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Widespread somatosensory sensitivity in naturally occurring canine model of osteoarthritis

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Abstract

Osteoarthritis (OA)-associated pain is a leading cause of disability. Central sensitization (CS), as a result of OA, is recognized as an important facet of human patients' chronic pain and has been measured in people using quantitative sensory testing (QST) testing. The spontaneous canine OA model has been suggested as a good translational model, but CS has not been explored in this model. In this study, QST was performed on dogs with and without spontaneous hip or stifle OA to determine whether OA is associated with CS in this model. Mechanical (von Frey and blunt pressure) and thermal (hot and cold) sensory thresholds obtained in dogs with chronic OA-associated pain (n = 31) were compared with those of normal dogs (n = 23). Dogs were phenotyped and joint-pain scored, and testing was performed at the OA-affected joint, cranial tibial muscle, and dorsal metatarsal region. QST summary data were evaluated using mixed-effect models to understand the influence of OA status and covariates, and dogs with OA and control dogs were compared. The presence of OA was strongly associated with hyperalgesia across all QST modalities at the index joint, cranial tibial muscle, and metatarsal site. Mechanical QST scores were significantly moderately negatively correlated with total joint-pain scores. The spontaneous canine OA model is associated with somatosensory sensitivity, likely indicative of CS. These data further validate the canine spontaneous OA model as an appropriate model of the human OA pain condition.

Keywords: Animal model, Osteoarthritis, Spontaneous osteoarthritis model, Quantitative sensory testing

1. Introduction

Osteoarthritis (OA) is the most common form of arthritis and is considered a leading cause of disability in humans (27 million U.S. adults and 8.5 million U.K. adults).[33] Osteoarthritis is a major contributor to the economic impact of chronic pain, and a 2011 report from the Institute of Medicine highlighted that *the economic cost of persistent pain (approximately $600 billion annually) was more than the economic cost of cardiovascular disease ($300 billion) and cancer ($250 billion) combined*.[11,18]

Additionally, numerous reviews[5,35,40,48] have highlighted a crippling lack of translation of basic research into new approved therapeutics for treatment of persistent pain in humans. The use of spontaneous painful disease in companion animals was highlighted as one of the changes that could be made to help improve translation of basic science to new therapeutics.[38]

In dogs, spontaneously occurring OA is a condition affecting a large percentage of the population, with an estimated 20% to 30% of the dog population having OA and associated clinical signs.[25] The pathophysiology of canine OA of the hip is considered to be very similar to human OA,[13] making dogs suitable candidates to be used as spontaneous disease models.[24] Additionally, the canine stifle joint is considered to be among the most similar to the human knee joint.[14] The pain associated with canine OA can be difficult to control,[31] and indeed, OA-associated pain is one of the most common reasons for euthanasia in dogs.[32,36] One of the reasons for the difficulty in controlling pain in dogs may be the presence of central plasticity, as in humans with OA.[2,4,15] Although many aspects of this spontaneous canine OA model have been well developed including objective measures of limb use,[19] objective measures of activity,[50] validated owner-completed clinical metrology instruments,[8,49] and measures of sleep disturbance,[28] measures of enhanced processing of nociceptive stimuli resulting from peripheral and central mechanisms associated with spontaneous canine OA have received little attention. Early work has evaluated the repeatability of quantitative sensory testing measures,[6,51] and 2 small studies have suggested the presence of enhanced pain processing in association with cruciate ligament rupture[10] and hind limb OA.[51] Additionally, one study concluded that unilateral total hip replacement in client-owned dogs with bilateral hip OA resulted in decreased central sensitization (CS), as measured using von Frey

thresholds 12 months after surgery[47]; however, controls were not evaluated. To optimally exploit this spontaneous canine model of OA for future comparative and translational research, more needs to be understood about nociceptive pain sensitivity and central plasticity in association with OA.

The primary aim of this study was to evaluate mechanical and thermal quantitative sensory testing (QST) in dogs with chronic OA-associated pain and in matched control dogs. We hypothesized that QST would be able to detect differences in somatosensory processing between normal dogs and dogs with naturally occurring OA, paralleling the findings in humans with OA.

2. Material and methods

The study protocol was approved by the Institutional Animal Care and Use Committee of North Carolina State University (protocol 11-073-O).

2.1. Animals

Client-owned dogs of both groups (OA affected and controls) were recruited over a 14-month period. Recruitment was conducted with the assistance of the Clinical Studies Core at the NC State Comparative Medicine Institute (https://cvm.ncsu.edu/research/centers/cmi/) using email advertisements within the veterinary college and NC State University community, email and paper flyer advertisements at local veterinary practices in Raleigh, and through local newspaper advertisements. All owners were shown pictures and videos of the proposed procedures and gave verbal and written informed consent before entering their pet into the study.

