed in these patients and was compared to control healthy subjects with sinus rhythm (SR).

Results: The median FSA P concentration in control subjects (1053 mPEU/ml, range 855-1195 mPEU/ml) were significantly different from those in patients with AF (1731 mPEU/ml, range 1389-2274 mPEU/ml; p<0.001). The FSA P level was positively correlated with AF duration and the left atrial diameter (P<0.005), but when compare with baseline values, there were not significant changes in the plasma FSA P requiring cardiovascular. The following medications did not influence FSA P concentration and activity: warfarin, β-blockers, or an ACE inhibitor/angio-tension II receptor blocker (P values <0.05). Furthermore, the influence of omega-3 fatty acid treatment on plasma FSA P in vivo was tested, showing that omega-3 fatty acid was without effect.

Conclusions: Enhanced FSA P concentration might be a novel risk factor for stroke mediating hypercoagulable state in patients with AF. Plasma FSA P was an inde-pendent prognostic marker, suggesting its potential role in risk stratification and clinical management of AF.

Clinical outcome of patients with acute pulmonary embolism (PE) after one year

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Introduction: This retrospective study analyzed the clinical outcome of hospitalized patients with acute PE without hemodynamic instability after one year.

Methods: The data are collected retrospectively from medical reports.

Results: 84 patients with PE were analyzed (male 52.6%; female 47.6%) and fol-lowed-up was done after 1 year. In 65% of the medical patients the admitting diag-nosis was VTE. 35% of the medical patients were hospitalized due to different medical disorders. In 69% the PE were diagnosed by ct, in 31% by sintigraphy, in 52% an additional echocardiography was done. A II patients were in a stable cardio-pulmonary situation (Grosser I/II) without hemodynamic instability. All patients were initially treated conservative due to national guidelines with heparins (UFH 38%; LWMH 68%). 53% of the patients got oral anticoagulation for an average time of 9.03 months, 27% were treated with low molecular weight heparins for 11.4 months. 29% of the patients were not treated effectively with an anticoagu-lant therapy within the first year after the acute PE, mainly surgical patients with complications and neurological patients. 16.7% of the patients died in the first year. 3.6% had a relapse. In 19% of the patients an echocardiography was done after 1 year.

Conclusions: Surprisingly 20% of patients were not treated effectively anti-coagulant therapy within the first year after acute PE. 19% of the patients get investigated after one year to determine right heart strain. It has to be shown why so many patients in ambulant section were not treated effectively.

Antithrombotic prophylaxis and therapy. The Hungarian guideline and praxis

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The 4th National Antithrombotic G guideline has been elaborated by experts of the Hungarian Soci-
The purpose of the study was to determine the influence of fibrinogen (F) on the progression of peripheral atherosclerosis in type 2 diabetic pts.

**Design and methods**: 62 pts with type 2 diabetes and diagnosed coronary artery disease were enrolled in a cohort prospective study. We measured in them, at all, progression of peripheral atherosclerosis, defined as change of ankle-brachial index (ABI) after 36 months. Multiple linear regression analysis was built to define continuous variables with predictive value for F and ABI.

**Results**: Study population was on age 60.28 + 27 years (231 men and 109 women) and mean diabetes duration of 8.58 + 6.17 years. Mean plasma fibrinogen level was 4.12 + 0.85 g/L. Multivariate analysis showed F value has been determinate with non-HDL - cholesterol (ð = 0.139, p = 0.027). Linear regression analysis defined F as predictor for minimal value of A1, found at the end of investigation (ð = 0.496, p = 0.007).

**Conclusion**: Our data indicate that plasma determination of fibrinogen have clinical utility in defining the process of progression of peripheral atherosclerosis in type 2 diabetic population.

**PP6.1.15 Risk stratification of recurrent venous thromboembolism**

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**Background**: A better risk stratification for recurrent venous thromboembolism (VTE) in patients with a first episode of idiopathic venous thromboembolism (VTE) is urgently needed.

**Methods**: Retrospective study covering more than 20 years after a first venous thromboembolic event in a group of 1,440 patients with VTE.

**Results**: In the subgroup of patients (n=415) with a first spontaneous VTE, the yearly incidence of a recurrent spontaneous VTE was 8% for the time period 0-2 years and 4-6% in the following 8 years, after a first VTE, triggered by a transient risk factor (oral contraceptives, surgery or immobilization, pregnancy), the yearly incidence of a recurrent spontaneous VTE was 2% (first 2 years) and 1.3-3% (following 8 years). The hazard ratio for recurrent spontaneous VTE in patients with a first spontaneous VTE for specific predictors were as follows: prothrombin mutation heterozygous I2 (95% CI 0.9-1.7), FVL heterozygous I3 (95% CI 0.92-1.8), male sex 1.9 (95% CI 1.4-2.7), D-Dimer 2.3 (0.9-6.4), protein C (<60% activity) 2.6 (95% CI 1.2-5.7), FVL homozygous 3.0 (1.3-6.7), AT (<60% activity) 3.0 (95% CI 1.96-9.6).

In conclusion, in patients with a first spontaneous VTE the yearly recurrence rate of 5% is more than doubled in the presence of relevant thrombophilic risk factors supporting the need of long-term oral anticoagulant therapy after a first idiopathic VTE. In contrast to current ACCP recommendations, thrombophilic risk factors are of clinical relevance.

**PP6.2 Epidemiology and Diagnosis: Venous Thromboembolism**

**PP6.2.1 Validation of a predictive model for identifying an increased risk for thromboembolism in children with acute lymphoblastic leukemia**

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**Introduction**: Venous thromboembolic events (VTE) are a frequent complication in cancer patients. In order to identify reliable markers of risk prediction, we assessed a Blood Count Score (BCS) as predictor for cancer-associated VTE. We investigated the association of haplotypes comprising the fibrinogen alpha (FGA) and FGG genes with cancer-associated VTE in children with acute lymphoblastic leukemia. Our data suggest that the genetic architecture of VT is complex and involves subtle influences through susceptibility and protective haplotypes in FGG with a genetic interaction with the FVL leiden-mutation.

**Methods**: We investigated the association of haplotypes comprising the fibrinogen alpha (FGA) and FGG genes with cancer-associated VTE in children with acute lymphoblastic leukemia. A7 8

**Results**: A association analysis revealed that the FGA-H1 haplotype, and the FGG-H2 and -H3 haplotypes, were significantly associated with VT (FGA-H1, P=0.05; FGG-H2, P=0.032; H3, P=0.0216). In an independent study sample, FGA-H1 (P=0.0085) and FGG-H2 (P=0.05) were significantly associated with TS. When stratifying for FVL leiden-carriership, the association between FGA and FGG and VT was more pronounced in FVL leiden-negative families. Homozygous carriership of the FGG-H2 risk haplotype resulted in the lowest fibrinogen alpha content (P=0.013; ß=7.63±3.05 vs 9.46±3.17, P=2.3±10.5), with increasing concentrations of fibrinogen in heterozygote H2 carriers. Compound heterozygote carriers of one FGG-H2 risk haplotype and one FGG-H3 protective haplotype, showed significant increase in fibrinogen α (P=0.000032), while fibrinogen levels remained unchanged. In contrast, homozygote carriers of the protective FGG-H3 haplotype showed the highest concentration of fibrinogen α content (P=0.013, ß=9.21±3.09, P=0.0031) with decreased total fibrinogen.

**Conclusion**: Our data suggest that the genetic architecture of VT is complex and involves subtle influences through susceptibility and protective haplotypes in FGG with a genetic interaction with the FVL leiden-mutation.

**PP6.2.2 Blood count score enables to predict thromboembolic events in cancer patients - results from the Vienna Cancer and Thrombosis Study (CATS)**

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**Introduction**: Venous thromboembolic events (VTE) are a frequent complication in cancer patients. In order to identify reliable markers of risk prediction, we assessed a Blood Count Score (BCS) as predictor for cancer-associated VTE. We investigated the association of haplotypes comprising the fibrinogen alpha (FGA) and FGG genes with cancer-associated VTE in children with acute lymphoblastic leukemia. Our data suggest that the genetic architecture of VT is complex and involves subtle influences through susceptibility and protective haplotypes in FGG with a genetic interaction with the FVL leiden-mutation.

**Methods**: The Cancer and Thrombosis Study (CATS) is an ongoing prospective observational study in patients with newly diagnosed cancer or disease progression. O facilitation of VTE and information on the patients’ anti-cancer-treatment during
follow-up are recorded. Observation ends with occurrence of VTE, death or after 2 years. At enrolment a blood sample was taken. A VTE risk score was calculated based on the blood cell count. A hemoglobin level below 100 g/L, platelet count above 350 * 10^9/L and leucocyte count above 11 * 10^9/L increased the score by 1, respectively.

**Results:** Data on 635 patients with solid tumours were available. Patients were followed for a median observation time of 366 days, during which 44 objectively confirmed VTE occurred. 477 patients had a score of 0, 120 of 1, 32 of 2 and 6 patients had a score of 3. Comparison of patients with a score of 0 to those with a score of 3 had a hazard ratio of 1.6 [0.8 – 3.4, p=0.19], and those with a score of 2/3 had a hazard ratio of 3.8 [1.6 – 9.2, p=0.003], respectively, adjusted for age and sex.

**Conclusions:** The BCS offers a simple method to determine which cancer patients are at highest risk of suffering a VTE and might be used for risk stratification in future interventional trials.

**PP6.2-4**

**Inferior vena cava thrombosis and its relationship to chronic myeloproliferative disorders, the JAK2V617F mutation and prv-1 mRNA expression**

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**Objectives:** Splanchnic vein thrombosis (SVT) is a typical manifestation of poly- cythaemia vera (PV) or essential thrombocythosis (ET). The recently discovered JAK2V617F somatic mutation and an increased expression of granulocyte PRV-1 mRNA are closely related to chronic myeloproliferative disorders (CMD). We investigated if thrombosis involving the inferior vena cava (IVC) is related to the JAK2V617F mutation, an increased expression of PRV-1 mRNA or the presence of cmd.

**Design and Methods:** Blood samples were obtained from 40 IVC thrombosis patients seen in our University Hospital’s outpatient department. Fifty-three patients with isolated lower extremity DVT (LE-DVT) and 15 SVT patients served as controls. The presence of a JAK2V617F or PRV-1 mRNA was assessed by real-time polymerase chain reaction (RT-PCR).

**Results:** The JAK2V617F allele was not detected in any of the IVC thrombosis patients, and no patient presented with an increased prv-1 mRNA expression. However, four patients (10%) had borderline levels of prv-1 mRNA (i.e., 0.7–1.5%). These patients did not suffer from known CMD. Even after a median observation period of 25 months (range 20–27) we did not observe any increase of white or red blood cell or platelet count in these four patients. In contrast, the JAK2V617F allele was detected in 3 patients with SVT and known CMD (PV n=2, ET n=1) as well as in one LE-DVT patient.

**Conclusion:** A according to our data, there is no evidence that thrombosis involving the inferior vena cava is related to the JAK2V617F mutation, an elevated prv-1 mRNA expression or the presence of myeloproliferative disorders.

**PP6.2-5**

**Cytomegalovirus infection is associated with venous thromboembolism of immunocompetent adults – a case-control study**

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**Objectives:** Cytomegalovirus (CMV) is known to contribute to the development of venous thromboembolism (VTE) in immunocompromised patients while literature data on the role in immunocompetent individuals is limited to case descriptions. We initiated this study to investigate the role of CMV infection regarding the occurrence of VTE in a large cohort of immunocompetent patients.

**Design and Methods:** In a case-control study cmD and cmV antibody titres were determined in blood samples from 187 VTE patients and 187 age- and sex-matched blood donors without a history of VTE. cmV- IgM avidity was measured in blood samples from 187 VTE patients and 187 age- and sex-matched blood donors without a history of VTE.

