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early (within 2 years from onset) diffuse cutaneous SSCs (dcSSC), 14 late dcSSC and 20 limited cutaneous SSCs (lcSSC). Levels of MCP-1 in serum were measured by commercial ELISA. Expression of MCP-1 and its major receptor CCR2 in snap-frozen skin biopsies was determined by immunohistochemistry. Co-localisation studies were performed to identify the cell types expressing MCP-1. Expression of chemokine receptors (CCR2, CXCXR2 and CCR5) on 9 SSCs and 9 normal fibroblasts cell lines was determined by flow cytometry. Results: MCP-1 serum levels (mean±sem) were significantly elevated in SSC patients (380±16 pg/ml) compared to controls (152.2±26 pg/ml, p=0.002) and were highest in the early dcSSC subset (557.1±61 pg/ml) compared to late-stage disease (307.4±41 pg/ml, p=0.01) or lcSSC (276.2±28 pg/ml, p=0.007). Immunohistochemistry confirmed strong expression of MCP-1 and CCR2 in skin from patients with early-stage dcSSC, but expression in late-stage dcSSC and lcSSC was minimal. In particular MCP-1 co-localised with endothelial cells, myofibroblasts, macrophages and T lymphocytes. Furthermore only SSC fibroblasts cell lines from early stage disease showed expression of CCR2 (5 of 9 lines) and CXCXR2 (6 of 9). No fibroblast lines expressed CCR5. Conclusions: Colocalisation of MCP-1 ligand with surface markers of cell type known to be activated in SSC may mark this region as a biomarker. This study demonstrates strong, replicated association of MCP-1 VNTR alleles with disease. We sought to determine whether polymorphisms within the IL1 gene cluster are associated with AS. Methods: We studied 29 polymorphic markers across the cluster in 305 parent-case trios (PCT), 223 affected sibling pair families and 199 ethnically matched unrelated healthy controls. Genotyping was performed using Sequenom MassArray or PCR/RFLP. Association was assessed by the transmission-disequilibrium test (TDT) using the program "Transmit" employing the robust variance option to control for the effect of linkage. Conditional extended TDT (CETDT), a form of logistic regression applied to TDT, was used to investigate the parent-case trio families to determine whether one or more SNP is the primary associated variant(s). Results: Comparing family probands with healthy controls, association was demonstrated with nine SNPs within IL1A, IL1B, IL1F6, IL1F8 and IL1F5 and the region known to be linked with AS, indicating that this region contains one or more genes involved in such patients. An unrestricted educational grant from Genzyme Corporation is gratefully acknowledged. Results: Most patients were female (84%) mean (±sd) age at disease onset was 44±13 years, mean follow-up duration 96±55 months. Mean baseline skin score skin score was 30±11, median of 30 (range 12 to 96) this decreased annually to a mean of 17±11, median of 17 at 5 years duration. Skin sclerosis was classified as severe in 33% of cases, based upon a peak mRSS ≥ 35, at a median of 12 months, whilst overall the median time taken to reach the peak skin score was 17 months. The first major organ involvement developed at a median of 13 months (1 to 130), Pulmonary involvement developed in 53% of cases, cardiac disease in 8%, scleroderma renal crisis in 11% and 5% developed myositis. Of the 42 patients who had died 17% occurred within 3 yrs of disease onset, 57% after 5 yrs. Patients with severe skin involvement had a higher cumulative frequency (72%) of developing another organ-based endpoint in comparison to those with less severe skin disease (66%) and were more likely to reach ≥2 additional major end-points (35%) compared to patients with mRSS < 35 (20% to 0.03 by fishers exact test). Although there was no correlation between having a skin score ≥ 35 and the development of individual organ-based complications there was a higher frequency of mortality in those patients (p = 0.03). Conclusions: Our study of a cohort of early-stage dCSS patients confirms that severe skin disease associates with mortality and a higher accumulation of organ-based complications, but suggests that peak skin score is often attained by 18 months from disease onset. The overall trend of stability or improvement in skin sclerosis within 2 years of diagnosis may confound interventional studies hoping to demonstrate prevention of skin score worsening in such patients. An unrestricted educational grant from Genzyme Corporation is gratefully acknowledged.