2.2. Screening

All dogs were required to be older than 2 years and weigh ≥15 kg. Prestudy in-hospital screening consisted of physical examination including body condition score (BCS),[30] orthopedic and neurologic examination, complete blood count, serum biochemical analysis, and voided urinalysis. Orthopedic examination (performed by a single individual [D.K.] trained by B.D.X.L.) evaluated gait (lameness, stiffness, and posture), joint manipulation (range of motion, pain, crepitus, effusion, and thickening), and the degree of muscle atrophy. The total number of painful appendicular joints and spinal segments was recorded. There is no validated subjective pain assessment scale, and in this study, pain responses detected during joint and spinal segment palpation were scored on a 5-point scale, with 0 = no pain and 4 = severe pain (**Table 1**), similar to other studies.[20,46] Total pain score was recorded as a sum of scores across all joints and spinal segments. In dogs in which OA was present bilaterally in hips and/or stifles or in both hip and stifle joints, the index joint was determined based on the highest pain score, confirmed by a second opinion evaluation (B.D.X.L.). Radiographic examination was performed under sedation to confirm the presence of OA. Two orthogonal views were taken of all clinically painful appendicular joints and only dogs with subjectively moderate or severe radiographic OA were included. Dogs were sedated using a combination of dexmedetomidine (Zoetis, Florham Park, NJ) (0.003 mg/kg intravenously) and hydromorphone (West-Ward Pharmaceuticals, Cherry Hill, NJ) (0.05 mg/kg intravenously) and reversal with atipamezole (Zoetis) (0.1 mg/kg intramuscularly). Dogs were excluded if there was evidence of clinically detectable neurologic disease or other systemic disease that could be associated with pain.

Eligible dogs with OA were required to have a 6-month history of impaired mobility as reported by the owner. Mobility impairment was assessed through a review of owner-completed validated clinical metrology instruments: the Liverpool Osteoarthritis in Dogs (LOAD)[21,49] and Canine Brief Pain Inventory (CBPI).[8] The LOAD[21,49] is a 13-item instrument with all items reported on a 5-point Likert-type scale. Each item is scored between 0 and 4, and the item scores are summed to give an overall instrument score. The CBPI[8,9] is a 2-part instrument: the pain severity score is the arithmetic mean of 4 items scored on an 11-point (0-10) numerical scale, and the pain interference score is the mean of 6 items scored similarly. One question asks about quality of life, scored 1 to 5 (poor to very good). Dogs with OA were required to not be currently receiving any anti-inflammatory medications or other analgesics (eg, amantadine and gabapentin), and also not be on nutritional supplements (eg, fish oil and "joint supplements"), or have been on them for 6 weeks or more before the time to be included in the study.

Control dogs were recruited with the aim of representing the typical phenotype of the average dog with clinically significant OA in terms of age, breed, and weight, characteristics that are well known from other studies[31,49,50] and clinical orthopedics. Additional inclusion criteria were no evidence of abnormalities on orthopedic examination (no pain, no decreased muscle mass, no neurological deficits, and no joint instability or other pathology), no history of impairment recognized by owner, and not receiving anti-inflammatory medications, other analgesics, or nutritional supplements. Canine Brief Pain Inventory was also used to assess normality of mobility, where mean CBPI scores of less than 0.75 for pain intensity and pain interference and quality of life scores of 4 (very good) or greater[7] were required.

2.3. Sensory testing design

Quantitative sensory testing was performed in a dedicated space, the Gait Laboratory (a 15 × 5 m isolated room with 2 doors and no windows) at the NCSU College of Veterinary Medicine. Quantitative sensory testing data were collected on each dog 7 days after screening.

2.3.1. Devices

2.3.1.1. Electronic von Frey

The electronic von Frey (EVF) device (IITC model Almemo 2450; IITC Life Sciences Inc, Woodland Hills, CA) consisted of a 1000-g internal load cell connected to a modified 0.5-mm diameter

Table 1

Pain scoring system used during orthopedic evaluation of pain severity of appendicular and axial segments.