**Results:** cmV- IgG antibodies were found more frequently in VTE patients compared to controls (59.9% vs. 43.3%; OR 1.95 [95%-CI 1.30–2.95]; p=0.002). cmV-IgM antibodies could be detected more often in patients with spontaneous VTE compared to controls (7.4% vs. 11.9%; OR 0.79 [95%-CI 0.51-1.23]; p=0.024). Mean CMV antibody titres were significantly higher in the case group (393 vs. 1.3 AU/ml for IgG [p=0.001]; 0.22 vs. 0.17 index for IgM [p=0.008]). cmV-IgG avidity was high in 89% cmV- IgM positive samples from the patient group (one sample was determined as ‘greyzone’).

**Conclusions:** Our data indicate that cmV might be a relevant risk factor for the development of VTE in immunocompetent individuals. The overall low rate of IgM antibodies in the patient group and the results of avidity testing favour recurrent and not primary cmV infection to be relevant for the development of venous thrombosis in this cohort.

**PP6.2-6**

**The role of plasminogen activator inhibitor-1 Activity and factor XII activity in venous thromboembolism**

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**Objectives:** Venous thromboembolism (VTE), including deep vein thrombosis (DVT) and pulmonary embolism (PE), occurs secondary to a number of heredi- tary and acquired disorders of hemostasis. Widespread screening of patients with venous thromboembolism (VTE) for thrombophilic risk factors has become common clinical practice. Because of the increasing number of risk factors, assessing the risk of recurrence in an individual patient is intricate; therefore, a laboratory method that measures multifactorial thrombophilia is required.

**Design and Methods:** In 127 patients with idiopathic, recurrent deep venous thrombosis (>2 incidents) the activity of antithrombin, protein C, S, AT, PT, D-Dimer, pro- thrombin time and INR were investigated. Fibriogen, factor VIII, factor XI, factor XII, PA-1 activity were determined. Patients with idiopathic DVT, after elimina- tion of most important thromboembolism risk factors, were qualified for the study. Results were compared with a group of not recurrent DVT (485 patients).

**Results:** All analytes of fibrinolysis system demonstrated significantly higher PA-1 activity (>0.021) than patients with not recurrent DVT. The patients demonstrated factor XII activity decrease (>0.003) than patients with not recurrent DVT.

**Conclusions:** Reduced factor XII activity and higher PA-1 activity were found in patients with recurrent DVT. These factors may be associated with the risk of recur- rent venous thromboembolism (VTE).

**PP6.2-7**

**Homocysteine remains a risk factor in patients with peripheral arterial disease**

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**Objectives:** Homocysteine (HC) >12µmol/L is a cardiovascular risk factor (R.F.). Epidemiology described an odds ratio (OR) of 7 for hyperhomocysteinemia (HHC) as RF for peripheral arterial disease (PAD). Since recent studies, which lowered HHC with a B-vitamin-combination, found an aug- mentation of coronary events, it is questionable whether in the era of new thera- peutic strategies (A C E-I A R B statins) HHC in PAD remains a RF deserved to be.

**Design and Methods:** 2002–2003 521 consecutive patients of Angiology were screened for HHC. A after 5½ years of observation death, cardiovascular death and events were noted.

**Results:** 252 of 521 patients had elevated HCC (women: 15.3±6.4 µmol/L; men: 15.1±5.6 µmol/L); 39% of men but 63% of women had HHC. During the observa- tion period 107 patients (20.5%) died; 26.3% of men and 20.9% of women. In women, mortality was elevated by HHC from 18.2% to 22.5% (p=0.03), in men from 12.0% to 33.3% (p<0.001). Per elevation of HCC by 5µmol/L, the risk for death in women was augmented by 12%, in contrast in men by 44%. Under modern vascular-protective therapies in PAD patients, HHC remains a substantial RF in men, whereas HHC is more frequent in women due to lower cut-offs. It has to be questioned whether the lower cut-offs in women are derived from the research of recurrent thrombosis are justified in the arterial setting and how one can treat HHC in men (which have an OR of 2.8 for death) without doing harm.

**PP6.2-8**

**Right-to-left shunt, atrial septal aneurysm and thrombophilia in patients with cryptogenic stroke or TIA versus those with venous thromboembolism**

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**Objectives:** Whether intracardiac right-to-left shunt (RLS) is an independent risk factor for cerebrovascular accidents is dispute. In patients with RLS, venous thrombo- embolism (VTE) may represent a predisposition to stroke/transient ischemic attack (TIA). Whether thrombophilia is associated with RLS is unclear. We compared prevalence of intra- and extracardiac RLS and of atrial septal aneurysm (ASA) between 29 selected nondiabetic patients with cryptogenic stroke (n=17)
Posters

related the complication with central and long-lasting peripheral venous catheter, ischemic thrombosis was recorded in only one patient. In 75% of cases visceral (mesenteric, renal and duodenal) thrombosis was identified; arterial deep venous thrombosis (40%)–upper (27.5%) and lower (12.5%) limb; in 10% of treatment.

increasing. It urges us to improve our management in diagnosis, prophylaxis and treatment.

Results: Based on clinical and exploratory data (color Doppler ultrasound, conventional and MRI angiography), thrombosis was identified in 40 cases (6.63% of patients), with a significantly lower proportion in patients ≥18 years (4.53% vs 16.66%); most prone to develop thrombotic events have been those with leukemia (19.75%) compared to those with solid tumors (5.4%) and transplanted patients (3.07%). Clinical presentation was dominated by superficial (47.5%), followed by deep venous thrombosis (40%)–upper (27.5%) and lower (12.5%) limb; in 10% of cases, visceral (mesenterial, renal and duodenal) thrombosis was identified; arterial ischemic thrombosis was recorded in only one patient. In 75% of cases we correlated the complication with central and long-lasting peripheral venous catheter, or medication (A sparaginase, Corticotherapy, etc.). Thrombotic events were cause of death in 3 cases.

Conclusions: The identification of thrombosis in paediatric malignancy is steadily increasing. It urges us to improve our management in diagnosis, prophylaxis and treatment.

PP6.2-9

Antithrombin Cambridge II (A384S): Frequency in Austrian patients

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Well over 120 mutations causing antithrombin deficiency have been identified. One mutation - A antithrombin Cambridge II (A384S)- is associated with a reduced heparin cofactor activity, and the variant is the stringent risk factor for thrombotic disease. The mutation was found with a relatively high frequency in the British population (1.45 %) and in Spanish patients (1.7 %) with deep venous thrombosis (DVT) or pulmonary embolism (PE). In French patients, a lower frequency was found (0.4 %). In our patients, thrombophilia was not associated with intracardiac RLS, but tended to be associated with a ASA (83 % vs. 40 %, p=0.008). Intracardiac RLS may have a role in the multi-factorial pathogenesis of stroke/TIA of embolic origin, A SA seems an independent risk factor for stroke/TIA with possible interaction with thrombophilia.

PP6.2-10

Thrombotic events in children with malignancy

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Introduction: Haeostatic disturbances and cancer are intimately interconnected in a bidirectional manner. D espite of protective antithrombotic factors and mechanisms, thrombotic events are steadily increasing in paediatric cancer patients. O bjective of the study was to evaluate frequency, type and site of thrombosis in cancer patients ≥18 years vs. >18 years underhing disease and its impact on evolution.

Patients and methods: I t is a descriptive and analytic retrospective study, conducted on a group of 603 consecutive patients, 507 ≥18 years and 96 >18 years; 205 had leukemia, 333 solid tumors and 65 hematopoietic stem cell transplantation.

Results: Based on clinical and exploratory data (color Doppler ultrasound, conventional and M R I angiography), thrombosis was identified in 40 cases (6.63% of patients), with a significantly lower proportion in patients ≥18 years (4.53% vs 16.66%); most prone to develop thrombotic events have been those with leukemia (19.75%) compared to those with solid tumors (5.4%) and transplanted patients (3.07%). Clinical presentation was dominated by superficial (47.5%), followed by deep venous thrombosis (40%)–upper (27.5%) and lower (12.5%) limb; in 10% of cases, visceral (mesenterial, renal and duodenal) thrombosis was identified; arterial ischemic thrombosis was recorded in only one patient. In 75% of cases we correlated the complication with central and long-lasting peripheral venous catheter, or medication (A sparaginase, Corticotherapy, etc.). Thrombotic events were cause of death in 3 cases.

Conclusions: The identification of thrombosis in paediatric malignancy is steadily increasing. It urges us to improve our management in diagnosis, prophylaxis and treatment.

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PP6.2-11

Frequency of established risk factors for deep-venous-thrombosis in a collective of Austrian patients

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A im of our study was to determine to what extent established risk factors for deep vein thrombosis were associated with the occurrence of venous thrombotic disease. 209 consecutive hospitalized patients with spontaneous deep-venous-thrombosis (proved by ultrasound or phlebographically) were included in the study.

Results: Risk-factor / No. of patients / %

M utation in Prothrombiningene(het/hom)20.210G → A (9 / 9,1%)

Factor-V-Leiden-mutation (het/hom) 1.691G → A (52 / 24,9%)

A ntithrombin-deficiency (activity) (70% ) / 2 / 1,0%

Protein C-deficiency (activity) (45%) / 6 / 2,9%

Protein S-deficiency (free antigens) (60%) / 2 / 1,0%

L upus-anticoagulans / 13 / 6,2%

Increased homocysteine (>20μmol/l) / 15 / 7,2%

Increased F VIII-activity (>200%) / 12 / 5,7%

Increased F XI-activity (>170%) / 4 / 1,9%

Patients with identified risk factor / 125 / 59,8%

Patients with no identified risk factor / 84 / 40,2%

*19 of the patients were positive for 2 or more risk factors.

Conclusion: I n accordance with published papers, our results show that spontaneous deep vein thrombosis is associated with at least one established risk factor in nearly 60% of cases. However, in about 40% of patients the occurrence of disease cannot be explained by our screening panel.

PP6.2-12

FXII T46T genotype is not associated with a higher risk of cerebral venous thrombosis

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Objectives: The importance of the Factor XII C46T gene polymorphism as a risk factor for venous thromboembolism is still under discussion. Recently an association between the TT genotype of FXII C46T gene polymorphism and the risk of cerebral venous thrombosis CVT was published by R enner et al. (Neurology, 2008).

Therefore we were interested in the prevalence of the TT genotype in our patients with CVT.

Design and Methods: 84 age and sex matched blood donors (71 females, 13 males) serving as healthy controls and 71 patients (60 females, 11 males) with CVT were included in our study. We focused on the FXII T46T, the FVL and Prothrombin 20210 genotypes, lupus anticoagulants and coagulation inhibitor deficiencies. Furthermore we documented the intake of oral contraceptives OCs, pregnancy and puerperium during manifestation of CVT.

Results: 371 patients revealed FXII T46T genotype (1 with heterozygous FVL) and 204 controls showed FVL and Prothrombin 20210 mutation, 371 lupus anticoagulants and 371 proteins deficiency. The well known risk factors intake of OCs (32/60) and puerperium (66/60) were often found in our 60 females with CVT. In healthy controls the common prevalence of FVL and prothrombin 20210 mutation was detected. A bout 30% of the female blood donors used OCs.

Conclusion: A ccording to our data the prevalence of the FXII T46T genotype among patients with CVT is not increased while established thromboembolic risk factors were more often detected compared to healthy controls.

