effect of PF1801 on the expression of antioxidant molecules in the myotubes was analyzed with quantitative real-time PCR.

**Results:** GLP-1R was expressed on the expanded muscle fibers of PM and CIM. The treatment with PF1801 in monotherapy or in combination with PSL suppressed CIM-induced muscle weakness and the muscle weight loss as well as the severity of histological myotubes while the monotherapy with PSL did not suppress muscle weakness and muscle weight loss. PF1801 decreased the levels of inflammatory mediators such as HMGB1, TNF-α, and IL-6 in the serum of CIM. In vitro, PF1801 inhibited FASLG-induced myobute necrosis and decreased the levels of HMGB1, TNF-α, and IL-6 in the supernatant. PF1801 activated AMPK and decreased the levels of PGAM5, which was crucial for FASLG-induced necrosis of the myotubes. The inhibitory effect of PF1801 on myobute necrosis was cancelled by compound C, an AMPK-kinase inhibitor, or MG132, a proteasome inhibitor, suggesting that PF1801 promoted ubiquitin-proteasome-mediated PGAM5 degradation through the activation of AMPK. Furthermore, PF1801 suppressed FASLG-induced reactive oxygen species (ROS) accumulation in myotubes, which was also crucial for the execution of necrosis, thorough up-regulating the antioxidant molecules such as Nrf2/22, Hmx1, Gclm, and Nqo1.

**Conclusion:** GLP-1R agonist could be a novel therapy for PM that restores muscle strength as well as suppresses muscle inflammation through inhibiting muscle fiber necrosis.

**REFERENCES:**

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POS0473

**INFLAMMATORY-PRIMED MUSCLE CELLS INFLUENCE MACROPHAGE CYTOKINE SECRETION IN A MYOSITIS CONTEXT**

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**Background:** Sporadic inclusion body myositis (sIBM) is a progressive disease causing muscle weakness and mobility issues. It is characterised by inflammatory and degenerative features; however, there is no clear single cause for sIBM symptoms. Inflammatory factors include macrophage infiltration into muscle fibres and presence of high levels of inflammatory cytokines such as IFNγ [1]. Degenerative features within muscle fibres include sarcoplasmic inclusions containing ubiquitin, p62 and TAR DNA-binding protein 43 (TDP-43) are two proteins that are found in inclusion bodies, and these have been suggested as sensitive diagnostic markers differentiating sIBM from other inflammatory myopathies [2]. Currently there is limited understanding of the pathogenic mechanisms underlying sIBM, and no effective treatments.

**Objectives:** The aim of this study is to investigate the interplay between inflammatory and degenerative features of sIBM with a focus on macrophage secreted factors, p62 and TDP-43 sarcoplasmic aggregation. Firstly, to examine whether skeletal muscle cells that have previously been exposed to an inflammatory environment can in turn influence the inflammatory response of macrophages. Secondly, by investigating if macrophage-secreted inflammatory factors influence p62 and TDP-43 sarcoplasmic aggregates. This will give insight into whether macrophage inflammation precludes skeletal muscle degeneration in an sIBM context or vice versa.

**Methods:** Primary human myogenic cells from healthy individuals were differentiated into myotubes for 7 days, with further 48-hour 20 ng/mL IL-1β and 750 ng/mL IFNγ incubation to form inflammatory-primed myotubes.Conditioned media (CM) was collected from washed myotubes after 24 hours. 6×108 THP-1 cells/mL were differentiated to unprimed (M/PM) macrophages with 150 nM phorbol 12-myristate 13-acetate (PMA) for 24 hours, followed by 72-hour PMA-free rest. Immunofluorescent-stained M/IFN-γ-LPS THP-1 were generated from M/PM with 48-hour IFNγ and lipopolysaccharide (LPS) incubation. 20 % CM from myotubes was added to M/PM or M(IFN-LPS) THP-1 for 24 hours, before washing and measuring IL-6 and TNFα release from macrophages via ELISA. 20 % CM from M/PM or M(IFN-LPS) was added to myotubes for 48 hours before quantifying TDP-43 and p62 aggregates with image analysis.

**Results:** M(IFN-LPS)-THP-1 secreted higher levels of IL-6 and TNFα than M(ISPMA), M/PM or M(IFN-LPS) exposed to conditioned media from control or inflammatory-primed myobutes had no detectable TNFα secretion. For both M/PM and M(IFN-LPS), addition of inflammatory-primed-myobute CM caused higher IL-6 secretion than adding control myobute CM. Incubating unprimed myotubes with M/PM or M(IFN-LPS) CM had no effect on the presence, size, or frequency of p62 or TDP-43 sarcoplasmic aggregates compared to control conditions.