Pain score	Description of responses
0	Does not notice manipulation (no pain)
1	Orients to site on manipulation, does not resist or mild resistance (mild)
2	Orients to site, slight objection to manipulation (moderate)
3	Withdraws from manipulation, may vocalize, may turn to guard area (significant)
4	Tries to escape from manipulation, or prevent manipulation, may bite or show aggression on manipulation (severe pain)

For this scoring, the manus and pes were considered a "single-joint area." Other joints scored were the carpi, elbows, shoulders, tarsi, stifles, and coxofemoral joints. The axial skeletal regions scored were the cervical, thoracic, lumbar, and lumbosacral regions. The addition of scores for each individual joint or axial skeletal region comprised the total pain score for that individual.

pipette tip, as described previously.[6] The amount of force applied to a test site was measured and displayed with a resolution of 0.1 g and maximum of 1000 g.

2.3.1.2. Pressure algometer

The blunt-probed pressure algometer (PA) (SMALGO algometer; Bioseb, Vitrolles, France) was equipped with a flat, 3-mm diameter tip attached to a recording unit. Internal software of the device recorded the maximum force applied at the test site up to a maximum of 2500 g.

2.3.1.3. Thermal probe

The thermal device (NTE-2A; Physitemp Instruments, Clifton, NJ) was composed of a hand-held probe equipped with a flat, 13-mm diameter tip on a 5-foot lead connected to a digital temperature control unit and recirculating pump with combined water reservoir. The temperature range of the probe tip could be altered to have a temperature between 0°C and 50°C, maintained to within 0.1°C. This device was used to deliver a hot thermal stimulus (49°C) and a cold thermal stimulus (0°C).

2.4. Testing sites

Three test sites on each hind limb were located and identified (**Figure 1**), and consisted of the index joint (hip or stifle), the cranial tibial muscle, and the metatarsal area. The locations of testing at the 3 sites were as follows: (1) index joint: either the hip joint—testing site located 2 cm craniodorsally from greater trochanter or the stifle joint, testing site located 2 cm laterally from apex of patella, (2) tibial muscle—center of cranial tibial muscle at dorsolateral position, and (3) metatarsal—dorsal

Figure 1. The locations of testing at the 3 sites were as follows: (1) index joint: either the hip joint—testing site located 2 cm craniodorsally from greater trochanter or the stifle joint, testing site located 2 cm laterally from apex of patella, (2) tibial muscle—center of cranial tibial muscle at dorsolateral position, and (3) metatarsal—dorsal surface of the metatarsus, between metatarsal bones III and IV.
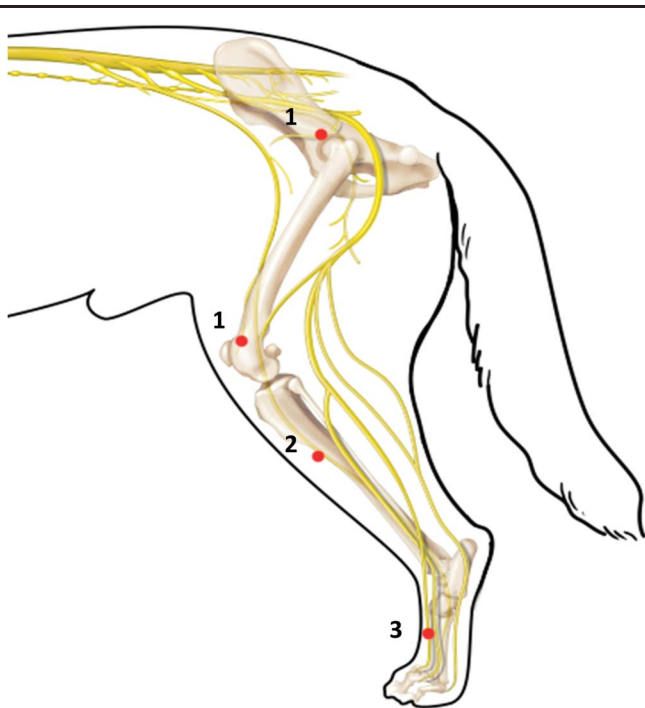
surface of the metatarsus between metatarsal bones III and IV, as previously described.[37]

The endpoint for both thermal and mechanical stimuli was defined as a behavioral response indicative of a conscious perception of the stimulus: movement of the limb away from the tip or probe with conscious perception, turning the head to look directly at the site, vocalization, or other consistent, clearly recognizable body movement indicating perception of the stimulus (see supplementary data: Video 1, available online at http://links.lww.com/PAIN/A240). Simple reflex movements (such as twitching) were not considered an endpoint. Each dog was assigned a feasibility score as previously described,[6] which represents the ease with which the data could be collected for each device and each site (0-5 scale with 0 indicating no problem in collecting the data and 5 indicating the data were impossible to collect).

