Capsaicin induced Central Neuronal Sensitization in the MIA model of OA pain
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Introduction:
Capsaicin, a natural irritant induces a painful burning sensation mediated by the activation of transient receptor potential vanilloid 1 (TRPV1). Intradermal injection of capsaicin produces a significant mechanical and thermal hyperalgesia in rats and increases BOLD signal in brain regions associated with nociceptive processing including thalamic, PAG, mesencephalic nuclei and the superior colliculus in anaesthetised rats. TRPV1 expression is up-regulated in experimental models of hyperalgesia (add review) and chronic pain conditions including osteoarthritis. The aim of the present study was to map cerebral pain processing network responses to intra-articular injection of capsaicin in an established model of OA pain and to identify associations with pain behavioural responses.

Materials and Method:
Experiments were conducted in accordance with the UK Animals (Scientific Procedures) Act of 1986. Sprague Dawley rats (160-200g) were anaesthetised and received intra-articular injection of MIA (1mg) into the left knee (n=16). Pain behaviour (weight bearing asymmetry and hindpaw withdrawal thresholds) was quantified up to day 28-32. Rats were randomly assigned to two groups: SAL-MIA/CAPS-MIA group (n=8) received an intra-articular injection of saline into the contralateral (right) knee joint followed by intra-articular injection of capsaicin (5µMµl) into the MIA pre-treated knee joint; SAL-MIA, CAPS-CON group (n=8) received intra-articular injection of saline in the MIA treated knee and intra-articular injection of capsaicin into the contralateral (right) knee. Rats were anaesthetised (isoflurane, maintenance ~ 2-2.5%) and MRI data were acquired using a Bruker 7T system with a receive-only head in combination with a volume coil for transmission. A total acquisition time of 1 hour was split into 10 minute baseline followed by 15 minute post vehicle saline and then 35minute post intra-articular capsaicin injection in a fixed order to avoid any potential carry over effects from capsaicin. The fMRI sequence was gradient echo EPI (TR=2000ms, TE=23ms, FOV=30mm, matrix: 64x64, 17 contiguous 1mm slices). fMRI data were analysed using FEAT. Data pre-processing was included brain extraction, motion correction, spatial smoothing of 1mm, global mean normalisation and registration to standard space. First level time-series statistical analyses were conducted using FILM with local autocorrelation corrections and block design. Within and between group level analyses were conducted with mixed effects analyses (FLAME), reported at corrected p-value of 0.05. BOLD responses were then correlated with pain behaviour. Functional connectivity analysis was performed using model free independent component (IC) analysis on the 35 minutes post intra-articular capsaicin injection fMRI time series. ICs reflecting the main networks were subjected to dual-regression analysis, and spatial differences in individual functional networks between the two groups were tested using the threshold free cluster enhancement permutation test, reported at corrected P<0.05.

Results:
Injection of capsaicin into the MIA-treated knee induced significant brain activation in the following brain areas: PAG, unilateral thalamus and bilateral mesencephalon, superior-colliculus and hippocampus (Figure 1); however capsaicin injection into the control (contralateral) knee did not yield significant brain activation. However, there was no significant difference between the two groups. In addition, there was no correlation between mechanical hindpaw withdrawal thresholds and brain activation. Model free independent component analysis identified 20 components. Dual regression analysis identified an exclusive effect of intra-articular injection of capsaicin on the mid brain network revealing increased functional connectivity between with the mediodorsal thalamic nucleus, hippocampus and globus pallidus in the CAP-MIA group compared to the SAL-MIA group (Figure 2). However, there was no correlation between mesencephalic functional connectivity and mechanical hindpaw withdrawal thresholds.

Discussion:
Intra-articular injection of capsaicin into the MIA knee resulted in significant, predominantly contralateral, brain activation in the thalamus and known regions involved in the processing of painful inputs. This effect was specific to the MIA knee and not replicated following intra-articular injection of capsaicin into the control knee, providing evidence for an augmented cerebral processing of this type of noxious stimulus. The lack of effect of capsaicin on brain activation under control conditions may arise as a result of the study being under-powered due to the small effect size of this treatment relative to the analgesic effect of the anaesthetic regimen that for animal welfare reasons had to be higher than in our previous studies. Alternatively, on-going noxious inputs from the MIA treated knee may have triggered descending inhibition control mechanisms that impact upon sensory inputs from the non-injected knee. The selective brain activation evoked by capsaicin when injected into the MIA-treated knee supports the mounting evidence for TRPV1 mediated sensitisation in osteoarthritis.

Figure 1: Group Mean fMRI activation map (red-yellow), for the MIA-group (n=8) that received capsaicin into the MIA knee and saline into the contralateral (right knee), overlaid on average T2-structural image (p<0.05 corrected). The blue line indicates the borders for a) PAG, b) superior-colliculus c) mesencephalic region d) thalamus e) hypothalamus medial f) hippocampus region.

Figure 2: Increased functional connectivity in the mediodorsal thalamic nucleus (top) and Globus Pallidus (bottom) overlaid on average T2-structural image; corrected P<0.05.

References: