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Scleroderma, myositis and related syndromes

FRI0363 KL6 AND NOT CCL-18 IS A PREDICTOR OF EARLY PROGRESSION IN SYSTEMIC SCLEROSIS RELATED INTERSTITIAL LUNG DISEASE

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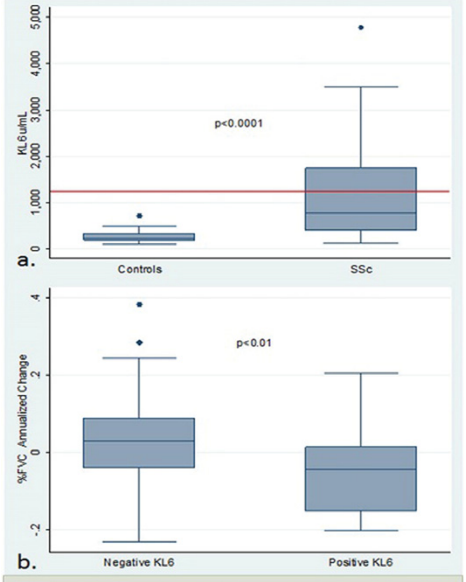
Background: Pneumoproteins are attractive biomarker candidates in systemic sclerosis (SSc) related interstitial lung disease (ILD) because they are easily obtainable and lung-specific. KL-6 and CCL-18 (PARC) have been previously reported as promising predictive biomarkers of lung parenchymal damage in multiple disorders including SSc-ILD.
Objectives: Our goal was to determine the predictive significance of these two pneumoproteins for forced vital capacity % (FVC) decline within the first year of follow-up in patients with early SSc-ILD, in order to inform individualized care in routine clinical practice and facilitate enrichment strategies in clinical trials.
Methods: GENISOS (Genetics versus ENvironment In Scleroderma Outcome Study) cohort patients who had ILD verified by imaging and available pulmonary function tests at enrollment plus 12–18 months thereafter, were included in this study. All patients had disease duration ≤5 years at enrollment. FVC, expressed as percentage of predicted value was used as surrogate for severity of ILD. Annualized percent change in FVC at one year follow up was calculated. Baseline demographic, clinical variables and two pneumo-proteins, KL-6 and CCL-18 were investigated. KL-6 and CCL-18 were measured in the plasma by commercially available, validated ELISA kits. Linear regression with baseline clinical and demographic variables as independent variables was performed in univariable and multivariable models. Only variables that reached p<0.1 were included in the multivariable analyses.
Results: A total of 82 patients with early SSc-ILD were included, 18 were male and 45 had diffuse cutaneous involvement. Mean disease duration was 2.3 years. Rate of FVC% predicted change over time ranged from -0.23 to 0.38, indicating a highly variable course. Baseline KL-6 levels were higher in patients than healthy controls (p<0.0001). Baseline higher KL-6 levels were predictive of faster FVC% decline at the one year follow-up (b=-0.03, *p*=0.04). Upon categorizing KL-6 using a previously determined optimal cut-off of 1273 u/mL (1), its predictive significance remained in the univariate (p=0.01) and multivariable analyses adjusted for Scl-70, disease type and gender (b=-0.03, *p*=0.04). Twenty nine (34.5%) patients had KL6 levels equal or above 1273 u/mL. Although CCL-18 was higher in patients than controls (<0.0001), its levels did not predict rate of FVC decline (provide p-value)

Figure 1. KL-6 levels between patients and controls and %FVC change



a) KL-6 levels (u/mL) in SSc patients versus controls (red line= 1273 cut-off). b) %FVC annualized change in KL-6 positive versus negative patients based on a cut-off of 1273 u/mL. Note: Lower values in %FVC change correspond to a faster FVC decline.

Conclusions: KL-6 but not CCl-18 is predictive of early SSc-ILD progression. In this study, we also validated the previously proposed cut-off of 1273 for KL-6 in an independent cohort. KL-6 is a promising pneumoprotein that can inform individualized clinical care and contribute to enrichment strategies in clinical trials of SSc-ILD.

References:
[1] Kuwana M, Shirai Y, Takeuchi T. Elevated Serum Krebs von den Lungen-6 in Early Disease Predicts Subsequent Deterioration of Pulmonary Function in Patients with Systemic Sclerosis and Interstitial Lung Disease. J Rheumatol 2016.