2.5. Protocol

Gait Laboratory room temperature was maintained between 21.7°C and 22.2°C. All dogs were introduced to the Gait Laboratory for acclimatization and were allowed to freely explore the environment for 20 minutes before testing. Two people were used for all QST procedures and data collection, but all sensory threshold measurements were collected by the same operator (D. K.). Food rewards were offered to dogs occasionally and fresh water was available in the room ad libitum. After acclimating, dogs were placed in lateral recumbency on a standard cushioned mat (0.5 cm thick, 2.0 m long, and 1.0 m wide). For measuring the thresholds on the left pelvic limb, dogs were placed in the right lateral recumbency, and vice versa. The devices were used in a set order at each site: EVF, PA, hot thermal, and then cold thermal. The order of testing sites for cases was randomized at the start of the study, using blinded drawing of numbers from a hat.

Mechanical stimuli (EVF and PA) were applied as ramped stimuli, with steadily increasing force applied until the behavioral response was elicited or the maximum value was reached (EVF, 1000 g; PA, 2500 g). The left hand of the operator (D.K.) was placed behind the paw for support during metatarsal-site testing and on the medial side of tibia during muscle-site testing. Thresholds were measured in grams.

The hot and cold stimuli were applied as fixed intensity stimuli. Hair on the 3 sites was clipped creating a 15-mm diameter area before any testing. Hot and cold stimuli represented probe settings of 49°C (cutoff time, 20 seconds) and 0°C (cutoff time, 60 seconds), respectively. The latency to respond was measured in seconds for both thermal stimuli using a digital stopwatch (Seiko W073; Seiko, Tokyo, Japan) with the precision of one-hundredth of a second.

Data were collected on both hind limbs, with 5 trials per site on each limb and a 60-second intertrial interval between the applications of stimuli.

2.6. Statistical analysis

Group comparisons of demographics were performed using Mann–Whitney tests. The primary measures of interest were the QST measures, and sample size calculations were based on previous von Frey threshold data. In pilot work in our laboratory, the expected mean difference between normal dogs and dogs with OA was found to be 158 g with a combined SD of 127 g, indicating that for a power of 90%, at an alpha of 0.05, 15 dogs would be needed per group.

Exploratory analyses tested for a linear effect across trials in a repeated-measures mixed-effects model, and this analysis did not reveal that a strong linear effect across trials was present. The QST data were evaluated using repeated-measures mixed-effects models to understand the influence of covariates. Separate mixed-effect repeated-measures models were made for each QST modality (EVF, PA, hot thermal probe, and cold thermal probe) at each of the 3 testing sites (index joint, muscle, and metatarsal) such that there were twelve models in total. In each model, the following covariates were included: OA status, sex, side (left or right), body condition score, and the feasibility of the QST measurement. The optimal covariance structure for the within-subject measures across trial and side was tested using likelihood ratio tests, starting with the most complicated co-variance structure first and then sequentially comparing simpler designs. The covariance designs that were compared for the within-subject measures across trial were random slopes with unstructured and Toeplitz covariance structure. The within-subject random intercept for side was compared with a model with just a random subject-specific intercept. At minimum, each model included a random subject-specific intercept.

The association between OA, sex, side, body condition score, and the feasibility of the QST measurement and the QST threshold was assessed using Wald tests. P values were not adjusted for multiple comparisons.

Following the above analysis, direct group comparisons for the average of each QST modality across trial and side were made using nonparametric analyses (Wilcoxon rank sums).

In exploratory analysis, correlations were performed between mechanical QST thresholds and clinical parameters (total pain score, number of painful joints, LOAD, and CBPI) using Spearman rank correlation coefficient (Spearman rho), denoted "r." Correlations were interpreted as very weak (0-0.19), weak (0.2-0.39), moderate (0.4-0.59), strong (0.6-0.79), or very strong (0.8-1.0).[16]

3. Results

Fifty-four dogs, 31 with OA, were recruited for the study. The demographics are shown in **Table 2**. Of the enrolled dogs with OA, 20 dogs had the hip joint identified as the index joint and 11 dogs had the stifle joint identified as the index joint. Most of the examined dogs had concurrent stifle and hip OA. Only 2 dogs had unilateral OA with only one joint affected.

3.1. Comparison of clinical parameters of dogs

When comparing all dogs from both groups (OA, controls), the age of the dogs with OA was significantly higher (P = 0.007), although the difference was only 2 years, with the control dogs being younger. Bodyweight was not significantly different (P = 0.523) when comparing all dogs from both groups (OA, controls).

