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Pain prediction by serum biomarkers of bone turnover in people with knee osteoarthritis: an observational study of TRAcP5b and cathepsin K in OA

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SUMMARY

Objectives: To investigate serum biomarkers, tartrate resistant acid phosphatase 5b (TRAcP5b) and cathepsin K (cath-K), indicative of osteoclastic bone resorption, and their relationship to pain and pain change in knee osteoarthritis (OA).
Methods: Sera and clinical data were collected from 129 people (97 with 3-year follow-up) with knee OA from the Prediction of Osteoarthritis Progression (POP) cohort. Knee OA-related outcomes in POP included: WOMAC pain, National Health and Nutrition Examination Survey (NHANES) I (pain, aching and stiffness), subchondral sclerosis, and radiographically determined tibiofemoral and patellofemoral OA. Two putative osteoclast biomarkers were measured in sera: TRAcP5b and cath-K. Medial tibia plateaux were donated at knee arthroplasty for symptomatic OA ($n = 84$) or from 16 post mortem (PM) controls from the Arthritis Research UK (ARUK) Pain Centre joint tissue repository. Osteoclasts were stained for tartrate resistant acid phosphatase (TRAcP) within the subchondral bone of the medial tibia plateaux.
Results: Serum TRAcP5b activity, but not cath-K-immunoreactivity, was associated with density of TRAcP-positive osteoclasts in the subchondral bone of medial tibia plateaux. TRAcP-positive osteoclasts were more abundant in people with symptomatic OA compared to controls. Serum TRAcP5b activity was associated with baseline pain and pain change.
Conclusions: Our observations support a role for subchondral osteoclast activity in the generation of OA pain. Serum TRAcP5b might be a clinically relevant biomarker of disease activity in OA.

Introduction

Pain is the reason for most osteoarthritis (OA)-related medical visits. OA knee pain substantially impacts quality of life and is a key determining factor for loss of joint function[1]. Available drug treatments focus on analgesia, but often do not have sustained benefit and many patients experience unwanted side effects[2].

Although OA affects articular cartilage, it is increasingly recognised as a disease of the whole joint. Changes in subchondral bone are key in the pathogenesis of knee OA, and associated with knee pain[3] and radiographic progression[4]. Bone remodelling and increased pain mediators (cyclooxygenase 2, substance P, TNF-α) in the subchondral bone might occur before overt OA cartilage degeneration[5]. Subchondral bone is densely innervated by sensory nerves[6], and might be a key source of OA pain.

Animal models of OA and imaging studies in man support associations between pain and subchondral structural pathology[7–9]. In particular, increased osteoclast activity indicative of subchondral bone turnover might be associated with OA and pain[7,10]. Osteoclasts are multinucleated giant cells responsible for homeostatic bone resorption that release enzymatic markers, including tartrate

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resistant acid phosphatase (TRAcP) and cathepsin K (cath-K). TRAcP, originally called type 5 acid phosphatase, can be expressed both by osteoclasts and macrophages[11]; it was identified in human serum and separated electrophoretically into two distinct bands: 5a and 5b. Electrophoretic studies suggest band 5b tartrate resistant acid phosphatase 5b (TRAcP5b) is derived from osteoclasts and 5a from macrophages[12]. Cath-K, a cysteine protease, has been implicated in OA pathogenesis, largely because of its upregulation in areas of cartilage damage and resorbed bone[13,14]. Roles of cath-K in the initial stages of bone resorption have led to it becoming a target for novel therapeutic approaches for diseases such as osteoporosis, where reduced bone resorption can increase bone mineral density and reduce fracture risk[15]. Circulating TRAcP5b activity and cath-K are reduced in clinical trials during bisphosphonate treatment[16,17].

Bone and cartilage biomarkers have been investigated in OA structural progression[18,19], and some circulating inflammation biomarkers have been associated with OA pain, including C reactive protein (CRP), tumour necrosis factor (TNF)-α, interleukin (IL)-6[20] and interleukin (IL)-1β[21]. One study reports concentrations of N-telopeptide of type I collagen (uNTX-I) being significantly increased in people with OA knee pain (VAS score) independent of radiographic severity[22]. However, validated biomarkers of subchondral osteoclast activity associated with OA pain, or pain progression, have yet to be reported.

We hypothesised that biomarkers which reflect subchondral osteoclast activity, will be associated with OA pain, and might be useful in predicting pain progression in OA. The objectives of this study were to identify and validate serum biomarkers of subchondral osteoclast activity in people with symptomatic knee OA and to evaluate the association of these markers with OA pain, structural severity, and progression.

Patients and methods

Data reports a cross-sectional, case–control, cohort study.