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Disclosure of Interest: None declared
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FRI0364 USEFULNESS OF SERUM HAPTOGLOBIN LEVELS AS A NOVEL MARKER FOR PULMONARY ARTERIAL HYPERTENSION COMPLICATED WITH CONNECTIVE TISSUE DISEASE

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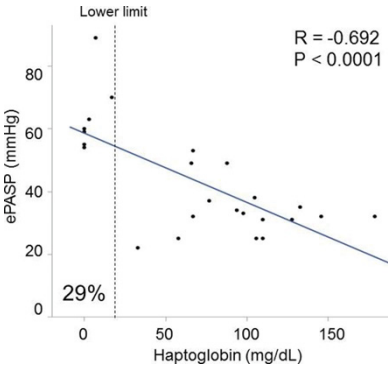
Background: Pulmonary arterial hypertension (PAH) is of great clinical significance as a life-threatening complication of connective tissue diseases (CTD). Pulmonary artery thrombotic microangiopathy (PATM) is an important pathophysiology of PAH. The concept of PATM refers to localized thrombotic microangiopathy to be defined histologically and should be discriminated from systemic thrombotic microangiopathy characterized by microangiopathic haemolysis and thrombocytopenia. The degree of PATM has been suggested to be associated with vasodilator response, severity, and prognosis of PAH, and anticoagulation therapy might be effective in PAH patients with features of PATM [1]. Haptoglobin (Hp) is a plasma protein mainly produced by hepatocytes, which binds free haemoglobin released from erythrocytes and protects the kidneys from damage induced by haemoglobin. The Hp is measured in clinical setting as a sensitive marker to detect intravascular haemolysis including thrombotic microangiopathy [2].
Objectives: We hypothesized that serum Hp levels decreased in patients with PAH due to pulmonary microangiopathic haemolysis. The aim of this study was to investigate the association between serum Hp levels and pulmonary artery systolic pressure estimated by echocardiography (ePASP) in patients with CTD.
Methods: This study included CTD patients with suspicion of PAH who were attending Rheumatology Department in Hokkaido University Hospital between August 2015 and August 2016 and underwent echocardiography. PAH was diagnosed based on right heart catheter findings. Serum Hp levels were measured by standardised turbidimetric immunoassay in all patients. Demographic data, laboratory results, and echocardiographic findings were extracted from the medical records. Decreased serum Hp levels were defined as below 19 mg/dL based on the 95th-percentile of healthy controls.
Results: Twenty-four CTD patients with confirmed PAH (CTD-PAH) and 32 CTD patients without PAH (non-PAH) were enrolled. Decreased serum Hp levels were significantly frequent in patients with CTD-PAH compared with non-PAH patients (29% vs 6%, p=0.03). In patients with CTD-PAH, serum Hp levels had a significant negative correlation (r = -0.692, p<0.0001, Figure 1) with ePASP, and serum lactate dehydrogenase (LDH) levels were significantly elevated in patients with decreased Hp levels (233±47 U/L vs 187±42 U/L, p=0.01). Follow up study showed lowering ePASP led to normalizing serum Hp levels.



Conclusions: Serum Hp levels correlated negatively with ePASP in patients with CTD-PAH, and serum LDH levels were higher in CTD-PAH patients with decreased Hp levels. These findings suggest that decreased Hp levels in CTD-PAH patients may reflect PATM and subsequent subclinical haemolysis. Serum Hp levels are a candidate of additional non-invasive marker of CTD-PAH to assess the degree of PATM.

References:
[1] Johnson SR, et al. Thrombotic arteriopathy and anticoagulation in pulmonary hypertension. Chest. 2006.
[2] Barcellini W, et al. Clinical Applications of Hemolytic Markers in the Differential Diagnosis and Management of Hemolytic Anemia. Dis Markers. 2015.
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FRI0365 NEW COLLAGEN BIOMARKERS PREDICT PROGRESSION OF FIBROSIS IN SYSTEMIC SCLEROSIS