3.2. Quantitative sensory testing data

3.2.1. Mixed-effects models to determine which factors affect thresholds

Presence of OA was a strong predictor of the QST area under the curve (AUC) summary measure in all of the models (see **Tables 3–6**) except for the PA at the metatarsal site (**Table 3**) and the EVF at the index joint site and the tibial muscle site (**Table 4**). No other factors consistently or significantly affected QST measures across all modalities. We found that feasibility may be an important

Table 2

Summary of characteristics of both groups (osteoarthritis [OA] and controls).

	OA (N = 31)	Control (N = 23)	All (N = 54)
Sex			
Male castrated	15	11	26
Female spayed	16	12	28
Age (y)			
Median	8.00	7.00	7.00
Range (min-max)	4-17	3-11	3-17
Mean	8.71	6.74	7.67
SD	2.77	1.91	2.71
SEM	0.50	0.40	0.37
Bodyweight (kg)			
Median	29.00	28.30	28.90
Range (min-max)	18-50.2	21-42	18-50.2
Mean	31.05	29.76	30.50
SD	7.69	7.03	7.37
SEM	1.38	1.47	1.00
Total pain score			
Median	7.00	0	4.50
Range (min-max)	3-23	0	0-23
Mean	9	0	5.17
SD	4.89	0	5.81
SEM	0.88	0	0.79
Total number of painful areas			
Median	4.00	0	2.00
Range (min-max)	2-12	0	0-12
Mean	4.84	0	2.78
SD	2.63	0	3.12
SEM	0.47	0	0.43
Breed			
Mixed breed	12	8	20
Labrador retriever	6	4	10
Golden retriever	3	6	9
American Staffordshire terrier	2	1	3
Boxer	1	0	1
Catahoula leopard	1	0	1
English setter	1	0	1
Australian cattle dog	1	1	2
Brittany spaniel	1	0	1
Otterhound	1	1	2
Rottweiler	1	0	1
Welsh springer spaniel	1	0	1
Australian shepherd dog	0	1	1
German shorthaired pointer	0	1	1

predictor for the EVF and hot thermal thresholds at the index joints (**Table 4 and 5**, respectively). We also found that BCS seemed to be strongly associated with the hot thermal measures at the metatarsal and tibial muscle sites (**Table 5** middle and right columns). Age, sex, and side did not seem to be strongly associated with any of the QST AUC measures at any of the 3 sites (index joint, tibial muscle, and metatarsal) for any QST modality.

3.2.2. Direct group comparisons

Following the aforementioned analysis, QST values were averaged over the left and right limbs, across all trials, for each dog. QST results for all dogs with OA and normal dogs are detailed in **Table 7**. P values were not adjusted for multiple comparisons; however, if a correction to the critical P value of 0.05/3 (=0.016) is used for each QST modality (3 different sites), it can be seen (**Table 7**) that all comparisons are significant at the 0.016 critical level.

Table 3

Fixed-effects estimates for predicting the blunt pressure thresholds (pressure algometer) at the affected joint, the metatarsal site, and the tibial muscle.

Effect	Index joint			Metatarsal			Tibial muscle		
	Estimate	95% CI	P	Estimate	95% CI	P	Estimate	95% CI	P
Intercept	2359.4			1407.4			1270.8		
OA	−423.0	−681.8 to −164.2	0.001	−193.7	−411.7 to 24.2	0.081	−374.0	−653.9 to −94.1	0.009
Age	−8.3	−53.5 to 36.9	0.718	−26.5	−63.8 to 10.8	0.163	−20.7	−69.6 to 28.2	0.405
Female	−61.5	−273.4 to 150.3	0.568	−62.9	−245.0 to 119.2	0.498	−79.2	−308.3 to 149.9	0.496
Side = L	−29.2	−106.4 to 48.1	0.452	2.3	−60.7 to 65.3	0.942	65.7	−13.7 to 145.2	0.103
BCS	−61.2	−235.2 to 112.7	0.489	25.6	−122.0 to 173.1	0.734	70.5	−117.6 to 258.7	0.461
Feasibility	26.5	−107.1 to 160.2	0.697	45.3	−51.7 to 142.4	0.359	11.6	−132.9 to 156.2	0.874

For the fixed-effects estimates, the reference value is the first trial, right side, and male gender. The intercept represents the expected average QST value for the first trial if the dog was a control male measured on the right side. BCS, body condition score; CI, confidence interval; L, left; OA, osteoarthritis.

3.2.2.1. Electronic von Frey

Dogs with OA showed significantly lower thresholds at the index joint and metatarsal sites ($P = 0.006$ and $P = 0.014$, respectively) (**Table 7**).