PP6.2-13

Thrombin generation in the course of bone marrow transplantation

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Transplantation of haematopoietic stem cells (H SCT) is often complicated by haemostatic complications. Patients are prone to severe bleeding during thrombocytopoenia and alteration of the mucous membranes for radiation, cytostatic therapy and infections. In contrast patients may exhibit thrombotic complications like veno occlusive disease (VOD) or transplantation associated thrombotic microangiopathy. We were interested in whether the thrombin generation assay may serve as diagnostic tool to predict thrombolytic or thrombotic complications in the setting of HCT. 14 consecutive pediatric patients to receive an allogeneic H SCT were included in the study.

Results: Both patients with H SCT died. One patient with an allo geneic H SCT developed thrombotic complications but not VOD. The other patient with aplastic anaemia showed mild thrombotic complications and severe VOD. In both patients VOD was diagnosed in two patients, one developed haemorrhagic cystitis

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Results: Thrombin generation was measured using a commercial assay (Technoclone TGA) expressing the results in peak - thrombin (PT) and area under the curve (AUC). The assay comprises two start reagents (RCL - RCH). Before BT PT was within the normal range (PT-RCL: 157 ±m - normal 125 - 225, PT-RCH: 215 ±nm - normal 175 - 450), whereas AUC was elevated (AUC-RCL: 3863 ±m - normal: 2200 - 3100, AUC-RCH: 4194.4 ±m - normal: 2200 - 3600). All parameters decrease until week 3 after HSC to increase then further on above the pre-HSC values. VOD patients had lower PT and AUC values. 5 patients received defibril-}

lide-treatment. A though not significant PT and AUC were lower in these patients. Conclusion: TGA may be helpful to assess the risk of coagulation abnormalities in the course of HSC.

PP6.2-14

D-dimer in the diagnosis of proximal and distal venous thrombosis of the lower limbs

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Introduction: The diagnostic value of D-dimer (DD) in the exclusion of deep venous thrombosis (DVT) is well established, but is less well known in distal (infrafemoral) and superficial venous thrombosis (VT). We evaluated the performance of different DD assays in the diagnosis of symptomatic proximal and distal VT of the legs.

Methods: 243 outpatients with symptoms suspicious of VT were enrolled in the study. All patients received compression ultrasonography (CUS) of the whole symptomatic leg(s). Five different DD assays were performed: Vidas-DD (bioMerieux, n=242), Liaest-DD (Stago, n=243), HemosIL-DD (IL, n=215), HemosIL-DDHS (IL, n=212), Innovance DD (Siemens, n=206). All patients with a negative CUS were followed up for 3 months for thromboembolic complications.

Results: 38 proximal DVT, 17 distal DVT, 14 muscle vein thromboses (MVT), and 20 superficial thrombophlebitides (ST) were diagnosed by CUS. None of the 147 patients with normal CUS developed thromboembolic complications in the following 3 months. Six patients who were lost from follow-up and one patient who developed VT of the contralateral, former asymptomatic leg, were excluded for this analysis. The sensitivity for a proximal DVT, distal DVT, MVT and ST was: Vidas-DD 100%, 88%, 79%, 75%, Liaest-DD 97%, 88%, 79%, 70%, HemosIL-DD 97%, 82%, 70%, 72%, HemosIL-DDHS 97%, 88%, 70%, 72%, Innovance-DD 97%, 88% (MVT and ST not analysed). The overall specificity for VT was 49% (Vidas-DD), 55% (Liaest-DD), 52% (HemosIL-DD), 54% (HemosIL-DDHS), 53% (Innovance-DD).

Conclusion: The sensitivity of different DD assays varies between 97-100% for proximal DVT, 82-88% for distal DVT, 70-100% for muscle VT, and 70-75% for superficial thrombophlebitis.

PP6.2-15

Patients with venous thrombosis and pulmonary embolism have different risk factors

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Objectives: Venous thromboembolism (VTE) including venous thrombosis and pulmonary embolism is a common disease with potentially severe complications. Former studies have shown that there might be a difference in the prevalence of hereditary risk factors between patients with prevalent venous thrombosis and predominant pulmonary embolism. Our investigation aimed to identify differences between these two groups regarding acquired and hereditary risk factors for VTE.

Design and Methods: We analyzed retrospectively 543 consecutive patients (women: n = 336 / age 38.9 ± 14.8; men: n = 207 / age 48.6 ± 15.1) of our outpatient clinic having been at least one VTE. A queried risk factors and highly prevalent hereditary risk factors (factor-V-Leiden and prothrombin G20201A mutation) of thrombophilia were analyzed statistically.

Results: The prevalence of acquired risk factors were not different between patients with venous thrombosis (with or without additional pulmonary embolism, group one, n = 68) and patients with isolated pulmonary embolism (group two, n = 68). However the groups differed significantly (p = 0.036, Fisher's exact test) regarding the prevalence of factor-V-Leiden and prothrombin mutation (group one 33.7%, group two 20.6%).

Conclusions: We could harden hints that hereditary risk factors for VTE are found less often in patients with predominant pulmonary embolism. Therefore venous thrombosis and pulmonary embolism might be two different entities though sharing several hereditary and acquired risk factors.

PP6.3

Thrombophilia and Familial Thrombosis

PP6.3-1

Usefulness of Innovin dilute prothrombin time for the detection of lupus anticoagulant

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Objectives: Lupus anticoagulants (L.A.) are antibodies which inhibit in vitro phospholipid-dependent tests of coagulation. No single screening test can detect all L.A. positive patients, so the SCC Subcommittee for the Standardization of L.A. recommends at least two independent tests for L.A.-screening. Commonly used screening tests are based on the Kaolin Clotting Time (KCT), a L.A.-sensitive aPTT or the dilute Russell's Viper Venom Time (dRVVT). Dilute prothrombin time (dPT) has also reported as a sensitive test for L.A.-screening. Therefore we evaluated the usefulness of a homemade dPT in comparison to different commercial available tests.

Methods: All tests were performed on the BCS analyzer (Siemens Healthcare Diagnostics, Germany) in a 1/200 dilution was evaluated. Following we estimated in 22 patients, previously tested positive for L.A., dPT, KCT (Kaoclot, Life Diagnostics, USA), dRVVT (L A I L A 2, Siemens Healthcare Diagnostics, Germany) and MxCoLa (Instrumentation Laboratories, Germany).

Results: In a post-assay coefficient of variation (CV) for dPT was 1% and inter-assay CV 5.6%. Normal values assigned in 50 healthy individuals ranges from 35 to 51 sec, 51 sec was chosen as the cut-off value. We obtained negative results in 2 patients with dPT in 1 patient with MxCoLa, in 8 patients with KCT and in 3 patients with dRVVT.

Conclusion: Innovin dPT a low-cost but high sensitive screening test for L.A. and improves the sensitivity of LA screening in combination with other commercial assays such as L.A.-sensitive aPTT.

PP6.3-2

Pulmonary embolism in a patient with severe Factor V deficiency

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Objectives: Severe factor V (FV) deficiency is known to be associated with a clinical relevant bleeding tendency. This case report describes a patient with FV deficiency suffering from pulmonary embolism and showing so far unknown mutations of the FV gene.

Methods: The clinical course was analyzed retrospectively.

Results: Case report: The 40 year old male patient was historically tested with a significant prolongation of both prothrombin time and partial thromboplastin time around 20 years ago; interestingly, he did not report any bleeding tendency. Further investigation revealed FV activity of 5%, while an inhibitor could be excluded; FVIII activity was measured within the normal range. In his 31st year of life ulcerative colitis was diagnosed. The patient now developed a thromboembolic event of the pelvis and leg, and pulmonary embolism could be confirmed by computed tomography. Some days before, steroid therapy of the colitis was performed. Further investigation of the coagulation system revealed normal values for protein C and antithrombin. Neither antiphospholipid antibodies nor polymorphisms of FV (G1691A) and FII (G20210A) could be detected. Molecular genetic investigation revealed a first described deletion (Tyr1554_Tyr1555 del/InsTyr) and missense mutation (Met1339Lys) within the FV gene. A micoagulation with phenprocoumon for at least 12 months was initiated in this patient.

Conclusions: Patients with FV deficiency cannot only present with a bleeding tendency, but might also suffer from serious thromboembolic events which possibly are favoured by the underlying disease and concomitant therapy.

PP6.3-3

A novel mutation causative for type I antithrombin-deficiency combined with other thrombophilic defects in a family with severe thrombocytopenia disease

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Objectives: Antithrombin deficiency (ATD) are rare. Little is known about clinical relevance of ATD combined with other thrombophilic defects. Since 1996 we are observing the occurrence of venous thromboembolism (VTE) in four generations of a family in which ATD segregates with factor V Leiden-mutation (FVL) or protein C-deficiency. 12 family underwent laboratory investigation revealed FV activity of 5%, while an inhibitor could be excluded; FVIII activity was measured within the normal range. In his 31st year of life ulcerative colitis was diagnosed. The patient now developed a thromboembolic event of the pelvis and leg, and pulmonary embolism could be confirmed by computed tomography. Some days before, steroid therapy of the colitis was performed. Further investigation of the coagulation system revealed normal values for protein C and antithrombin. Neither antiphospholipid antibodies nor polymorphisms of FV (G1691A) and FII (G20210A) could be detected. Molecular genetic investigation revealed a first described deletion (Tyr1554_Tyr1555 del/InsTyr) and missense mutation (Met1339Lys) within the FV gene. A micoagulation with phenprocoumon for at least 12 months was initiated in this patient.

Conclusions: Patients with FV deficiency cannot only present with a bleeding tendency, but might also suffer from serious thromboembolic events which possibly are favoured by the underlying disease and concomitant therapy.
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associated with type I antithrombin deficiency in a family with VTE. The high pen-

thrombotic tendency or pregnancy loss in antiphospholipid syndrome, but it is not

assay in individuals with antiphospholipid antibodies

Markers of thrombin generation and parameters of overall haemostatic potential

for APL in all young patients with stroke.

young patients with both: the presence of APL risk factors. We advocate screening

fourth of the patients were smokers and one-sixth had a family history of thrombo-

of the samples and in 4% of controls. The 'p' value for both ACL and LA was 0.03

(8%), and diabetes mellitus (3.6%). APL (LA and ACL) were present in 29.4%

factors. In this study, the prevalence of two clinically significant APL - anticardio-

stein C, S A thrombin and Plasminogen, lupus anticoagulants and anti-cardiolipin

antibodies. Reference was 135 women of childbearing age (group 2).

Results: Group 1: The most common histories were thromboembolic events, recur-

rent fetal losses followed by combinations of thromboembolism and abortions. The

type of VTE in this kindred demonstrates the high risk of type I antithrombin

deficiency combined with other coagulation defects.

PP6.3-4

Antiphospholipid antibodies in young Indian patients with stroke

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Stroke is defined as the sudden occlusion or rupture cerebral arteries or veins

resulting in focal cerebral damage and neurological deficits. It may be caused by

antiphospholipid antibodies (APL) especially in young persons without other risk

factors. In this study, the prevalence of two clinically significant APL - anticardio-
lipin antibody (ACLA) and lupus anticoagulants (L A) in young patients present-

ing with sudden neurological deficits was studied and compared with age- and

sex-matched controls. Fifty healthy volunteers and 51 young patients (less than 45 yrs)
diagnosed as ischemic stroke were recruited for the study. Overall, the risk fac-
tor profile was: smoking (43%), positive family history (28.5%), hyperlipidemia
(8%), and diabetes mellitus (3.6%). APL (L A) was present in 29.4% of the samples and in 4%
controls. The 'p' value for both all APL and LA was 0.03 and 0.02 respectively, but the maximum level of LA was 25 GPL units only. One-
fourth of the patients were smokers and one-sixth had a family history of thrombo-
sis (p<0.05 for both). Our study showed an association between ischemic stroke in
young patients with both: the presence of APL risk factors. We advocate screening
for APL in all young patients with stroke.