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Background: Systemic sclerosis (SSc) is a complex autoimmune disease with extensive fibrosis of the skin and internal organs in which extracellular matrix (ECM) remodeling is a key pathogenic process. Imbalance in the formation and degradation of collagens results in fibrosis. Quantifying the tissue turnover in a highly fibrotic disease such as SSc is very important for the prediction of disease progression and therapeutic efficacy. Given the clinical heterogeneity of SSc patients, biomarkers facilitating personalized medicine approaches are highly needed.
Objectives: To evaluate the potential of selected ECM neo-epitopes as serological biomarkers for diagnosis, prediction of clinical outcomes and disease progression in SSc.
Methods: Healthy controls (HC; n=29), stable SSc (n=149), and progressive SSc patients (n=23, progression defined either as 10% decrease in FVC% predicted or increase in mRSS ≥25% and 5 points on one year clinical follow up), meeting the 2013 ACR/EULAR classification criteria were analyzed. Longitudinal clinical assessment, data recording and sera collection were done according to EUSTAR standards. ECM-degradation (C3M, VICM, C4M2, BGM) and ECM-formation biomarkers (P1NP, P4NP7S, Pro-C3, Pro-C5, Pro-C6) were measured in serum using newly developed ELISA-based assays (Nordic Bioscience). Differences in biomarker levels were analyzed with respect to several fibrosis-related clinical outcomes. Statistical analysis was performed by Man-Whitney U, Kruskal-Wallis and Spearman tests. Biomarkers' sensitivity and specificity was examined by ROC analysis.
Results: Both ECM-degradation and ECM-formation biomarkers differed between SSc patients and HC: The expression of C4M2, Pro-C3, BGM and C3M was significantly increased in SSc patients compared to HC ($p<0.0001$, AUC=0.93; $p<0.0001$, AUC=0.74; $p=0.003$, AUC=0.67; $p<0.0001$; AUC=0.94, respectively), whereas P1NP was significantly lower ($p<0.0001$, AUC=0.78), Figure 1. Furthermore, Pro-C3, VICM and Pro-C6 levels were significantly higher in SSc progressors vs. HC ($p<0.0001$, AUC=0.86; $p=0.003$, AUC=0.75; $p=0.0005$, AUC=0.81, respectively).
Currently, there is no fully validated biomarker predicting worsening of fibrosis. In this regard, most interestingly, the ECM-degradation markers C4M2, BGM, C3M were significantly lower in SSc patients showing progression of fibrosis on follow up versus SSc patients being stable on follow up ($p<0.0001$; $p<0.008$; $p<0.0001$, respectively). Consistently, the formation marker Pro-C6 was significantly increased ($p=0.001$, AUC=0.71) indicating a profound imbalance of ECM turnover in progressors. The strongest difference between progressive and stable SSc patients was seen for the ratio between the formation and degradation biomarkers Pro-C3/C3M which showed an AUC of 0.86 in progressive vs. stable SSc patients ($p<0.0001$).
Conclusions: These data support ECM neo-epitopes as potential new biomarkers of prognostic interest in SSc. This could help identifying patients at risk of progression of their fibrotic disease at one year follow up. The significant decrease in ECM-degradation markers in SSc progressors compared to stable patients suggests an impairment of collagen degradation in this group. The ratio of Pro-C3/C3M is as a new potential predictive index for differentiation of stable vs. progressive patients.
Disclosure of Interest: R. Dobrota: None declared, S. Jordan: None declared, P. Juhl Employee of: Nordic Bioscience, B. Maurer: None declared, L. Wildi: None declared, A.-C. Bay-Jensen Employee of: Nordic Bioscience, M. A. Karsdal Shareholder of: Nordic Bioscience, Employee of: Nordic Bioscience, A. S. Siebuhr Employee of: Nordic Bioscience, O. Distler Grant/research support from: Actelion, Bayer, Boehringer Ingelheim, Pfizer, Sanofi, Consultant for: 4D Science, Actelion, Active Biotec, Bayer, BiogenIdec, BMS, Boehringer Ingelheim, ChemomAb, EpiPharm, espeRare foundation, Genentech/Roche,GSK, Intentiva, Lilly,medac, Mepha, MedImmune, Mitschubishi Tanabe Pharma, Pharmacyclics, Pfizer, Sanofi, Serodapharm, Sinoxa, Speakers bureau: AbbVie, iQone Healthcare, Mepha
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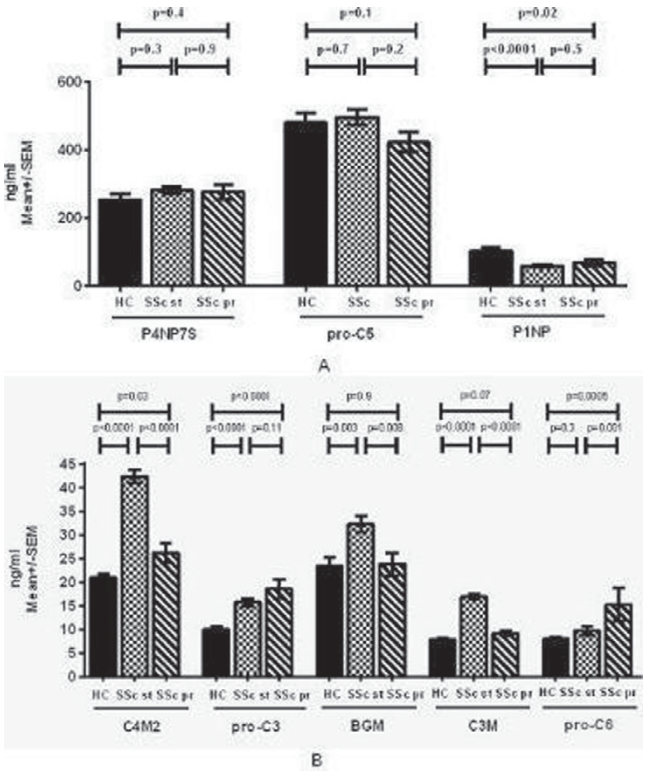