3.2.2.2. Pressure algometer

Dogs with OA showed significantly lower thresholds at the index joint, tibial muscle, and metatarsal sites ($P < 0.0001$, $P = 0.001$, and $P = 0.005$, respectively) (**Table 7**).

3.2.2.3. Thermal probe—heat

Dogs with OA showed significantly lower thresholds at the index joint, tibial muscle, and metatarsal sites ($P < 0.0001$, $P = <0.001$, and $P < 0.0001$, respectively) (**Table 7**).

3.2.2.4. Thermal probe—cold

Dogs with OA showed significantly lower thresholds at the index joint, tibial muscle, and metatarsal sites ($P < 0.0001$, $P = 0.001$, and $P = <0.001$, respectively) (**Table 7**).

3.3. Correlations

There were significant moderate negative correlations between mechanical thresholds at the metatarsal site and total pain scores (EVF [$r = −0.49$, $P = 0.006$] and PA [$r = −0.37$, $P = 0.04$]). There were significant weak or moderate negative correlations between the total number of painful joints and mechanical thresholds at the index joint site (EVF [$r = −0.38$, weak correlation, $P = 0.04$] and PA [$r = −0.44$, moderate correlation, $P = 0.02$]) and also the metatarsal site (EVF [$r = −0.44$, moderate correlation, $P = 0.02$]).

Total pain score and LOAD and CBPI scores were significantly positively correlated (LOAD [$r = 0.42$, moderate correlation, $P = 0.02$] and CBPI [$r = 0.37$, weak correlation, $P = 0.04$]). The number of painful joints was significantly and weakly positively correlated with CBPI scores ($r = 0.39$, $P = 0.03$), but not with LOAD scores ($r = 0.34$, $P = 0.06$). There were no significant correlations between owner-completed clinical metrology instruments (CMIs) and QST values for any modality.

4. Discussion

We found significantly lower mechanical and thermal thresholds at index joints and at sites remote from the affected joints (tibial muscle and metatarsal) in dogs with OA as compared with control dogs without OA. This increased sensitivity to mechanical and thermal stimuli remote from the OA-affected joint could indicate the presence of enhanced pain processing resulting from central and, or, peripheral sensitization, with peripheral sensitization driven by circulating proinflammatory or pain-promoting substances. Our results support the findings of facilitated nociceptive transmission because of central plasticity in rodent models of OA[22,26] and humans with OA.[2,23,45] However, it is not known from this work whether the sensitization is driven by spinal cord changes, or alterations in endogenous descending pathways as have been described in humans,[41] or, indeed, is driven by peripheral mechanisms. The facilitated nociceptive transmission could be a product of all 3 mechanisms. These data further validate the canine spontaneous OA model as an appropriate model of the human OA pain condition. Previous work has shown mechanical and thermal QST testing to be highly feasible and repeatable in normal, client-owned dogs,[6] and our present work indicates that QST data can be collected easily in dogs with OA,

Table 4

Fixed-effects estimates for predicting the von Frey thresholds (electronic von Frey) at the affected joint, the metatarsal site, and the tibial muscle.

Effect	Index joint			Metatarsal			Tibial muscle		
	Estimate	95% CI	P	Estimate	95% CI	P	Estimate	95% CI	P
Intercept	1114.3			532.5			471.8		
OA	−118.0	−245.4 to 9.5	0.070	−141.6	−266.9 to −16.2	0.027	−144.6	−280.3 to −8.9	0.037
Age	7.6	−14.7 to 29.9	0.503	−0.6	−22.0 to 20.7	0.952	−2.9	−26.6 to 20.9	0.813
Female	−20.9	−124.5 to 82.6	0.692	−19.8	−124.9 to 85.3	0.711	12.1	−98.1 to 122.3	0.829
Side = L	−19.4	−64.8 to 26.0	0.396	27.5	−7.5 to 62.6	0.121	−22.9	−62.3 to 16.6	0.250
BCS	−73.9	−159.5 to 11.8	0.091	5.1	−79.3 to 89.6	0.905	15.5	−75.7 to 106.7	0.739
Feasibility	−64.4	−125.4 to −3.5	0.038	6.5	−49.2 to 62.1	0.819	3.1	−61.8 to 68.0	0.925

For the fixed-effects estimates, the reference value is the first trial, right side, and male gender. The intercept represents the expected average QST value for the first trial if the dog was a control male measured on the right side. BCS, body condition score; CI, confidence interval; L, left; OA, osteoarthritis.