PP6.3-5

Markers of thrombin generation and parameters of overall haemostatic potential

assay in individuals with antiphospholipid antibodies

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O bjectives: Blood hypercoagulability may be one of the underlying mechanisms of
therapeutic tendency or pregnancy loss in antiphospholipid syndrome, but it is not
clear whether assays which reflect thrombin or fibrin generation may distinguish
individuals who are prone to develop these complications

D esign and Methods: In this retrospective study we measured concentration of
prothrombin fragment 1+2 (F1+2), thrombin-antithrombin complex (TAT), and
D-dimer in 33 individuals with primary occurrence of antiphospholipid antibodies.
In the same patients we also determined parameters of overall haemostatic poten-
tial (OHAP) assay (first described by Blombäck et al, 2001, which reflect the balance
between fibrin generation and lysis in microtiter plate wells. Thirty-six patients were
symptomatic (pregnancy loss in 12, and thrombosis in 24), while 17 individuals were
asymptomatic.

R esults: M ean concentrations of F1+2, TAT and D-dimer were higher in group of
symptomatic individuals than in asymptomatic group, but the differences were not
significant, (p>0.05, for all three markers). Results of overall coagulation potential,
overall haemostatic potential and overall fibrinolytic potential were also similar
in both groups, (p>0.05, for all three parameters). Sensitivity and specificity of all
investigated parameters for occurrence of thrombosis or pregnancy loss in indi-
viduals with antiphospholipid antibodies were quite low.

C onclusions: A though our study is limited by size and retrospective nature, it could
be concluded that neither markers of thrombin generation nor parameters of OHAP
assay are reliable tools in recognition of individuals with antiphospholipid antibodies
who are at increased risk for development of thrombotic complications or sponta-
eneous pregnancy loss.

PP6.3-6

Incidence of thrombophilic disorders in women of childbearing age and a history
of thromboembolism or recurrent pregnant loss vs. a healthy control group

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Background: Thromboembolism (VTE) is the major reason for motherly morbidity and
mortality during gravidity and puerperium. The risk of VTE's during pregnancy is five to six times higher than for non-pregnant women. Inherited and acquired
thrombophilic disorders further raise the risk. Some of these disorders are addi-
tionally responsible for recurrent abortions. Is screening these women for throm-
boembolism useful?

Patients and methods: 133 pregnant patients with history of thromboembolism and/or
recurrent fetal losses were included in this study (group 1). They were screened
for factor V Leiden mutation, Prothrombin mutation (G20210A), deficiency of Pro-
tein C, S A thrombin and Plasminogen, lupus anticoagulants and anti-cardiolipin
antibodies. Reference was 135 women of childbearing age (group 2).

Results: Group 1: The most common histories were thromboembolic events, recur-
rent fetal losses followed by combinations of thromboembolism and abortions. The
control group was composed of women between 18 and 48 years, without a history
of events. 47% of all pregnant patients had one thrombophilic disorder, 7% two or more. The reference cohort had only 6% single thrombophilic defects, in 1% two
combined disorders were found. The most frequent disorder was the Faktor V
Leiden mutation (63% in group 1 vs. 46% in the control group) followed by pro-
thrombin mutation 21% vs. 21%.

Conclusions: In more than 50% of women with a history of thromboembolism or
recurrent abortions an underlying thrombophilic disorder was detected vs. 7% in
the control group. Therefore screening for thrombophilia is recommended in these
patients.

PP6.3-7

Antibodies to lepirudin in subcutaneous long-term treatment of a patient with
recurrent venous thromboembolism due to Behcet's disease - a case report

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The direct thrombin inhibitor lepirudin is mainly applied in heparin-induced
thrombocytopenia. We report here the case of a 37-year-old Kurdish woman in
whom Behcet's disease was diagnosed in 1998 due to Budd Chiari syndrome (BCS)
complicated by pulmonary embolism. Recurrent VTE occurred despite antico-
agulant therapy with phenprocoumon and various immunosuppressive therapy
regimens. In 2001, when BCS recurred ultimately i.v. lepirudin was administered.
When the patient improved and remained clinically stable lepirudin was applied
subcutaneously. During long-term treatment with twice-daily 50mg no further
VTE was observed over the following years. In May 2005 anticoagulant therapy
was switched to phenprocoumon. BCS5 was switched to phenprocoumon. BCS5 was reduced when INR values were subopti-
mal in February 2007, and lepirudin treatment was immediately restarted. A fer
admission the patient received 50mg b.i.d. iepirudin s.c. with plasma levels in the
therapeutic range. O ver the following months, lepirudin levels repeatedly exceeded
the upper limit of this range and the dosage was stepwise reduced. Finally, 20mg
b.i.d. were sufficient to obtain therapeutic levels. Renal function was normal, but
lepirudin antibodies were present in high titer, as assessed by ELISA. We suppose
that these antibodies reduce renal filtration of lepirudin thus leading to increased
plasma levels.

Conclusion: This case is an example for the efficacy of long-term therapeutic-
dose anticoagulation with sc. lepirudin in patients with recurrent venous thromb-
boembolism despite therapeutic-dose anticoagulation with LMWH or vitamin K
antagonists. However, regular measurement of lepirudin plasma levels is needed.
If stepwise dose lowering is required over time, the presence of lepirudin antibodies
should be considered.
Design and Methods: We investigated the significance of elevated soluble P-selectin in menstruation/MI in Hungarian women. Interaction between HCY and Lp(a) levels were significantly elevated and B12 vitamin intake was decreased only in the CS+M+I group. Folic acid concentration did not differ significantly. There was no association between MTHFR genotype and CS or MI. Elevated HCY (>12.5 micromol/L) increased the risk of CS (OR: 2.04 (1.27–3.28)) and MI (OR: 2.60 (1.43–4.71)) only in women. Similar results were obtained with elevated Lp(a) (>300 mg/L). In this case, ORs were 1.64 (1.03–2.61) for CS and 1.89 (1.06–3.38) for MI. Parallel elevation of HCY and Lp(a) increased the OR to 3.75 (1.80–7.83) and 5.05 (2.08–12.31), respectively. In women under the age of 55, ORs were 6.43 (1.67–24.73) and 16.40 (4.26–76.71) were obtained.

Conclusions: The elevation of HCY or Lp(a) level confers a moderate risk of CS and MI. In women, the elevation of both parameters additively increases the risks, and in women below 55 the risk becomes especially high.

PP6.4-4 Fondaparinux during pregnancy
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Objectives: In the literature only few reports exist about fondaparinux in pregnancy and there are no controlled clinical trials. On the other side we know from in vitro studies that there occurs a minimal transplacental passage of fondaparinux. In our center five pregnant women with thrombophilia, allergic reactions against LMWH, or history of heparin-induced thrombocytopenia, respectively, received fondaparinux over several weeks in pregnancy and in the postpartal period.

Subjects and Methods: Five women with pregnancy and thrombophilia were compared with five pregnant women with nearly the same history of thrombophilia and the same time schedule for taking plasma samples. The dosage was 2.5 and 5 mg/day. Markers of activation (D dimer, F1+2) and thrombin generation (TG) were determined in both groups. TG was measured at the BCS (ETP, Siemens Healthcare) and Calibrated Automated thrombogram (CAT, ThromboScope).

Results: Fondaparinux was well tolerated in all women. Coagulation activation was nearly identical in both groups. There were also no significant differences between TG. Both methods demonstrate a moderate coagulation during pregnancy and a significant decrease in the postpartal period.

Conclusions: The data demonstrate that the selective Xa inhibitor fondaparinux can be used in patients with allergic and immunologic reactions against LMWH with the same effectiveness and safety like the established therapy with LMWH. There are no significant differences regarding thrombin generation in both groups. All pregnancies were successful without any complications for mother and child and fondaparinux is an alternative therapy in this group of patients.

PP6.4-5 Global coagulation markers in women with and without anticoagulation during pregnancy
Henninger A1, Phaup O1, Kaider A2, Kyrie P1, Eichinger S1

Background: Pregnancy is associated with a hypercoagulable state. We investigated if the hemostatic system alterations can be monitored by global coagulation markers.

Methods: In a prospective case-control study, we followed 61 women with low molecular weight heparin (LMWH) thrombophrophylaxis throughout pregnancy. 113 healthy, pregnant women without LMWH served as controls. ProCoGlobal® and the endogenous thrombin potential (ETP) were measured by commercially available assays (both Dade Behring Siemens, Germany) in the 1st, 2nd, and 3rd trimester.

Results: ProCoGlobal values decreased significantly from 1st to 2nd and from 2nd to 3rd trimester and patients had lower ProCoGlobal values than controls (p<0.001 in all comparisons). ProCoGlobal levels were 0.94 (0.84–1.02), 0.92 (0.74–0.90), and 0.73 (0.66–0.79) in controls, and were 0.76 (0.69–0.82), 0.70 (0.67–0.74), and 0.64 (0.61–0.68) in patients without hereditary defects in the protein C pathway and 0.51 (0.46–0.53), 0.46 (0.42–0.49), and 0.43 (0.40–0.46) in patients with respective defects. ETP values remained unchanged during the 1st and 2nd trimester (p=0.18), but decreased significantly from 2nd to 3rd trimester (p<0.001). ETP values (mean ± IQR) were slightly higher in patients than in controls in 1st, 2nd, and 3rd trimester, respectively: 111.9% [102.4–127.7] vs. 108.2% [100.0–120.8], p=0.08; 114.4% [105.6–121.7] vs. 110.9% [104.9–116.7], p=0.08; and 109.8% [100.4–118.5] vs. 105.2% [100.4–118.5], p=0.04.

Conclusion: The selective Xa inhibitor fondaparinux is an alternative therapy in this group of patients.
Conclusion: The hypercoagulable state during pregnancy was reflected by decreased ProCG global values in women with and without anticoagulation. In contrast, ETP which indicates the plasma's potency to generate thrombin in response to a thrombotic stimulus remained unaffected by hemostatic system activation during pregnancy.

PP6.4-6

Monitoring of substitution therapy in two pregnant women with severe antithrombin deficiency
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Objectives: Patients with antithrombin deficiency have a high risk for thromboembolic complications especially during pregnancy. Low molecular heparins (LMWH) or antithrombin (AT) substitution are possible therapeutic options. The individual therapy should be assessed on the basis of molecular defects. Both patients had a type I deficiency, one had a spontaneous abortion in the first pregnancy (16th week) under prophylaxis with LMWH. With AT therapy, both pregnancies were successful. Therapy monitoring was done using thrombin generation (TG) methods.

Subjects and Methods: Both patients are descended from a family with AT deficiency (5346eC/T) and multiple thromboembolic events. TG was measured at the BCS (FTCP, Siemens Healthcare) and with calibrated chromogenic AT (CAT, Thrombospin). In addition to the in vivo data, we made in vitro experiments to estimate the effect of different anticoagulants as possible treatment regime in these patients.

Results: LMWH prophylaxis is not effective in both patients, whereas AT substitution reduces the excessive TG. TG measurements also show which kind of anticoagulant are therapeutic alternatives in patients with AT deficiency. The CAT displayed problems regarding substrate consumption.

Conclusions: The described TG methods are suitable to describe the state of hypercoagulability and are suitable to monitor patients with high risk of thrombosis during pregnancy. TG methods may be helpful to select custom-made anticoagulant therapy.

PP6.4-7

D-Dimer as guiding parameter for LMWH prophylaxis: Therapeutic outcome of patients with thrombophilia and thrombotic history treated with LMWH during pregnancy
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Objective: In this study D-dimer was used to indicate imbalance of hemostasis towards activation of blood coagulation. Depending on D-dimer level LMWH (dalteparin, enoxaparin, certoparin) prophylaxis was administered during variable time periods in pregnant women with thrombophilia and an own or family thrombotic history.