Figure 1: Expression of biomarkers in healthy controls (HC), stable SSc patients (SSc) and progressive SSc patients (SSc pr). A) P4NP7S- amino terminal pro-peptide of procollagen type IV; Pro-C5- formation of collagen type V; P1NP-amino terminal pro-peptide of procollagen type I; B) C4M2-MMP degraded collagen type IV ; Pro-C3- formation of collagen type III ; BGM -biglycan degraded by MMP-2/9; C3M-collagen III degraded by MMP-9; Pro-C6- formation of collagen type VI;

FRI0366 A PILOT STUDY ON ISCHEMIA AND REPERFUSION INJURY DURING A RAYNAUD'S ATTACK: SEQUENTIAL ASSESSMENT OF REDOX STRESS PARAMETERS IN A UNIQUE COOLING AND REWARMING EXPERIMENT

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Background: Oxidative stress plays a role in systemic sclerosis (SSc), but the molecular mechanisms involved are incompletely understood. During an attack of Raynaud's phenomenon (RP) a period of ischemia (I), followed by reperfusion (R) occurs frequently, associated with the severity of vasculopathy. [1] Only in secondary RP digital ulcers develop. We hypothesized that I/R injury may play a role in the pathogenesis and could offer new therapeutic targets.
Objectives: To explore the course of oxidative stress in patients with SSc compared with primary RP and healthy controls.
Methods: A total of 30 patients were included: 10 with limited cutaneous SSc (age: 57 (53–61) yr, male/female 5/5), 10 with primary RP (age: 54 (41–58), 2/8), and 10 healthy controls (age: 25 (22–25), 3/7). A standardized cooling experiment was performed and digital perfusion was assessed in all 5 fingers using photo-electric plethysmography: at baseline (T=0) the dominant hand was submerged in water at 33℃, followed by cooling in steps of 3℃ every 4 minutes, until 6℃ or when pain became intolerable (T=1). Recording was continued 10 (T=2) and 30 (T=3) minutes of rewarming to ambient temperature (23℃). Blood was drawn from ipsilateral cubital vein at T0, 1, 2 and 3, markers for tissue injury (lactate, LDH, creatinine phosphokinase (CPK) [routine methods]), redox status (free thiols corrected for total protein) and nitric oxide (NO) activity (NO_2^-, NO_3^-, RXNO) were measured in plasma. [1–3] Numbers are in median (IQR).
Results: Baseline free thiols were significantly decreased in RP vs. controls (5.18 (4.79–5.63) vs 5.87 (5.41–5.99) umol/g, p=0.013), with no differences in lactate, LDH, CPK, and NO activity. Raynaud's attack was induced in all RP patients but not in controls. Median duration of hypoperfusion was greater in SSc vs. PRP (30 (27–35) vs. 12 (9–14) min, p=0.010), with a considerably longer recovery time (8 (4–10) vs. 0 (0–1) min, p=0.006). No changes were observed in lactate, LDH, and CPK levels. A rise in free thiols occurred at recovery (T4) in all 3 groups (figure 1). The concentrations of NO-related products did not change during cooling or recovery. No association was detected between the extent of I/R and plasma parameters.
Conclusions: In patients with RP free thiols were significantly reduced, indicating increased redox stress. During the cooling and rewarming experiment, a clear rise in free thiols was observed during rewarming, irrespective of the underlying disease or finger perfusion. Meanwhile, NO-related products remained stable.