Table 5

Fixed-effects estimates for predicting hot thermal latencies (hot thermal) at the affected joint, the metatarsal site, and the tibial muscle.

Effect	Index joint			Metatarsal			Tibial muscle		
	Estimate	95% CI	P	Estimate	95% CI	P	Estimate	95% CI	P
Intercept	9.6			3.9			1.8		
OA	−6.8	−9.0 to −4.6	<0.001	−6.5	−9.1 to −3.8	<0.001	−7.1	−9.7 to −4.4	<0.001
Age	0.2	−0.2 to 0.6	0.301	0.1	−0.3 to 0.6	0.652	0.2	−0.3 to 0.7	0.393
Female	−0.3	−2.1 to 1.6	0.767	0.6	−1.6 to 2.8	0.610	−0.8	−3.0 to 1.4	0.473
Side = L	0.3	−0.7 to 1.2	0.548	−0.3	−1.3 to 0.7	0.583	−0.8	−2.1 to 0.6	0.267
BCS	1.5	0.0 to 3.1	0.055	2.6	0.7 to 4.4	0.006	2.6	0.7 to 4.5	0.007
Feasibility	−1.0	−1.9 to −0.1	0.038	−0.6	−1.6 to 0.4	0.208	−1.0	−2.1 to 0.1	0.080

For the fixed-effects estimates, the reference value is the first trial, right side, and male gender. The intercept represents the expected average QST value for the first trial if the dog was a control male measured on the right side. BCS, body condition score; CI, confidence interval; L, left; OA, osteoarthritis.

making this canine model of spontaneous OA useful for the evaluation of somatosensory processing.

Lower thresholds at the affected joint of dogs with OA compared with normal controls could be due to a combination of peripheral and CS, but lower thresholds remote from the affected joints are generally inferred to be due to CS. However, it is plausible that this remote hyperalgesia may be partly driven by generalized changes in the periphery, although this concept has not received any discussion, even in recent reviews.[3,12]

QST is the most common method used to assess pain sensitivity through application of a standardized stimulus to a peripheral tissue and the recording of a subject's response.[1] We found that dogs with long-standing OA-associated pain showed lower thresholds across all QST modalities (EVF, PA, hot thermal, and cold thermal). Most studies in human subjects assessing mechanical thresholds have reported lower thresholds in patients with OA than in controls, regardless of whether the devices were applied at the affected joint (eg, knee) or remote sites, including thumb and shoulder[34] or forearm.[53] For example, Imamura et al. reported significantly lower mechanical thresholds in people with knee OA, than in controls, at 18 anatomical test sites including upper thigh and lower back.[23] A recent systematic review and meta-analysis concluded that there was a large difference in pain pressure thresholds between patients with knee OA and normal controls, with patients with knee OA showing increased sensitivity at both the local site (joint) and remote sites.[17] Evaluating thermal thresholds, one study reported that human patients with OA had significantly higher median warm and cold detection thresholds (hypoesthesia) than healthy participants at the knee, but not the forearm.[53] Recording of any threshold in nonverbal species is dependent on the behavioral endpoints of reactions to the stimulus. Whether these reactions represent thermal detection thresholds or tolerance thresholds is currently unknown. We assume that our recordings relate to "hot pain thresholds" as measured in humans.[29,52,53] We detected hot thermal hyperalgesia at all 3 testing locations in dogs with OA compared with controls. The recent systematic review of QST in patients with knee OA compared with controls concluded that patients with knee OA have significant hot pain sensitivity at remote sites, but not at local sites (affected joints).[17] However, increased sensitivity to hot pain has been demonstrated in human subjects with chronic musculoskeletal pain[39,44] and in patients with high-symptom OA.[27] The dogs in this study represented moderate-to-severe OA, suggesting similarity to the findings in humans. Our finding of cold hypersensitivity also supports the finding of previous work in another spontaneous pain model in dogs, cruciate ligament rupture.[10] In humans, widespread cold hypersensitivity was detected in subjects with chronic whiplash-associated pain,[42] and although a recent meta-analysis was not able to draw conclusions on cold pain sensitivity in patients with knee OA because of insufficient data, increased cold pain sensitivity has been demonstrated in patients with high-symptom OA.[27]

Our statistical models indicated that the dominant factor affecting QST values across all modalities was OA status. Overall, this naturally occurring model of OA in dogs would seem to be a valid model of OA-associated somatosensory sensitivity indicative of CS. In this study, dogs were recruited to the control group to be generally similar in age, bodyweight, and breed as the OA population (which is well characterized in veterinary medicine). The control group was significantly younger, by 2 years; however, the statistical models indicated little influence of age on QST thresholds in our cohorts. Our recommendation is that future

Table 6

Fixed-effects estimates for predicting cold thermal latencies (cold thermal) at the affected joint, the metatarsal site, and the tibial muscle.