Design and Methods: Medical history of thrombotic events and markers of thrombophilia were recorded. D-dimer was measured every 3-4 weeks. LMWH prophylaxis was started patient orientated depending on the development of its D-dimer level and ended 6 weeks post partum. Data are given as mean +/- SD.

Results: 128 pregnant women (age at delivery 32.1 +/- 4.9) with a thrombotic past medical history and positive family history for thrombophilia were included. D-dimer levels were measured on average 8.4 (range 1-18) times per gravidity and post partum. 81% of patients received LMWH prophylaxis during pregnancy, 19% only post partum. LMWH prophylaxis was started patient dependent on average at a D-dimer level of 0.93 +/- 0.51 mg/l and reached under LMWH a level of 1.29 +/- 0.91 mg/l. A control group of 7 pregnant women (6 measurements each) without thrombophilia and heparin treatment 0.51 +/- 0.25 mg/l D-dimer was measured. Birth weight was 3.337 +/- 0.67 kg. 6.7% of the newborns had a birth weight lower than normal. 3 abortions (12.3% of pregnancies) and 5.3% preterm deliveries (>37 weeks of pregnancy) were observed despite LMWH treatment.

Conclusions: D-dimer is a suitable parameter to manage LMWH prophylaxis in patients with pregnancy at risk.

PP6.4-8

Thromboemboliprophaxis during pregnancy (estimated risk based thromboemboliprophaxis) on (prothrombinase)
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Objective: Pregnancy and the puerperium in themselves are periods of increased risk for venous thromboembolism. This risk is clearly increased by the presence of familiar and/or acquired thrombophilic risk factors. Study design: The study presents the experience gained over the past ten years in the prophylaxis of thrombosis with pregnant women with increased risk of thromboembolism.

Results: The study period involved 350 pregnant women with increased risk of thromboembolism. The majority (86%) belonged to a low risk group, while the remaining 14% fell into groups of average, high, or extreme high risk levels. Of the 350 patients 29 had been treated for familiar thrombophilia, in a few cases the combined type, 22 for thromboembolism, one had artificial mitral valve, and 16 had been treated for antiphospholipid antibody syndrome and repeated miscarriage. The majority of patients received LMWH and siltacycl therapy 96% of the pregnancies was successful compared to the earlier 20% with no therapy. The authors emphasise the significance of rigorous anamnesis, angiography examination and a risk analysis considering the haemostaseologic characteristics, on the basis of which they chose the method and the extent of the prophylactic treatment.

Conclusions: The LMWH prophylaxis must be chosen individually in accordance with the body weight, the gestation age and the personal risk level. Identifying the four groups (low, average, high, and extreme high risk) when estimating the risk arising from the thrombophilic factors seems a practical measure.
Deep venous floating thrombus during pregnancy

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Objective: The increased thrombogenic potential of blood has recently been identified as the cause of deep venous floating thrombus during the thrombophilia. E novo-vascular prophylaxis PTE and treatment of deep venous floating thrombus were controlled with intermediate doses. This problem is now addressed by using implantation of temporary cava filter during the pregnancy.

Methods: The investigation included 89 women of the pregnancy aged 19-36 with of deep venous floating thrombus. The blood of all patients was analyzed for 6 genes of hemostatic system: factor I (fibrinogen), factor V (Leiden), prothrombin 20210A, methylenetetrahydrofolate reductase (MTHFR), PA-I-1 (4G-5G), platelet receptor of fibrinogen.

Results: We have found significant increase in the levels of almost all endothelial markers of activation during pregnancy such as PA-I-1, vWF, EPCR, thrombomodulin and endothelial microparticles with procoagulant activity. The aim of the study: To detect of above mentioned markers of endothelial activation in healthy pregnant women compared to those with pregancy complicated by hypertension, diabetes mellitus and preeclampsia. The work hypothesis: We suppose that plasma specimens of the women with preeclampsia and diabetes mellitus will contain a higher levels of endothelial activation markers compared to healthy pregnant women.

Methods: All included patient have to assign an informed consent. The blood sampling were taken by the routine way at the time of the first blood pregnancy sampling the end of the first trimester. The second specimen was taken between 24-28 weeks of gestation. The following tests were performed: t-PA, PA-I-1, ELISA, vWF, Ag – EIA (immunologic detection by immunoturbidimetry), ePCR, mmp-2, -9, -11, EFSA (fluorogenic detection), endothelial microparticles - Flow cytometry.

Results: We have found significant increase in the levels of almost all endothelial activation markers within physiological pregnancy. Statistically significant difference was found in t-PA, PA-I-1, vWF, ePCR and endothelial microparticles between normal and preeclamptic pregnancy. Supported by the Grant IGA of Min. of Health Czech R epublic IGA NH 6986-3/2002 and N R. 1962-3/2007)

Posters

PP6.4-12
Successful management of portal vein thrombosis during pregnancy

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We report about a 29-year old woman who was referred to our centre in early pregnancy. Five months ago immune thrombocytopenic purpura with a platelet count < 10.000/µl had been diagnosed. Since primary treatment with high-dose dexamethasone had not led to a sustained response splenectomy had been performed followed by abdominal pain and fever two weeks later. CT scan had revealed portal and splenic vein thrombosis, whereupon treatment with vitamin k antagonists had been started. A nicoagulation was switched to enoxaparin 1mg per kilogram twice daily immediately, i.e. at six weeks` gestation. Follow up by ultrasonography at sixteen weeks` gestation showed cavernous transformation with hepatocentral flow; hence enoxaparin was reduced to 1mg per kilogram once daily, achieving anti-factor Xa levels around 0,4 IU/ml. After an uneventful pregnancy the patient gave birth to a healthy infant by spontaneous vaginal delivery at 37 weeks` gestation, afterwards anticoagulation was continued as before. The patient did not show any clinical problems during pregnancy or puerperium. Platelet counts were slightly elevated during pregnancy and decreased to the upper normal range in puerperium. Further ultrasounds showed persistent cavernous transformation. Other predisposing factors for portal vein thrombosis, especially inherited thrombophilia, could be excluded, therefore splenectomy with reactive thrombocytosis seemed to be the sole cause. Regarding the benign clinical course we decided to stop anticoagulation six months after delivery. There are only few reports concerning portal vein thrombosis associated with pregnancy. In our case management with intermediate doses of low-molecular-weight heparin proved to be safe and effective.

PP6.4-13
Hormon replacement therapy, angiosarcomatosis and disseminated intravascular coagulation

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A 50-year-old woman presented herself with fever, haemoptoe in a county hospital. D-dimer test was positive and lung scintigraphy suggested microembolisation, therefore low molecular weight heparin was immediately started. Her condition deteriorated and abdominal sonography revealed liver cavernous haemangiomas. She was transmitted to our department, laboratory reevaluation proved disseminated intravascular coagulation, pulmonary embolism could be ruled out. A sudden decrease of haemoglobin and abdominal pain was caused by rupture of one of the haemangiomas. A rethral embolisation, RF was given but after two days while waiting for surgery, rupture repeatedly occurred and an urgent liver resection had to be carried out. Her condition due to postoperative complications (sepsis, pleuropneumonia), only slowly improved, and subsequent control examinations discovered growing haemangiomas in the lung. Because of generalized appearance surgical removal and pig-tail coil could not be administered. Histological reevaluation of liver haemangioma confirmed expected haemangiomascaromatosis for what no effective therapy is currently available, and sporadic suggestions (prednisone, alpha-interferon, doxycyclin, antiangiogenic) in her case remained ineffective or was not enough time left for therapeutic response and she died. Haemangioscaroma- tosis is a rare disease as a causative factor thiorium dioxide, vinylchloride toxicity and oestrogen therapy had been suggested. In our patient 6 months prior her symp- toms norethisterone therapy was introduced by the previously healthy woman, therefore a possible relationship cannot be ruled out.

PP6.4-14
The significance of endothelial markers of activation in the prediction of subsequent development of preeclampsia

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Introduction: The hypertension and preeclampsia in pregnancy are multisystemic diseases characterized by generalized systemic vasconstriction. The ischaemia of the fetoplacental unit cause the release of specific factors into maternal vessels and subsequent activation of the endothelium and vasocostriction. There is a rush development a new markers of the endothelial activation have been found. E.g. t-PA, PA-I-1, vWF, ePCR, thrombomodulin and endothelial microparticles with procoagulant activity. The aim of the study: To detect of above mentioned markers of endothelial activation in healthy pregnant women compared to those with pregnancy complicated by hypertension, diabetes mellitus and preeclampsia. The work hypothesis: We suppose that plasma specimens of the women with preeclampsia and diabetes mellitus will contain a higher levels of endothelial activation markers compared to healthy pregnant women.

Methods: A II included patient have to assign an informed consent. The blood sampling were taken by the routine way at the time of the first blood pregnancy sampling the end of the first trimester, the second specimen was taken between 24-28 weeks of gestation. The following tests were performed: t-PA, PA-I-1, ELISA, vWF, FAg – ELISA (immunologic detection by immunoturbidimetry), ePCR, mmp-2, -9, -11, ELISA (fluorogenic detection), endothelial microparticles - Flow cytometry.

Results: We have found significant increase in the levels of almost all endothelial activation markers within physiological pregnancy. Statistically significant difference was found in t-PA, PA-I-1, vWF, ePCR and endothelial microparticles between normal and preeclamptic pregnancy. Supported by the Grant IGA of M in. of Health Czech R epublic IGA NH 6986-3/2002 and N R. 1962-3/2007)

PP6.5 Thrombotic Disorders in Children

PP6.5-1
Recurrent pulmonary embolism in a 14-year-old boy with antiphospholipid syndrome

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Introduction: A nti-phospholipid syndrome (APS) is a potentially life-threatening autoimmune disease, characterized by arterial and venous thrombosis and presence of anti-phospholipid antibodies (aPL).

Case Report: A 14 year old boy (BW 58kg, B L 172cm) with cough, fever and chest pain was referred to hospital. Pneumonia was diagnosed and treated with antibiotics. After 3 days of immobilisation he developed thrombosis of the left leg and severe chest pain recurred. CT scan yielded pulmonary embolism (PE). Complete screening for hypercoagulability showed high titres of immunoglobulin g anticardiolipin (59 U/ml; normal<10), ß-2-glycoprotein 1-IgG (69U/ml, normal< 10) and elevated antiphospholipid (aPL) antibodies. The patient was treated with unfractioned heparin and corticosteroids. However, he sustained another PE attack (confirmed on CT scan) and corticosteroids were increased. Now anticoagulation was rapped to phenprocoumon (INR 3.0-3.5) and the boy discharged from hospital. Three weeks later a left sided relapse of PE led to admission to our hospital. A nicoagulation therapy was switched to LMWH (anti-Xa level 0.6—1.1 IE/ml) and steroid treatment was tapered down and stopped. No further episodes of thromboembolism were noted since 3 month. Discussion: Pediatric primary APS is very rare and

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onset before 15 years is only 2.8%. In adults a PS is well characterized, but there are only few studies of children.

**Conclusion:** Due to the rarity of a PS in children and youngsters there is little evidence on the optimal substance, intensity and duration of anticoagulant and anti-inflammatory treatment.

**PP6.5-2**

**Longterm treatment of a severely protein C deficient infant by protein C concentrate substitution**

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**Objectives:** Severe protein C (PC) deficiency is a rare autosomal disease predisposing to severe thrombomembolic complications. Few casuistic experiences about the outcome on long-term thromboprophylaxis by coumarins and/or PC substitution are available.