**Conclusion:** Inflammatory priming myotubes with IL-1β and IFNγ caused a response in macrophages that increased IL-6 production. However, CM from inflammatory-primed myobutes did not influence TNFα secretion from macrophages compared to control myobute CM. IL-6 has pleiotropic effects on skeletal muscle [3], promoting both hypertrophy and atrophy under different conditions. Therefore the effects of macrophage IL-6 upregulation on muscle is unclear. Culturing myotubes in unprimed or inflammatory-primed macrophage conditioned media did not influence p62 or TDP-43 sarcoplasmic aggregation. This suggests macrophage secretory factors are not responsible for the sIBM degenerative features of p62 and TDP-43 aggregation.

**REFERENCES:**

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POS0474

**ACTIVATION AND HYPERSENSITIVIZATION OF THE ANGIOTENSIN II TYPE 1 AND ENDOTHELIN-1 TYPE A RECEPTORS BY AGONISTIC AUTOANTIBODIES CONTRIBUTES TO VASCULAR INJURY IN SCLERODERMA RENAL CRISIS**

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**Background:** Scleroderma renal crisis (SRC) is a vascular complication of systemic sclerosis (SSc) with substantial risks for end-stage renal disease and death. Activating autoantibodies (Abs) targeting the angiotensin II type 1 (AT1R) and the endothelin-1 type A receptor (ETAR) are suggested to contribute to the vasculopathy in SSc (1, 2).

**Objectives:** Here, we sought to determine their pathogenic significance for acute renal vascular injury.

**Methods:** IgG proteins with Abs to SRC were studied for AT1R and ETAR dependent biologic effects on isolated rat renal interlobar arteries and vessels including contraction, signaling, and mechanisms of receptor activation. A cohort of ten patients with refractory SRC received multimodal treatment including AT1R and ETAR inhibition and plasma exchange and was followed for improvement of kidney function.

**Results:** In myography experiments, patient IgG exerted vasoconstriction (mean 6.5% of KCl induced contraction [95% confidence interval (95 CI) 5.0-8.1]) whereas control IgG did not (0.6% [95 CI 0.3-1.0]). The response was sensitive to inhibition of AT1R (3.0% [95 CI 1.4-4.7]) and ETAR (1.0% [95 CI 0.6-1.3]) and relied on MEK-ERK signaling. Contraction induced by angiotensin II and endothelin-1 was amplified by anti-AT1R and anti-ETAR Abs with substantial cross-talk between both receptors implicating autocrine receptor hypersensitization. Co-immunoprecipitation experiments indicated heterodimerization between both receptor types enabling functional interrelation by structural inter- actions. 30% of patients with refractory SRC had improved kidney function after multimodal therapy.
Conclusion: We provide experimental and clinical evidence that agonistic Abs may contribute to SRC. Novel therapies targeted at autoimmune hyperactivation of AT1R and ETAR might improve outcomes in severe cases of SRC.

REFERENCES:

Disclosure of Interests: None declared.

Methods: Data from 277 SSc patients, fulfilling the 2013 ACR/EULAR classification criteria, attending our Scleroderma Clinic were retrospectively analysed. We selected patients with EKG trace and a blood sample available, collected after the SSc diagnosis. The sera levels of sST2 (ELISA Kit, Abcam), IL-33 (ELISA kit, RayBiotech) and NT-pro-BNP (ELISA Kit, Abcam) were measured. Patients with a history of heart diseases occurring before the diagnosis of SSc or features of secondary cardiac involvement (pulmonary arterial hypertension, severe interstitial lung disease or renal disease) were excluded.

RESULTS: Forty-six SSc patients showed significant EKG abnormalities (rhythm and conduction disorders). Thirty-one SSc patients without pathologic finding at EKG traces were recruited as the control group. From the analysis of the clinical characteristic of the disease at the moment of serum collection, patients with EKG abnormalities have more frequently both a diffuse form of disease (n=12; 23-50 vs 7-23; p=0.01), with a mean value of mRSS higher than controls (11±9 vs 6±6; p=0.01), and a scleroderma “late” pattern at the nailfold capillaroscopy (n=12; 23-50 vs 6-19; p=0.027). Significantly higher median values of serum levels of sST2 in SSc patients with EKG disorders compared to the control group (4289 pg/mL vs 1455 pg/mL, p=0.0002) were detected, while opposite results were found analyzing serum levels of IL-33 (2.89 pg/mL vs 4289 pg/mL, IQR 2383 vs 2560 pg/mL, IQR 1455 vs 0.0002) were detected. These values correlated with sST2 serum levels (rho Spearman correlation 0.37; p=0.0006).