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OP61. UPRREGULATION OF THE MONOCYTE CHEMOTACTANT PROTEIN-1 LIGAND-RECEPTOR AXIS IN EARLY STAGE DIFFUSE CUTANEOUS SCLERODERMA

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Background: Fibroblast derived monocyte chemotactant protein-1 (MCP-1 or CCL2) is a candidate mediator linking inflammatory and fibrotic processes in scleroderma (SSc). The present study addresses the hypothesis that MCP-1 is a key mediator of intercellular cross-talk in the pathogenesis of early stage SSc and that fibroblasts are a target for activation for this cytokine. Methods: Serum samples and skin biopsies were examined from 64 patients with SSc and 12 healthy controls. Of our cohort of patients 30 had
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**OP63. ALTERED LIPID RAFT EXPRESSION AND ASSOCIATED SIGNALING IN T LYMPHOCYTES FROM PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS (SLE)**

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**Background:** We have shown previously that LCK is reduced in T cell lipid rafts from patients with SLE and that this reduction is associated with disease activity and a parallel increase in LCK ubiquitination independent of T cell activation. Our results indicate that lipid raft expression is increased in T cells from patients with SLE and LCK may be differentially regulated owing to alteration in the association of CD45 with lipid raft domains. CD45 tyrosine phosphatase, which regulates LCK activity, is also differentially expressed and its localisation into lipid rafts increased in T cells from patients with SLE. Therefore the reasons underlying these alterations in CD45 and LCK association in T cells from patients with SLE were examined.

**Methods:** T cells from 19 patients with SLE, 15 healthy controls and 6 patients with RA were isolated from PBMC. T cells, either ex vivo, rested overnight or activated using beads coated with antibodies to CD3 and CD28 for 1, 5, 10, 30 and 60 minutes were then examined for expression of lipid rafts and the partition and expression of LCK and CD45 in raft domains using confocal microscopy. Flow cytometric analysis of intracellular marker expression was also conducted using a BLAST analysis system.

**Results:** The results revealed that ex vivo changes in LCK, CD45 and lipid raft expression in SLE T cells were reversed by overnight rest. The results also revealed that alterations in the level of lipid raft expression and occupancy were not induced by serum factors from patients with SLE. However, T cell receptor (TCR) activation appeared to influence CD45/Lipid raft localisation in lupus patients. T cells from patients with SLE reacted more rapidly in terms of lipid raft and CD45 recruitment to the site of TCR activation and after its initial recruitment CD45 was excluded more rapidly from the activation site.

**Conclusions:** These results indicate that cell contact, activating aberrant proximal signalling pathways, may be important in influencing abnormalities in T cell signalling, and therefore function, in patients with SLE. These results show that T cells from patients with SLE are ’primed’ for activation and respond more rapidly to antigenic triggers than T cells from normal controls.

**OP64. SEROLOGICAL INVESTIGATION OF AUTOANTIBODIES IN 577 FAMILIES WITH SYSTEMIC LUPUS ERYTHEMATOSUS**

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**Background:** The BXS B mouse strain is an excellent model of the proliferative glomerulonephritis seen in human SLE. We previously carried out linkage analyses between BXS/B and C57BL/10 mice to identify genetic intervals containing susceptibility loci. We have shown that four chromosomes contain disease-susceptibility loci and that four loci were associated with different aspects of the disease phenotype. Bxs3 mapped to an interval that had also been mapped in the NZB/NZW F1 model of lupus (Sle1, Nba2), however, Bxs1, 2 and 4 represented novel disease-associated intervals. Methods: In order to dissect the individual effects of the four non-MHC, autosomal loci (Bxs1-4) that contribute to SLE susceptibility in BXS/B mice, we generated congenic lines on a C57BL/10, B10.Yaa background, for Bxs1(46.3-89.2Mb), Bxs2/3 (20.0-65.9Mb), Bxs1/4 (64.4-159.0Mb) and Bxs2/3 (105.4-189.0Mb). Results: Glomerulonephritis, qualitatively similar to that seen in the parental BXS/B strain, developed in three of these congenic strains. Early-onset, severe disease was observed in B10.Yaa.BXS/B-Bxs1/4 congenic mice and caused 50% mortality by 12 months. In B10.Yaa.BXS/B-Bxs1/4 mice disease progressed more slowly, resulting in 13% mortality at 12 months. The progression of renal disease in both of these strains was correlated with the level of anti-dsDNA Abs. B10.Yaa.BXS/B-Bxs1 mice, despite their genetic similarity to B10.Yaa.BXS/B-Bxs1/4 mice, developed a low-grade glomerulonephritis in the absence of anti-dsDNA Abs, whereas B10.Yaa.BXS/B-Bxs1/2 mice did not develop nephritis in spite of the presence of anti-dsDNA Abs. Furthermore, we have shown that Yaa can cause breakdown of tolerance in a non-autoimmune prone strain, permitting development of ANA and anti-chromatin Abs, without provoking nephritis.