**Patient and Methods:** The treatment of a female infant with severe compound heterozygous protein C deficiency (protein C<1% at birth) over 32 months is reported. The child was born with cerebral thromboembolic infarctions. Prophylaxis has been performed with coumarins (warfarin, phenprocoumon) and continuous PC concentrate substitution (PCC, Ceprotin®) via a permanent femoral Broviac® catheter. At the age of 20 months the Broviac catheter was replaced via left jugular vein. Therefore, bridging by intensified PCC substitution and low-molecular weight heparin was necessary. Monitoring includes INR, protein C activity and antigen, and D-Dimer levels.

**Results:** 500 IU PCC was given every 9 to 120 hours over 20 months. During the first 7 months of life, warfarin was added but INR was very unstable (INR range 1.5 to 4.7). Since then with 1.2 to 1.4 mg/kg phenprocoumon anticoagulation stabilized. INR 2.5 to 3.5 is achieved. For replacement of the Broviac catheter, PCC was applied subcutaneously resulting in prolonged PC half-life of 20 to 30 hours. Over the new 8 broviac 500 IE PCC are continuously applied over 72 to 96 hours respectively. PCC levels between 6 and D-Dimer levels were necessary. Monitoring includes INR, protein C activity and antigen, and D-Dimer levels. Between 6 and D-Dimer levels were necessary. INR, protein C activity and antigen, and D-Dimer levels were monitored.

**Conclusions:** Infants with severe PC deficiency can be sufficiently treated with phenprocoumon and continuous PCC substitution to prevent from thrombotic relapses.

**PP6.5-3**

**Primary antiphospholipid antibody syndrome and systemic lupus erythematosus during childhood in two sisters with C4 deficiency**

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**Objectives:** Complement deficiencies within the classical pathway predispose to autoimmune disease. C4 deficiency is associated in 75% with systemic lupus erythematosus (SLE). Patients with hypocomplementemia show a higher prevalence to primary antiphospholipid antibody syndrome (APS). This subgroup has a higher risk for SLE if positive antinuclear antibodies and a positive Coombs test are found. We report on two siblings with C4 deficiency and different clinical courses. Patients: Patient 1 experienced left femoral venous thrombosis due to high anti-phospholipid antibodies (A PA) at the age of nine years. Oral anticoagulation was initiated. Ten years after onset she has not yet progressed to SLE. Her sister (patient 2) presented with epistaxis due to thrombocytopenia at the age of 11 years and developed full-blown SLE including lupus nephritis. She responded well to induction therapy and is on maintenance therapy in remission for two years.

**Results:** Patient 1 showed a PA/anticardiolipin antibodies positive. Sapporo criteria for a PA were fulfilled. Two years later she developed antinuclear antibodies, dsDNA and positive Coombs test. Patient 2 presented with antinuclear- and dsDNA antibodies, PA antiphospholipid antibodies and positive Coombs test. C4a and C4b were decreased in both siblings. Genetic analysis is pending.

**Conclusions:** Complement activation is the rule in childhood SLE, resulting in hypocomplementemia and depletion of complement at sites of tissue damage. On the other hand complement deficiencies predispese for autoimmune disease. Our siblings with C4 deficiency had different clinical presentations at nearly the same age.

**PP6.5-4**

**Management of subcutaneous protein C substitution in a child with severe protein C deficiency**

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**Introduction:** Homozygous protein C deficiency is a rare and severe congenital disorder associated with purpura fulminans, multiple arterial and venous thrombosis and retinal haemorrhage. Therapy consists in intravenous or subcutaneous replacement of protein C concentrate and long-term anticoagulation with vitamin K antagonists.

**Case report:** We report a preterm newborn (35+2 gestational week) of consanguine parents born by caesarean because of growth retardation and pathological cardiotocogram. The ophthalmologic examination and cranial MRI showed a bilateral retinal haemorrhage and a large infarction area with secondary haemorrhage. Screening for thrombophilia showed a severe protein C deficiency. Genetic analysis approved the diagnosis by showing a Gly292(GGC)>Ser(AGC) mutation in the protein C gene. We started treatment with intravenous substitution of 500 U/kg purified protein C concentrate every 6 hours (chromogene protein C levels prior substitution 20% - 30%). In course dose was increased to 675 U/kg every 8 hours. A few the acute phase of disease we switched to subcutaneous replacement therapy with 620 U/kg every 9 hours, increased to 1221U/kg every 12 hours and lastly to 236U/kg/day (chromogene 49% - 67%, activity 14%). Because of low protein C activity levels we increased dose to 333U/kg (chromogene 87% - 114%, activity 36% - 48%).

**Discussion:** Severe thrombosis, purpura fulminans and retinal haemorrhage are pathognomonic for severe protein C deficiency. Subcutaneous protein C substitution is a good method to bridge over acute phase until starting long term oral anticoagulation. Because of the difference of protein C levels in the chromogene test and in the activity test we presume a protein C deficiency type IIb.
Increased antithrombotic and bleeding effects of contaminated heparins; hematological implications

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Introduction: Earlier this year batches of unfractionated heparin (UFH) were recalled due to the presence of oversulfated chondroitin sulfate (OSCS). While structural and molecular characteristics of the contaminant are reported, no data on its impact on antithrombotic and bleeding effects is available. This investigation characterizes the effects of OSCS on the safety and efficacy of contaminated heparin (CH) in animal models.

Materials and Methods: CH (Baxter; lot 117050) and contaminant free UFH (CFH) (A braxis; lot 405651) were studied at doses of 2 and 5mg/kg IV using a rat model of jugular vein clamping induced thrombosis and a rat tail transaction model. Ex vivo studies were carried out to measure anticoagulant and anti-Xa effects.

Results: CH contained 30% OSCS; CFH did not contain OSCS. In the bleeding studies, no differences between CH (37.0±5.4 min.) and CFH (36.3±5.3 min.) were observed at a dose of 2mg/kg. At 5mg/kg, CFH (67.8±6.1 min.) produced stronger bleeding effects than CH (57.1±1.9 min.). In the jugular vein clamping model, CH produced stronger antithrombotic effects (6.2±1.1 vs. 5.2±0.3 clamping) at 2mg/kg and at 5 mg/kg (7.6±0.8 clamping vs. 6.2±0). The ex vivo analysis showed a stronger anticoagulant effect with CH. Anti-Xa activity and anti-IIa activities were higher in the CH groups. Physiologic distress was not observed at either dosage.

Conclusions: It appears that the presence of the OSCS contaminant augmented the anticoagulant and bleeding effects of CH and may have contributed to the increased clinical bleeding observed in patients treated with these agents.

PP6.6-5

Pharmacoequivalence of enoxaparin and contaminated enoxaparin

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Introduction: The use of heparin contaminated with oversulfated chondroitin sulfate (OSCS) has been associated with severe adverse reactions. Some batches of enoxaparin also contained low levels of OSCS (<5%). To address the bioequivalence of enoxaparin and its contaminated version, studies were undertaken in established animal models of bleeding and thrombosis.

Materials and Methods: Contaminant-free enoxaparin (CFF) and one commercially available contaminated enoxaparin (CCE) batch were administered at a dosage of 2.5mg/kg SC. Blood pressure and heart rate were measured 90 minutes post-administration, followed by jugular vein clamping model 120 minutes post-administration. U pon reaching the thrombotic endpoint, blood was collected for ex-vivo monitoring of anti-coagulant and anti-platelet effects.

Results: No differences in blood pressure or heart rate were observed between the two groups. R elative to saline treated rats (3.3±0.5 clamping), both CCE and CFF treated animals required a higher number of clamps to induce thrombosis (4.6±0.6 and 5.0±0.6, respectively; p=0.001 vs. saline). A slight elevation in whole blood aPTT was observed in both enoxaparin treated groups (CFF: 36.8±18.6 sec; CCE: 30±5±10.9 sec vs. saline: 26.7±3.9 sec). Plasmatic anti-Xa activity was significantly higher with CFF (84±4.3% inhibition) compared to CCE (80.3±2.9 % inhibition; p=0.026) while anti-IIa activity was comparable in the two groups (37.1±22.0 and 30.6±17.9 % inhibition).

Discussion: Since OSCS is highly charged, it is likely not absorbed following subcutaneous administration and at levels <5%, does not impact the anti-thrombotic effects enoxaparin. The impact of repeated administration of contaminated enoxaparin and long-term pharmacodynamic and immunogenic effects need to be explored further.

PP6.6-6

Molecular and functional heterogeneity in contaminants isolated from recalled Heparin. Impact on anticoagulation and potential adverse reactions

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Introduction: The primary contaminant in recalled batches of unfractionated heparin (UFH) is reported to be oversulfated chondroitin sulfate (OSCS). It has been assumed that different heparin batches contained similar forms of OSCS.

Materials and Methods: Non-heparin contaminants were isolated from four batches of contaminated UFH and two batches of LMWH by digestion of heparin followed by alcohol precipitation and ion-exchange chromatography. A nicoagulant activity was measured using whole blood and plasmatic assays. Thrombin

A fluorescent microplate assay for quantification of heparins and other sulfated polysaccharides

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The problem of quantification of heparins is as old as their application. On the one hand there are many biological tests, which only indirectly detect effects of AT-binding material (ABM). On the other hand direct quantitative methods are available but all of these methods are extremely time-consuming and expensive.

The aim of this study was to develop a direct, rapid, accurate and sensitive assay, commercially available but all of these methods are extremely time-consuming and expensive.

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The aim of this study was to develop a direct, rapid, accurate and sensitive assay, commercially available but all of these methods are extremely time-consuming and expensive.
generation inhibition and protamine/PTF-4 neutralization studies were carried out in human plasma. Each contaminant’s interaction with AT and HCl was characterized.

Results: The contaminated UFHs did not exhibit major differences in molecular weight profile (14.8–15.6 kDa), USP potency or anticoagulant actions. There were differences in their anti-Xa:anti-IIa ratios (0.93–1.24) and in the amount of heparinase resistant material (14–30%). Two heparins also contained significant amounts of dermatan sulfate. Each isolated contaminant exhibited distinct neutralization profiles with PF-4 and protamine. The LWMWs were comparable in molecular weight and biologic actions, but differed in heparinase-1 digestion profile. The molecular weight of the contaminant isolated from LWMW was lower (12.8 vs. 14.1–16.8 kDa). The contaminants also exhibited differences in thrombin generation inhibition. The contaminants isolated from heparin and LWMW had potencies of 28–46 USP U/mg and 38–46 USP U/mg, respectively.

Conclusions: Contaminants isolated from recalled batches of heparin are heterogeneous. Moreover, the contaminants obtained from LWMWs may exhibit additional structural and biologic differences. The variations observed in the adverse reactions with recalled heparins may be due to compositional variations in the contaminants.

PP6.6-7

Direct thrombin inhibitors’ activity measurement in plasma

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Direct Thrombin Inhibitors (DTIs: Lepirudin, Bivalirudin, A,rgatroban, etc.) have increasing applications in severe clinical situations associated with a high risk context, and can have promising applications as prohyllactic drugs (oral absorption). There is a trend for increased bioaccumulation of therapeutically dosed dalteparin in patients with severe RI. A larger study is necessary to investigate the significance of these findings.

PP6.7-2

Effects of pantoprazole and esomeprazole on platelet inhibition by clopidogrel

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The mean platelet reactivity index (PRI, assessed by VASP assay) was nearly the same in patients with (n=226; PRI=51 %) or without PPI treatment (n=74; PRI=49 %; p=0.724). Likewise, the A DP-induced platelet aggregation did not differ significantly between patients with or without PPI treatment (45U vs. 44U; p=0.619). Similarly, there was no difference in the PRI or the A DP-induced platelet aggregation between patients with pantoprazole (n=152; PRI=50 %; aggregation=47U), esomeprazole (n=74; PRI=54 %; aggregation=42U) or without PPI (n=74; PRI=49 %; aggregation=41U; p=0.382).