Effect	Index joint			Metatarsal			Tibial muscle		
	Estimate	95% CI	P	Estimate	95% CI	P	Estimate	95% CI	P
Intercept	53.0			25.7			46.0		
OA	2.0	1.0 to 3.0	<0.001	−17.9	−28.0 to −7.8	0.001	−13.7	−20.5 to −6.8	<0.001
Age	−12.4	−17.9 to −6.9	<0.001	0.5	−1.3 to 2.3	0.591	−0.3	−1.6 to 0.9	0.581
Female	0.7	−0.3 to 1.6	0.197	−1.8	−11.5 to 7.9	0.716	−2.8	−9.3 to 3.7	0.396
Side = L	−4.0	−9.3 to 1.3	0.140	3.3	0.1 to 6.5	0.043	−0.9	−5.2 to 3.3	0.664
BCS	2.4	−0.3 to 5.1	0.076	3.2	−6.1 to 12.4	0.500	2.4	−3.9 to 8.8	0.453
Feasibility	−0.7	−5.9 to 4.4	0.773	−2.7	−8.0 to 2.6	0.316	−1.0	−4.2 to 2.3	0.554

For the fixed-effects estimates, the reference value is the first trial, right side, and male gender. The intercept represents the expected average QST value for the first trial if the dog was a control male measured on the right side. BCS, body condition score; CI, confidence interval; L, left; OA, osteoarthritis.

	OA	Controls	*P*
EVF (g)			
I	558.2 (191.5)	721.1 (200.4)	0.006
T	370.4 (144.3)	514.9 (251.3)	0.055
M	380.9 (159.1)	521.1 (216.8)	0.014
PA (g)			
I	1482 (368.2)	1959 (376.6)	<0.0001
T	1100 (358.2)	1523 (514.1)	0.001
M	1106 (344.2)	1338 (308.6)	0.005
Hot thermal (s)			
I	13.07 (4.193)	18.5 (2.098)	<0.0001
T	10.35 (4.589)	15.54 (4.112)	<0.001
M	12.79 (4.936)	17.31 (3.548)	<0.0001
Cold thermal (s)			
I	44.17 (11.68)	57.29 (5.729)	<0.0001
T	41.81 (11.2)	52.77 (9.574)	0.001
M	34.26 (13.41)	50.89 (12.62)	<0.001

EVF, electronic von Frey; I, index joint; M, metatarsal; OA, osteoarthritis; PA, pressure algometer; T, tibial.

studies should recruit controls that are similar to the affected population, and consider formal matching of controls for age, bodyweight, and BCS. Increasing BCS (correlating to increasing obesity) was associated with increased sensitivity to mechanical stimuli in this study, and although the relationship between obesity and QST has not been evaluated in humans, obesity is a risk factor for increased OA pain in humans.[43]

Validated clinical metrology instruments for owners of dogs (CBPI and LOAD)[8,9,21,49] were used in this study to measure owner evaluations of pain and mobility impairment and to ensure inclusion of normal, healthy, control dogs. Furthermore, these scores were found to positively correlate with total pain scores (assessed during evaluation of joints). Although increasing pain scores were negatively correlated with mechanical thresholds (as are self-reported pain scores in humans with OA[27]), there was no correlation between CBPI or LOAD values and QST values. Although the CBPI and LOAD primarily assess aspects of mobility and a dog's ability to perform certain activities that may be influenced by pain, these instruments are not measuring pain directly, although the CBPI pain severity subscale does ask about pain.

5. Conclusions

Our study indicates that dogs with long-standing OA and associated pain show increased somatosensory sensitivity, which is inferred to be, at least in part, due to central plasticity. Our study further supports spontaneous OA and associated pain in dogs as a valid model of human OA-associated pain, increasing the potential for use of this model in the development of therapies for human OA pain.

Conflict of interest statement

B.D.X. Lascelles has acted as a paid consultant for Boehringer Ingelheim Vetmedica and received honoraria for continuing education lectures. No product sold by Boehringer Ingelheim Vetmedica was used in this study, and Boehringer Ingelheim Vetmedica did not have input into the study design, data collection, or analysis. The other authors have no conflicts of interest to declare.

Acknowledgements

Appendix A. Supplemental Digital Content

Supplemental Digital Content associated with this article can be found online at http://links.lww.com/PAIN/A240.

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