**Phenotypic features of BXS chromosome 1 congenic mice**

<table>
<thead>
<tr>
<th>Mouse strain</th>
<th>Mortality</th>
<th>Anti-chromatin Abs</th>
<th>Anti-nuclear Abs</th>
<th>Anti-dsDNA Abs</th>
<th>Anti-ssDNA Abs</th>
<th>Nephritis</th>
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All data for mice aged 12 months except BXS/B (6 months)

**Conclusions:** Bxs1 directed an increase in titre and spectrum of autoAbs, whilst Bxs1/4, promoted the development of nephritis. Bxs2/3 was linked to the production of anti-dsDNA Abs without concomitant glomerulonephritis. In contrast, the Bxs3 interval was sufficient to generate classic lupus nephritis in a non-autoimmune prone strain, although disease was delayed in comparison to the BXS/B strain, emphasising the necessity for multiple interactions in the production of the full BXS/B phenotype.
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**OP66. RECONSTITUTION OF C1q-DEFICIENCY WITH BONE MARROW DERIVED CELLS AMELIORATES THE AUTOIMMUNE ASSOCIATED WITH C1q-DEFICIENCY**

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**Background:** Previous studies have shown that bone marrow cells (BMC) can reconstitute C1q serum levels in C1q-deficient mice (C1qa-/-) suggesting that transplantation of BMC might be a potential treatment for patients with C1q-deficiency. C1q-deficiency is strongly associated with the development of SLE. C1qa-/- mice backcrossed onto a lupus-prone MRL/Mp background showed an acceleration of the onset and the severity of the disease. We investigated whether the reconstitution of C1q by BMC is sufficient to ameliorate the development of autoimmunity.

**Methods:** Recipient MRL/Mp female mice (C1qa-/- or C1qa+/+) were irradiated (800 rads) and received a single graft of 10⁶ donor cells from MRL/Mp or C3H/HeJ or C3H/HeN-129. B cell depletion was monitored by anti-CD20 antibodies.

**Results:** Serum C1q levels increased rapidly when the C1qa-/- mice received BMC from C1qa+/+ animals, while the levels decreased more slowly in C1qa+/- mice transplanted with C1qa-/- BMC. As expected, the C1qa-/-/transplanted with C1qa+/+ BMC showed a marked acceleration of the autoimmune disease associated with an increased mortality compared with systemic wild-type controls (64% vs. 10%, p<0.0001). Notably, in the C1qa-/-/animals reconstituted with C1qa+/+ BMC a reduction in the levels of autoantibodies were observed when compared with C1qa-/- mice transplanted with C1qa-/- BMC (ANA 1/1200 (0-1/5120) vs. 1/640 (0-1/10240) (median, range), p<0.02; gp70C 0.7 (0.1-6.3) vs. 3 (0.5-7.9), p<0.06, respectively). In addition, a significant delay in the development of proteinuria and expression of glomerulonephritis was also observed [1 (1-4) vs. 2 (1-4), p=0.04 for proteinuria and 1 (1-3) vs. 2 (1-4), p=0.04 for glomerulonephritis, respectively]. By contrast, disease was accelerated in the C1qa+/- animals reconstituted with C1qa+/+ BMC compared with the animals transplanted with C1qa+/- cells (1/5120 (1/80-1/10240) vs. 1/800 (0-1/5120), p<0.0001 for ANA; 1.26 (0.2-6.3) vs. 0.18 (0.5-3.6), p= 0.002 for gp70C levels; 2 (1-4) vs. 1 (1-3), p=0.002 for proteinuria and 2 (1-4) vs. 1 (1-2), p=0.005 for glomerulonephritis). Impaired clearance of apoptotic cells, previously reported in C1qa-/- mice, was also reversed by BMC.

**Conclusions:** These data suggest that bone marrow transplant may ameliorate the autoimmune disease associated with C1q-deficiency and provide supporting evidence to the stem cell transplantation as a therapy for patients with hereditary C1q deficiency.

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**OP67. SEROLOGICAL CHANGES FOLLOWING B LYMPHOCYTE DEPLETION THERAPY IN SYSTEMIC LUPUS ERYTHEMATOSUS**

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**Background:** Systemic lupus erythematosus (SLE) is an autoimmune disorder in which autoantibodies are thought to cause tissue damage and dysfunction. B lymphocyte depletion therapy (BlyD) may induce long term remission if pathogenic, autoreactive B lymphocyte clones can be removed. An open label phase II trial of BlyD was conducted in SLE, although an RCT differentiating wild-type from C1q-deficient genotype. C1q levels were assessed by ELISA. Proteinuria, serum levels of autoantibodies and gp70 immune complexes (gp70IC) were measured periodically. All mice were sacrificed for total immunoglobulins.

