neutrophil elastase (NE), proteinase 3 (PR3), and cathepsin G (CG) and myeloperoxidase (MPO) expression by qRT-PCR. NE+/PR3+/CG+ triple knockout mice and in vitro depletion of neutrophils approaches were performed to determine the role of NSPs and neutrophils in estrogen-mediated inflammatory responses. The splenic neutrophil and NSPs expression in lupus-prone MRL-lpr, B6-lpr and NZB/W F1 and their respective controls were also analysed.

**Results** Although oestrogen reduced total splenocytes number, it markedly increased the splenic neutrophil numbers, NSPs and MPO expression in B6 mice (Figure 1). Splenic neutrophils, NSPs and MPO were also significantly increased in MRL-lpr, B6-lpr and NZB/W F1 mice (Figure 2). Despite of the critical role of NSPs and neutrophils in inflammation, depletion of NSPs in vitro did neither affect oestrogen’s ability to increase in splenic neutrophils nor the induction of inflammatory mediators from ex vivo activated splenocytes, and depletion of splenic neutrophils in vitro had also no obvious effect on NSPs expression (due to the increase of NSPs in cells other than neutrophils) and LPS-induced IFNγ and MCP-1 (Figure 3).

**Conclusions** Overall, we demonstrated a remarkable commonality with regards to the increase of neutrophils and NSPs in the spleens of autoimmune-prone mice and estrogen-treated B6 mice.

**114 N-ACETYL-L-CYSTEINE (NAC) CONTROLS OSTEOCLASTOGENESIS THROUGH REGULATING TH17 DIFFERENTIATION AND RANKL PRODUCTION IN RHEUMATOID ARTHRITIS**

**Background and aims** This study aimed to determine the regulatory role of N-Acetyl-l-cysteine (NAC), an antioxidant, in IL-17-induced osteoclast differentiation in rheumatoid arthritis (RA).

**Methods** After RA synovial fibroblasts were stimulated by IL-17, the expression and production of RANKL was determined by real-time PCR and ELISA. Human peripheral blood monocytes were cultured with M-CSF, IL-17, RANKL, and/or various concentrations of NAC, followed by counting of the cells for tartrate-resistant acid phosphatase activity to determine osteoclast formation. Osteoclastogenesis was also determined after cocultures of IL-17-stimulated RA synovial fibroblasts, Th17 cells and various concentrations of NAC with monocytes. After human peripheral CD4+ T cells were cultured with NAC under Th17 condition, IL-17, IFN-g, IL-4, Foxp3, RANKL and IL-2 expression and production was determined by flow cytometry or ELISA.

**Results** When RA synovial fibroblasts were stimulated by IL-17, IL-17 stimulated the production of RANKL, and NAC reduced the IL-17-induced RANKL production in a dose-dependent manner. NAC decreased IL-17-activated phosphorylation of mTOR, JNK and IκB. When human peripheral blood CD14+ monocytes were cultured with M-CSF and IL-17 or RANKL, osteoclasts were differentiated, and NAC reduced the osteoclastogenesis. After human peripheral CD4+ T cells were co-cultured with IL-17-pre-treated RA synovial fibroblasts or Th17 cells, NAC reduced their osteoclastogenesis. Under Th17 polarising condition, NAC decreased Th17 cell differentiation and IL-17 and RANKL production.

**Conclusions** NAC inhibits the IL-17-induced RANKL production in RA synovial fibroblasts and IL-17-induced osteoclast differentiation. NAC also reduced Th17 polarisation. NAC could be a supplementary therapeutic option for inflammatory and bony destructive processes in RA.
SLE samples have lower PMPs than control samples. (A) PMP gate setup using 0.22, 0.45, 0.88 and 1.34μm fluorescent polystyrene beads. (B) A representative example of control PMP (left) and patient PMP (right) diluted 1:10. Events falling outside the gate were not considered as PMP. Threshold for PMP collection set according to SSC. (C) Time-matched backgrounds of the diluents binding buffer (BB, left) and 1% bovine serum albumin diluted in phosphate buffered solution (BSA/PBS, right). (D) PMP counts for control samples (n=39), patient samples (n=56), BB background (n=16) and BSA/PBS background (n=13). Controls had significantly higher PMP counts than patients (p=0.01). Each data point shown represents 1 unique control or patient sample. Bars represent median with interquartile range.
Background and Aims

Plasma microparticles (PMPs) are small (0.1 to 1.0 μm) membrane bound vesicles, released from plasma membrane during cell activation and apoptosis. The cytoplasmic and surface contents of PMPs vary according to the parental cell’s lineage and physiological state. Altered numbers and profiles have been associated with thrombotic and inflammatory disorders including rheumatic diseases.

There have been variable reports on PMPs levels and profiles in Systemic Lupus Erythematosus (SLE). Some studies have reported higher levels while others showed similar or lower numbers with altered profiles. We compared PMPs in a multi-ethnic SLE cohort with a healthy control group, and explored their role in pathophysiology of SLE.

Methods

Clinical data and blood samples were collected from 56 consented SLE patients (median 45 years, 95% females) who fulfilled the ACR criteria, and 39 healthy adults. PMP pellet was obtained by high speed centrifugation. Flow cytometry was used to enumerate and profile the PMPs, utilising fluorescent polystyrene nanobeads, lipophilic dyes and fluorescent-labelled antibodies. HL60, a promyelocytic cell line, was utilised to study functional effects of PMPs in a smaller subgroup of samples (8 from each arm).

Results

SLE patients have lower PMP numbers compared to healthy controls (median: 12,925 vs 26,490 respectively, p=0.01, Figure 1). SLE PMPs also exhibited significantly higher proportions of leukocyte, and endothelial markers (Figure 2). SLE PMPs induced significantly higher p38 phosphorylation in HL60 cells compared to control PMPs (p=0.04, Figure 3).

Conclusions

Our data suggest that there are quantitative and functional differences in PMPs between SLE patients and healthy controls.