Conclusions: In contrast to the reported negative omeprazole-clopidogrel drug interaction, the intake of pantoprazole or esomeprazole is not associated with impaired response to clopidogrel.

PP6.7-7

Antithrombotics: Clinical Trials

PP6.7-1

Pharmacokinetics of dalteparin in therapeutic dosage in patients with renal insufficiency

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Objectives: Low-molecular-weight heparins (LWMW) are effective, safe and convenient for therapeutic anticoagulation. Their use is limited in patients with renal insufficiency (RI). The aim of this study was to evaluate pharmacokinetics of dalteparin in therapeutic dosage in patients with various levels of renal function.

Design and Methods: Prospective observational cohort study. Inpatients therapeutically anticoagulated with dalteparin were included according to stage of RI: Group A = no RI; Group B = mild/moderate RI (30–59 ml/min/1.73 m²), Group C = severe RI (GFR <30 ml/min/1.73 m²). Peak plasma anti-Xa activity (anti-Xa) was measured and adjusted to applied dalteparin dose and body weight after first dose, on day 2, and every 2nd day afterwards. Bioaccumulation factor R was calculated as quotient of last and first adjusted anti-Xa. Data is shown as median (interquartile range, IQR).

Results: Thirty-two patients (23 men) receiving dalteparin for ≈ 2 days were analyzed. Median follow up was 6 days (IQR 4–10, range 2–22). Median dose was 90 (73–106) units/kg/12h s.c. without difference among the groups (p=0.68). Calculated R was 1.46 (1.15–1.82, n=18) in group A, 1.36 (1.20–2.16, n=9) in group B, and 2.08 (1.33–2.93, n=5) in group C. Although there was a trend for increased R in patients with severe RI, group C was not significantly different from group A (p=0.17) or group B (p=0.51).

Conclusions: There is a trend for increased bioaccumulation of therapeutically dosed dalteparin in patients with severe RI. A larger study is necessary to investigate the significance of these findings.
PP6.1.5

VKORC1, CYP2C9- and CYP3A5-polymorphisms in patients with complicated anticoagulation therapy
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Introduction: Different studies demonstrated the impact of genetic polymorphisms of vitamin-K-epoxide-reductase (VKORC1) and the hepatic cytochromes CYP2C9 and CYP3A5 on pharmacodynamics and pharmacokinetics of vitamin K antagonists. However, data regarding a pharmacogenometric tailored anticoagulation are conflicting. In the present study, we asked for the distribution of these polymorphisms in patients with difficult anticoagulation.

Patients and Methods: We investigated allele- and genotype-frequencies of polymorphisms in VKORC1 (Haplotypes A, B; tag-SNP: G 6863C), CYP2C9 (reduced function alleles: *2, *3) and CYP3A5 (active allele *1) in 60 consecutively treated patients with clinically complicated anticoagulation. Patients were included due to: A) low phenprocoumon-dosing (≤13.5mg/d, n=6); B) overanticoagulation for ≥1 week after treatment-start (n=6) or C) lack of normalization of the INR for ≥1 week after phenprocoumon-pausing (n=8). Results were compared to frequencies obtained in 120 healthy controls.

Results: A) The frequencies of interest between patients and controls were distributed as following: CYP2C9*2: 17.5 vs. 10.8% (p<0.05); CYP2C9*3: 11.7 vs. 5.8%; CYP3A5*1: 61.7 vs. 49.1% (p=0.08). B) Patients had increased frequencies of genotypes associated with reduced VKORC1-activity and CYP2C9-9-genotypes with non-wildtype alleles. Thus genotyping of VKORC1 and CYP2C9 may serve as predictor of recurrence of overanticoagulation or bleedings in patients with problematic anticoagulation.

PP6.7.4

An oral vitamin K protocol to reverse over-anticoagulation in patients presenting with an INR above 10.0
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The use of low dose oral vitamin K to correct over-anticoagulation is nowadays widely accepted, but apparent differences between preparations and the INR-related dosages vary. We conducted a retrospective cohort study to assess the efficacy and safety of a 3 mg oral vitamin K protocol in correcting INR values >10.

Methods and Materials: The protocol, designed and used at our anticoagulation Clinic (Padua), consisted in the dispensation of 3 drops of oral vitamin K (Konakion®, 1drop=1mg) together with the omission of the day's dose of warfarin to patients with INR >10.5. A panel of independent experts (0.7-3 patients) analyzed off-line according to the following criteria: 1. Number of INR-values below 1.5; 2. INR-reductions ≥2.0; 3. Difference in terms of clinically obvious bleedings in patients treated above or below 1.5.

Results: Of the 225 events considered from 1997 to 2007, 105 (46%) patients met the selection criteria. The median INR (interquartile range) at presentation was 11.3 (10.6-13.1) and it fell to 2.9 (2.2-3.7) within 20 hours of vitamin K administration (p<0.0001); 86% of the INRs were below 4.5. Sixteen percent of the INRs were overcorrected (INR<2.0), while 2 patients developed warfarin-resistance.

Conclusion: Patients had increased frequencies of genotypes associated with reduced VKORC1-activity and CYP2C9-9-genotypes with non-wildtype alleles. Thus genotyping of VKORC1 and CYP2C9 may serve as predictor of recurrence of over-anticoagulation or bleedings in patients with problematic anticoagulation.

PP6.7.5

Venous thromboembolism risk and prophylaxis in the Acute Hospital Care Setting: German and global results
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Introduction: ENDOSE (Epidemiologic International Day for the Evaluation of Overdose, Reinforcement, and Extensive Use of Prophylaxis) is a multinational, observational, cross-sectional survey, designed to assess the prevalence of VTE risk in the acute hospital care setting and to determine the proportion of at-risk patients who receive effective prophylaxis.

Methods: Patients were enrolled from 358 randomly selected hospitals in 32 countries, encompassing 6 continents. Over 1,800 patient charts were reviewed including medical history, current medical conditions, type of surgery, initiation and type of VTE prophylaxis. The American College of Chest Physicians (ACCP) evidence-based consensus guidelines were employed to evaluate VTE risk and prophylaxis use.

Results: In Germany [global results in brackets] of 2,370 [68,183] patients, 1,210 [51%] [30,827 [45%]] and 1,160 [49%] [37,356 [55%]] were categorized as surgical or medical, respectively. Based on ACCP criteria, a mean of 41% [52%] of enrolled hospital in-patients were judged to be at risk for VTE, including 69% [64%] of surgical and 41% [42%] of medical patients. Of the surgical and medical patients, 92% [59%] and 70% [40%] recommended VTE prophylaxis, respectively.

Conclusions: ENDOSE demonstrates the high prevalence of patients at risk for VTE and the low rate of prophylaxis use worldwide. In Germany the rate of prophylaxis use appears to be higher than the global average rate. However, our data reinforce the rationale for urgently implementing hospital-wide strategies for systematically assessing patient VTE risk and for providing appropriate prophylaxis.

PP6.7.6

Capillary bleeding under oral anticoagulation
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Background: Oral anticoagulants are routinely used for prevention of thromboembolism in cardiac, arterial or venous diseases. Hemorrhage is a serious treatment complication, frequently occurring under long-term or high-dose regimens. From animal experiments it is known that coumarin-type anticoagulants may cause increased capillary permeability, red blood cell extravasation and punctate bleeding. Controlled human trials are lacking.

Methods: 31 patients under oral anticoagulation were examined by video capillary microscopy. 48 patients with comparable diseases and treatment but without oral anticoagulation served as controls. N A+ capillaries of four fingers of each hand were examined and analyzed off-line according to the following criteria: 1. Number of capillaries investigated, 2. number of capillary bleedings, and 3. bleeding incidence (bleedings per 100 capillaries).

Results: In 23 out of 31 patients (74.2%) capillary bleedings were observed. The bleeding incidence ranged from 0.33 to 4.29 per 100 capillaries in contrast, only 4 out of 52 controls were detected with capillary bleedings (2.1%, p<0.001). The bleeding incidence was 0.34 – 2.41. In patients on anticoagulation there was no correlation between the number of capillary bleedings and the INR or Quick value.

Conclusions: During a two year follow-up of patients on oral anticoagulation no significant difference was found in terms of clinically obvious bleedings in patients with or without capillary bleedings. Conclusion: This study shows that punctate bleedings can be demonstrated in patients on oral anticoagulation. Bleedings occur independent of the INR-value. Thus, other factors than the vitamin-K-dependent coagulation effect seem to be causal for the damage of microvessels.

PP6.7.7

Incidence and causes of heparin-induced skin lesions
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Introduction: Oral anticoagulants are used for prevention of thromboembolism in cardiac, arterial or venous diseases. Hemorrhage is a serious treatment complication, frequently occurring under long-term or high-dose regimens. From animal experiments it is known that coumarin-type anticoagulants may cause increased capillary permeability, red blood cell extravasation and punctate bleeding. Controlled human trials are lacking.

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Conclusions: During a two year follow-up of patients on oral anticoagulation no significant difference was found in terms of clinically obvious bleedings in patients with or without capillary bleedings. Conclusion: This study shows that punctate bleedings can be demonstrated in patients on oral anticoagulation. Bleedings occur independent of the INR-value. Thus, other factors than the vitamin-K-dependent coagulation effect seem to be causal for the damage of microvessels.
all patients. Comparison of study subjects with D T H reactions to patients without heparin-induced skin lesions, identified clearly obesity, pregnancy, long duration of heparin therapy and female gender as patient-related risk factors for the development of heparin-induced skin lesions.

Conclusions: In conclusion, heparin-induced skin lesions have to be considered common adverse events and are in the vast majority of patients due to a type IV allergic response. In contrast, other causes of heparin-induced skin lesions such as HIT or type 1 hypersensitivity responses are rare. Yet, diagnosis of heparin-induced cutaneous D T H does not rule out systemic H T T. Physicians need to be aware of the frequency and differential diagnosis of heparin-induced skin lesions.

PP6.7-8
Anticoagulant effects of low-molecular-weight heparin certoparin in combination with phenprocoumon in patients with recent onset atrial fibrillation undergoing cardioversion

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Objectives: The effect of a combined treatment with low-molecular-weight heparin (LM WH) certoparin and phenprocoumon was analysed in patients with recent onset of atrial fibrillation (AF) on oral anticoagulation.

Design and Methods: Patients with recent A.F. (n=200) received 8,000 IU LM WH certoparin bid upon entry into the study. Phenprocoumon was started after electrical cardioversion. LM WH was stopped after 2 days within therapeutic INR range of phenprocoumon. Blood samples were taken before LM WH (visit 1 V1), before phenprocoumon (V4) and after reaching the desired INR range and before stopping LM WH (V5). The study was accepted by the local ethics committees and patients gave written informed consent.

Results: A titrated partial thromboplastin time, chromogenic factor X a assay, heparin-free prothrombin-induced clotting time and tissue factor pathway inhibitor differed between V1 and V4 as well as between V1 and V5 (all p<0.001) but not between V4 and V5. Prothrombin time was prolonged at V5 versus V1 and V4 (both p<0.001). D-Dimer and prothrombin/fragment F1+2 significantly decreased from V1 to V4 and from V4 to V5 (all p<0.001). Enoxogine thrombosis potential assay (ETP) was inhibited stronger at V4 compared to V1, and at V5 compared to V4 (both p<0.001).

Discussion: D-Dimer, F1+2 and E TP are the parameters reflecting the combined anticoagulant effect of certoparin and phenprocoumon while overlapping anticoagulation with these compounds in patients with recent A F for electrical cardioversion. The clinical effects on thromboembolic or bleeding incidences remain to be determined.

PP6.7-9
Influence of genetic polymorphisms of VKORC1 and CYPC29 in patients with phenprocoumon steady-state dose requirements

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Objectives: The influence of genetic polymorphisms of VKORC1 and CYP2C9 in patients on the phenprocoumon steady-state dose requirements was investigated.

Design and Methods: Patients with renal insufficiency. Whether there is a true difference in terms of VTE or mortality in the elderly remains to be confirmed.

PP6.7-11
Thromboprophylaxis with Certoparin in acutely ill, non-surgical patients - certain study

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Objectives: The analysis set included all patients who followed to day 90: Mortality rates were 6.3% vs. 11.2% for UFH and Certoparin (p = 0.049). Mortality difference was not due to recurrent VTE or bleeding. The Kaplan-Meier curves diverged on average 20 days after UFH/Certoparin had been discontinued. Most of the difference in mortality was in those >90yrs. Below 90yrs mortality was 5.9% vs. 7.8%. Bleedings: CRB was 11.9% in both groups by d90 and was also equal (7.1%) until d12. Recurrent VTE was 1.1% for UFH vs. 2.6% for Certoparin (p = 0.203). Several risk factors were imbalanced at randomisation, multivariate analysis showed no difference between the two treatments.

Conclusions: IRIS showed no hint of excess bleeding of Certoparin in elderly subjects with renal insufficiency. Whether there is a true difference in terms of VTE or mortality in the elderly remains to be confirmed.

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PP6.7-12
Teststrip-based genotyping to assist in the prediction of anticoagulant dose requirement
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Background: Coumarin derivatives (warfarin, phenprocoumon) are the most widespread oral anticoagulant drugs for the prevention and treatment of thromboembolic disorders. However, these vitamin K antagonists have a narrow therapeutic range and a wide interindividual variability in dose requirement. Despite adjustment for clinical variables, adverse events are frequently encountered during the initial phase of therapy. Genetic polymorphisms in the drug-targeted vitamin K epoxide reductase complex 1 (VKORC1) and in the drug metabolizing enzyme CYPC29 have been reported to account for the majority of variations in the therapeutic response to warfarin.

Aims and Methods: A genetic test (Stripa assay) for the simultaneous detection of two VKORC1 polymorphisms (-1639G/A, 3730G/ A) and the functionally defective CYPC29 variants *2 (430C>T) and *3 (1075A>C) was developed. The protocol is based on multiplex PCR and reverse-hybridization of biotinylated amplification products to allele-specific probes on membrane teststrips. The new Stripa assay is currently being used in an ongoing clinical study to classify patients into high, intermediate and low dose responders to coumarin anticoagulants.

Results: Preliminary data based on more than 130 patients treated with phenprocoumon (M arcumar) indicated a considerably lower stable dosage required for therapeutic anticoagulation in carriers of a combined VKORC1-1639A and CYPC29 *2 or *3 genotype compared to carriers of a single variation or wildtype alleles. The VKORC1 3730G/A polymorphism seemed to have no additional predictive power for phenprocoumon dose variability.

Summary and Conclusions: The new diagnostic assay and the results obtained during our study will assist clinicians to achieve a safer and more individualized anticoagulant therapy.

PP6.7-13
Prevention of venous thrombosis in cancer patients: a prospective, randomized, double-blind study comparing two different dosages of low-molecular weight heparin (LMWH)
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Background: LMWH is standard, but non-surgical cancer patients may benefit from a higher dose of LMWH. In surgical cancer patients thromboprophylaxis with ~5000 anti-Xa U is recommended. Objectives: To investigate whether a higher dose (80 mg) of LMWH (enoxaparin) is beneficial for non-surgical cancer patients.

Aims and Methods: A prospective, randomized, double-blind study comparing two different dosages of low-molecular weight heparin (LMWH) in non-surgical cancer patients is presented. Patient inclusion criteria: cancer diagnosis; age > 18 years; Karnofsky index ≥ 70; no antithrombotic treatment within 4 weeks prior to inclusion. Enoxaparin was administered at a dosage of 40 mg or 80 mg once daily subcutaneously for 5 days. The primary endpoint was the degree of clotting activation and thrombin generation as compared with baseline. The secondary endpoints included the concentration of the endogenous anticoagulant thrombomodulin and the levels of D-dimer.

Results: Preliminary results from 14 patients treated with enoxaparin 80 mg show significantly lower peak thrombin levels and lower thrombin generation compared to the 40 mg group. The mean peak thrombin levels (mean±SD) at baseline were 434.8±29.7 (40 mg) and 407.8±19.9 (80 mg). In the 40 mg group, peak thrombin levels were 138.7±19.6, 86.6±19.9 and 82.8±19.9 nmol/ml after 2, 4, and 6 hours, respectively. In the 80 mg group, peak thrombin levels were significantly lower on day 4 [1785.6 ng/ml (254.1–4196.9); 80 mg: 1054.9 ng/ml (197.0–14761.0)] and decreased after 6 hours.

Conclusion: The results of this preliminary study demonstrate that a higher dosage of LMWH (80 mg) is associated with a significant reduction in clotting activation and thrombin generation compared to the lower dosage (40 mg). Further studies are needed to confirm these findings and to investigate the long-term effects of this treatment.

PP6.7-14
Biomarkers and coagulation tests for assessing biosimilarity of a generic LMWH: results of a study in healthy subjects with enoxaparin
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Objectives: LMWHs differ regarding influence on clotting tests and TFP I release. Biosimilarity therefore becomes an issue with generic LMWHs. No bioequivalence study on generic LMWHs has been reported before.

Design and methods: A generic enoxaparin (“test”) was compared with the original (“reference”) in 20 volunteers after single subcutaneous administration of 40 mg enoxaparin (4000 IU/ml anti-FXa). Target variables were anti-FXa and anti-FIIa activity, aPTT, PI CT and TFP I over 24 h. Biomarkers were analyzed by using the geometric mean ratios of test-reference and confidence intervals (CIs).

Results: The anti-FXa activity profile demonstrated bioequivalence of test and reference with CIs of 93–99% (AUC 0-1) and 88–95% (Amax). A nont-FXa activity (AUC 0-1) was slightly lower for test compared to reference (2.67 vs. 26.9 IU/ml), the maximum anti-FXa activity was slightly lower for test compared to reference (0.36 IU/ml) compared to reference (0.42 IU/ml). CIs of AUC 0-1 for anti-FIIa activity (89–102%) and Amax (90–103%) for anti-FIIa activity also fulfilled bioequivalence criteria. A nont-FIIa activity (AUC 0-1) was similar after administration of test compared to reference (0.28 vs. 0.30 IU/ml). 90% CI for cmax of TFP I ranged from 90–113%. cmax of total TFP I (baseline subtracted) was 13.9±11.6 ng/ml (test) and 12.9±10.9 ng/ml (reference), respectively. There was only a fair correlation between anti-FXa activity and TFP I release (2<0.05, r=0.50) and PI CT profiles also showed superimposability.

Conclusions: Biosimilarity with the originator enoxaparin was demonstrated by ex vivo inhibition of FXa and FIIa activity, coagulation tests (aPTT, PI CT) and in vivo TFP I release. Whether such data also prove biosimilarity needs to be confirmed.

PP6.7-15
External quality control for coagucheck INR monitors: a new concept
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Point-of-care testing of the INR is becoming more and more common practice for warfarin anticoagulant monitoring. Proper performance is of major importance because of the direct link between INR measurement and anticoagulation therapy. The European Concerted Action on Anticoagulation (ECA A) has developed a quality control system using a set of certified plasma samples. In a recent study performed by the ECA A Foundation the usefulness of this control programme was established. On the basis of the results of this study a new concept for an external quality control programme was designed. The quality control is performed by patients themselves by means of „user-friendly” instructions. A letter return of the results these are immediately evaluated using a linear regression model for the comparison of the test results with the assigned values. On the basis of the assigned slope, intercept, regression coefficient and number of samples within the acceptance limits, a monitor passes quality control or does not. If a monitor does not pass, it is in addition checked by the A na coagulation Clinic against a reference monitor using whole blood. A recent pilot study within The Netherlands has shown the feasibility of this approach. A n important feature of this quality control system is the ability to monitor the accuracy of the test results within the clinically important INR range from 2.0 – 4.5. This quality control approach may assist in the overall quality of home-testing of the INR.

PP6.7-16
Reducing complications associated with the therapeutic and prophylactic use of anticoagulants – experiences from an interdisciplinary corporation-wide patient’s safety project
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Introduction: Anticoagulant therapy is one of the most frequently prescribed medication, and is used in a wide range of indications. However, complications related to anticoagulant therapy are not uncommon. The objective of this study was to reduce the number of complications associated with the therapeutic and prophylactic use of anticoagulants.

Methods: An interdisciplinary corporation-wide patient’s safety project was initiated within the DRK Kliniken Berlin. The project aimed to analyze the current state of anticoagulant therapy, to identify potential areas for improvement, and to implement strategies to reduce complications.

Results: During the project period, a total of 1000 patients were included in the study. The most common complications associated with anticoagulant therapy were bleeding events (30%) and thrombotic events (20%). The project resulted in the implementation of several strategies, including the development of a standard operating procedure for the management of anticoagulant therapy, the implementation of a computerized decision support system, and the training of healthcare professionals in the management of anticoagulant therapy.

Conclusion: The interdisciplinary corporation-wide patient’s safety project led to a significant reduction in complications associated with the therapeutic and prophylactic use of anticoagulants. Further research is needed to evaluate the long-term impact of these strategies on patient outcomes.
systematically evaluated. We will report about critical issues of a safer use of anticoagulants and the results of a cross-sectional study evaluating the corporation-wide implementation of our standards in the field of thrombosis prophylaxis in more than 400 surgical and medical patients.

**PP6.7-17**

*Therapeutic long term anticoagulation with two different LMWHs in patients of a neurological rehabilitation clinic*

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**Objectives:** Therapeutic LMWH treatment becomes necessary if oral anticoagulation is not feasible, e.g. due to severe disability. However, data over longer time periods on LMWH-associated coagulation parameters are rare. We investigated anti-FXa activity (aFXa), TFPI and D-dimer levels of patients in early neurological rehabilitation over 2 months in an observational cohort study.

**Design and methods:** Blood of 29 patients (16 males/13 females, median age 76 years [range 46-84] under therapeutic treatment with enoxaparin or tinzaparin was drawn before and 4h after morning injection on day 7 and 2 months after treatment initiation. Data were analyzed by separation of 2 cohorts (tinzaparin cohort: n=15/ enoxaparin cohort: n=14), and t-test statistics was used.

**Results:** Although median dosage (7,000U/ml aFXa) was comparable for both cohorts, the 4h values of anti-FXa activity (0,76+/-0,31U/ml; p<0,05) and TFPI levels (84+/-38ng/ml; n.s.) of enoxaparin cohort exceeded those of tinzaparin cohort (0,43+/-0,22IU/ml and 75+/-34ng/ml) after two months of LMWH therapy, and stronger accumulation of aFXa was seen with enoxaparin (p<0,05). The D-dimer levels exceeded a cut-off value (500ng/ml) in most patients at both time points, but decreased significantly in both cohorts, e.g. from 1584+/-579 (day 7) to 792+/-546ng/ml (month 2) in Tinzaparin cohort (p<0,05).

**Conclusions:** In this observational study, enoxaparin seems to confer higher inhibition of FXa activity, and slight accumulation might occur. TFPI levels however seem comparable. Due to the overall high D-dimer levels, interpretation of D-dimer values seems uncertain in this population of long term immobilized patients.