Participants

129 participants from the Prediction of Osteoarthritis Progression (POP) cohort[19] and knee tissue from 100 subjects from the Arthritis Research UK (ARUK) Pain Centre joint tissue repository[23] were available (Table I). Included participants met the American College of Rheumatology (ACR) criteria for symptomatic OA[24]. Samples from 129 of the POP cohort were available at baseline and from 97 at 3-year follow up. Participants in the POP cohort who had unilateral total knee replacement (TKR) surgery before baseline blood and data collection were excluded, and those who had TKR before follow up were excluded from longitudinal analyses. Cases from the joint tissue repository had knee tissue taken at TKR surgery for symptomatic OA ($n = 84$), or post mortem (PM) ($n = 16$) from people who had not sought help for knee pain during the last year of life (asymptomatic control group). Sixteen cases from each of the TKR and PM groups were matched for macroscopic chondropathy scores, age and gender. Macroscopic chondropathy was scored by a single observer as previously described[25], taking account of severity (graded from 0 (normal unbroken surface) to 4 (subchondral bone exposure)) and extent (percentage of area involved by each grade) to calculate a chondropathy score from 0 to 100. Scores for all 4 compartments (medial and lateral tibial plateaux and femoral condyles) were summed to give a total chondropathy score from 0 to 400. Participants were excluded if they had specific bone disease known to affect bone turnover (e.g., Paget's disease of the bone, osteomalacia), or non-OA diagnoses as a cause of knee pain (e.g., rheumatoid arthritis, acute gout), but not according to medication use (Table I). Cases with self-reported osteoporosis were also included (Table I).

Table I
Demographics of patient study groups

	POP cohort		ARUK Pain Centre joint repository	
Number	Baseline; 129	Follow up; 97	TKR; 84*	PM; 16
Age (mean ± SD years)	64 ± 11	67 ± 11	66 ± 10	69 ± 12
Female (%)	72	72	57	56
BMI (mean ± SD kg/m^2)	31.4 ± 6.6	31.6 ± 6.7	31.3 ± 6.8	n/a
Osteoporosis (%)	17	16	0	0
Bisphosphonate use (%)	11	9	0	0

* Matched TKR cases ($n = 16$) were a subgroup of the total TKR cases used.

Imaging

Postero-anterior weight-bearing knee radiographs were obtained as previously described[25–27]. Radiographs of the POP cohort were scored by observers blinded to patient details for Kellgren–Lawrence (K/L) grade (0–4)[28] and individual radiographic features of OA including joint space narrowing (JSN 0–3), osteophytes (OST 0–3), subchondral sclerosis (0 or 1) and patellofemoral OA (0–3) using a standardized atlas[29]. Total scores were summed scores for both knees (right + left) and compartments (tibia − medial, lateral; femur − medial, lateral)[19]. Knee radiographs for cases providing joint tissues at TKR were scored using an atlas of line drawings of medial and lateral JSN and OST[30]. JSN (range 0–6) and OST (range 0–12) scores were summed to provide a total radiographic OA severity score for each knee (range 0–18). Radiographs were not available for PM cases.

Scintigraphic imaging of knees and whole body was performed as previously described[19,27]. The radiotracer methylene-diphosphonate labelled with technetium-99m was administered 2 h prior to imaging. Sixteen joint sites were scored semi-quantitatively by 2 experienced observers blinded to patient detail, on a scale of 0–3, where 0 = normal to 3 = intense. The scores were summed for each joint site. Scored sites included knees, shoulders, elbows, wrists, hands, hips, sacroiliac joints, ankles, forefeet, first metatarsophalangeal joints, sternoclavicular joints, acromioclavicular joints, the sternomanubrial joint, the cervical spine, the thoracic spine, and the lumbar spine.

Pain assessment

In the POP cohort, pain was assessed using the Likert pain scale of the Western Ontario and McMaster Universities Osteoarthritis index (WOMAC-A)[31]. It consists of 5 summed items (pain on walking, stair climbing, nocturnal, rest and weight bearing) scored from 0 = none, 1 = mild, 2 = moderate, 3 = severe and 4 = extreme, to give a total subscore ranging from 0 to 20. Knee symptoms were also ascertained by the National Health and Nutrition Examination Survey (NHANES) I criterion[32] of pain, aching or stiffness on most days of any 1 month in the last year; for subjects answering yes, symptoms were quantified as mild, moderate, or severe, yielding a total score of 0–3 for each knee. Change scores were calculated separately for WOMAC pain and NHANES I pain as follow-up score minus baseline score, summed across both knees, and used to define pain worsening or improvement in participants over 3 years as previously published[19]. Pain scores were not available for ARUK Pain Centre joint tissue repository cases, and sera were not available for PM cases.

Biomarker quantification

TRAcP5b activity and cath-K concentrations were analysed in serum stored at −80°C from participants in the POP cohort and