Conclusion: SSc patients with EKG abnormalities showed an increased skin and vascular involvement with respect to the control group. These associations could help clinicians in clinically stratifying SSc patients at risk of EKG abnormalities. To our knowledge, this is the first report evaluating the serum concentration of sST2 in SSc patients. Based on these results, we can speculate on the role of this molecule as potential biomarkers of early cardiac injury during SSc, although further studies involving a larger cohort of patients are needed.

REFERENCES:

Figure 1: Graph 1. Serum sST2 (a) and IL-33 (b) concentrations in patients with EKG abnormalities (EKG+) vs control group (EKG-).

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POS0475
ROLE OF IL-33/ST2 AXIS IN SYSTEMIC SCLEROSIS PATIENTS WITH ELECTROCARDIOGRAPHIC ABNORMALITIES

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Background: Electrocardiographic (EKG) abnormalities are described in 25-75% Systemic Sclerosis (SSc) patients and they are associated with other systemic manifestations as well as with a worse prognosis. There is an increasing need for clinical and laboratory biomarkers to ameliorate the diagnostic approaches to patients with EKG abnormalities. In the last decade, many studies focused on the components of the interleukin (IL)-33/ST2 axis. Under physiological conditions, IL-33 is released by apoptotic cardiac cells, promoting a protective mechanism of cell survival, thanks to the binding with its transmembrane receptor ST2. During pathological cardiovascular events, an abnormal secretion of the IL-33 soluble receptor (sST2) by TH2 cells occurs. It binds IL-33 not allowing the physiological mechanism driven by the IL-33/ST2 binding previously described. For these reasons, sST2 has been proposed as a biomarker of cardiac injury in a variety of diseases.

Objectives: Aim of this study was to analyse clinical and demographical parameters in a group of SSc patients, trying to define any possible feature associated with EKG abnormalities. Furthermore, the role of IL-33/ST2 axis components as biomarkers of cardiac injury in patients with SSc-related EKG abnormalities was evaluated, also assessing the possible correlation with serum concentration of NT-pro-BNP, a well-known cardiac injury biomarker in SSc.

Methods: Data from 277 SSc patients, fulfilling the 2013 ACR/EULAR classification criteria, attending our Scleroderma Clinic were retrospectively analysed. We selected patients with EKG trace and a blood sample available, collected after the SSc diagnosis. The sera levels of sST2 (ELISA Kit, Abcam), IL-33 (ELISA kit, RayBiotech) and NT-pro-BNP (ELISA Kit, Abcam) were measured. Patients with a history of heart diseases occurring before the diagnosis of SSc or features of secondary cardiac involvement (pulmonary arterial hypertension, severe interstitial lung disease or renal disease) were excluded.

Results: Forty-six SSc patients showed significant EKG abnormalities (rhythm and conduction disorders). Thirty-one SSc patients without pathologic finding at EKG traces were recruited as the control group. From the analysis of the clinical characteristic of the disease at the moment of serum collection, patients with EKG abnormalities have more frequently both a diffuse form of disease (n=12; 23-50 vs 7-23; p=0.01), with a mean value of mRSS higher than controls (11±9 vs 6±6; p=0.01), and a scleroderma “late” pattern at the nailfold capillaroscopy (n=12; 23-50 vs 6-19; p=0.027). Significantly higher median values of serum levels of sST2 in SSc patients with EKG disorders compared to the control group (4289 pg/mL vs 1455 pg/mL, p=0.0002) were detected, while opposite results were found analyzing serum levels of IL-33 (2.89 pg/mL vs 4289 pg/mL, IQR 2383 vs 2560 pg/mL, IQR 1455 vs 0.0002) were detected. These values correlated with sST2 serum levels (rho Spearman correlation 0.37; p=0.0006).

Conclusion: SSc patients with EKG abnormalities showed an increased skin and vascular involvement with respect to the control group. These associations could help clinicians in clinically stratifying SSc patients at risk of EKG abnormalities. To our knowledge, this is the first report evaluating the serum concentration of sST2 in SSc patients. Based on these results, we can speculate on the role of this molecule as potential biomarkers of early cardiac injury during SSc, although further studies involving a larger cohort of patients are needed.

REFERENCES:

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POS0476
THE NEUTRAL RECEPTOR TR4 ORCHESTRATES CYTOSKELETAL ORGANIZATION IN A Gα12/13 ROCK-DEPENDENT MANNER TO PROMOTE MYOFIBROBLAST DIFFERENTIATION AND TISSUE FIBROSIS IN SYSTEMIC SCLEROSIS

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