**Methods:** Serum C1q levels increased rapidly when the C1qa-/- mice received BMC from C1qa+/+ animals, while the levels decreased more slowly in C1qa+/- mice transplanted with C1qa-/- BMC. As expected, the C1qa-/- /transplanted with C1qa+/+ BMC showed a marked acceleration of the autoimmune disease associated with an increased mortality compared with systemic wild-type controls (64% vs. 10%, p<0.0001). Notably, in the C1qa-/-/animals reconstituted with C1qa+/+ BMC a reduction in the levels of autoantibodies were observed when compared with C1qa-/- mice transplanted with C1qa-/- BMC (ANA 1/1200 (0-1/5120) vs. 1/640 (0-1/10240) (median, range), p<0.02; gp70C 0.7 (0.1-6.3) vs. 3 (0.5-7.9), p<0.06, respectively). In addition, a significant delay in the development of proteinuria and expression of glomerulonephritis was also observed [1 (1-4) vs. 2 (1-4), p=0.04 for proteinuria and 1 (1-3) vs. 2 (1-4), p=0.04 for glomerulonephritis, respectively]. By contrast, disease was accelerated in the C1qa+/- animals reconstituted with C1qa+/+ BMC compared with the animals transplanted with C1qa+/- cells (1/5120 (1/80-1/10240) vs. 1/800 (0-1/5120), p<0.0001 for ANA; 1.26 (0.2-6.3) vs. 0.18 (0.5-3.6), p= 0.002 for gp70C levels; 2 (1-4) vs. 1 (1-3), p=0.002 for proteinuria and 2 (1-4) vs. 1 (1-2), p=0.005 for glomerulonephritis). Impaired clearance of apoptotic cells, previously reported in C1qa-/- mice, was also reversed by BMT.

**Conclusions:** These data suggest that bone marrow transplant may ameliorate the autoimmune disease associated with C1q-deficiency and provide supporting evidence to the stem cell transplantation as a therapy for patients with hereditary C1q deficiency.

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**OP68. QUALITY OF LIFE, FUNCTIONAL STATUS AND CLINICAL FEATURES AT PRESENTATION IN POLYMYALGIA RHEUMATICA (PMR): RESULTS FROM A MULTI-CENTRE PROSPECTIVE COHORT STUDY**

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**Background:** Little is known about quality of life (QOL) and functional status in patients with PMR and their association with clinical features. Patients with rheumatological conditions report poorer physical but similar mental quality of life compared with patients with many other chronic conditions, though there is wide variation in mental QOL across studies of patients with rheumatoid arthritis (RA) and osteoarthritis (OA).

**Methods:** We recruited 129 patients with newly diagnosed PMR fulfilling modified Jones & Hazleman criteria (minimum duration of disease was excluded) and without clinical features of GCA. QOL and functional status were assessed before starting steroid therapy using the SF-36v2 and Modified Health Assessment Questionnaire (MHAQ), respectively. The SF-36 produces summary scores for physical (PCS) and mental (MCS) components. We compared QOL in PMR patients with norms for 65-74 year-olds in the general population and patients with RA and OA. Peripheral signs/symptoms were defined as hand/wrist pain, joint involvement (swelling or restriction of active or passive movements) or oedema. We used multivariable regression to assess whether QOL and peripheral signs/symptoms were associated with functional status, demographic (sex, age) and clinical (ESR, CRP, EMS) data.

**Results:** Mean age was 70.9 years (range 52-92); 59.7% of patients were female. Mean PCS 31.5 (95% CI 30.0 to 32.9) and MCS 38.9 (95% CI 36.8 to 40.9) scores were substantially lower than population norms (44.7 and 53.2, respectively). PCS scores were similar to those in patients with RA and OA, though MCS scores were at the lower end of the range reported for RA and OA. Mean MHAQ score was 1.20 (95% CI 1.10 to 1.30). Poor physical and mental QOL was associated with lower functional status (both p<0.001), whereas poor mental (not physical) QOL was associated with lower age (p=0.003) and being female (p=0.025). Peripheral symptoms/signs were present in 43 (33.3%) patients, all with normal rheumatoid factor, and were associated with increased CRP (p=0.03) and decreased functional status (p=0.04). Longer EMS was associated with higher CRP (p=0.01) but not ESR (p=0.47).

**Conclusions:** Our findings demonstrate the impact of disease on patients with PMR. Follow-up will enable us to evaluate changes in QOL in response to steroid therapy, examine the relationship between QOL and clinical measures of disease activity and determine if patients with peripheral signs/symptoms represent a separate disease subgroup.