Original Article

Interleukin-6 and Interleukin-8 Gene Expressions Differ Between Male and Female Patients at Time of Hip Arthroscopy for Femoroacetabular Impingement Syndrome

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Purpose: To identify key molecular components within the femoroacetabular impingement hip and compare the findings between male and female patients across varying age groups. **Methods:** All patients undergoing hip arthroscopy for femoroacetabular impingement syndrome (FAIS) without hip dysplasia were included. During hip arthroscopy, performed at University of Wisconsin Health, loose articular cartilage, excess synovium, damaged labral tissue, and minimal adipose tissue were debrided only as needed for visualization and tissue repair purposes and collected. Tissue was processed and used for quantitative polymerase chain reaction (qPCR). Genes were selected for qPCR on the basis of their associated function in inflammation and/or extracellular matrix remodeling during the progression of osteoarthritis. **Results:** A total of 91 male (M) and female (F) patients 15 to 58 years old were included in the study. qPCR results indicated that Interleukin-6 ($P < .05$, 95% confidence interval [CI] 0.047-0.083 F, 0.070-0.12 M) and Interleukin-8 ($P = .04$, 95% CI 0.059-0.10 F, 0.082-0.18 M) were significantly greater in male patients compared with female patients regardless of age, and *IL6* ($P = .02$, 95% CI [0.026-0.070] F, [0.067-0.17] M), Interleukin-1ß ($P < .01$ 95% CI [0.013-0.063] F, [0.073-0.25] M), and Matrix metalloproteinase-13 ($P = .047$, 95% CI [0.0051-0.017] F, [0.0084-0.052] M) were significantly greater in male patients younger than 20 years old compared with female patients younger than 20 years old. **Conclusions:** In patients with FAIS, there are significant differences between male and female patients in the biomarkers present in the affected hip at the time of surgery. Male patients have greater levels of *IL6* and *IL8* and male patients younger than 20 years of age have greater levels of *IL1β*, *IL6*, and *MMP13* compared with age-matched female patients. **Clinical Relevance:** A better understanding of the molecular markers present during varying stages of FAIS and in patients of different ages will help characterize the pathologic process behind FAIS. This may also help define future methods of targeted treatment and prevention of disease progression.

Femoroacetabular impingement (FAI) is believed to be a leading cause of early hip osteoarthritis (OA). Femoroacetabular impingement syndrome (FAIS) occurs when excess bone on the femoral head-neck junction or acetabular rim results in abnormal femoroacetabular contact and subsequent labral injury and cartilage damage.[1-3] A better understanding characterizing the mechanism of OA development from FAI could improve outcomes from interventions. One specific intervention,[1] hip arthroscopic surgery, is a proven minimally invasive, safe, and effective means to treat FAIS.[4]

Hip arthroscopy with labral repair and removal of impinging bone relieves a patient's pain related to labral and impingement pathologies and allows patients to return to their desired levels of function.[4-7] Unfortunately, the time for a definitive diagnosis of FAIS can average 3.1 years and 4.2 visits to health care providers before the appropriate diagnosis is made.[8,9] As time progresses, FAIS-induced, irreversible cartilage damage greatly impedes patients' chances for improvement after arthroscopic surgery and may contribute to the

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development of OA.[10-12] Early intervention in FAIS is associated with improved patient-reported outcomes.[13-15] Therefore, the ability to provide quantifiable evidence that early surgical intervention of FAIS can prevent irreversible cartilage damage and OA progression would lessen the burden of disease on future patients with FAIS. Because the pathophysiology of hip OA is composed of several distinct processes, understanding the mechanisms associated with OA progression may offer the greatest potential to treat or prevent its advancement.

A number of molecular factors are presumed to contribute to OA progression. These include factors relating to bone and cartilage metabolism, collagen production, inflammation, and pain regulation (Table 1).[16] For example aggrecan (ACAN) is an integral proteoglycan component of the extracellular matrix in cartilaginous tissues.[12] ACAN is actively degraded by enzymes, including matrix metalloproteinases, during development of OA and synthesized by chondrocytes to stabilize cartilage damage and essential for skeletal development.[17] Type II collagen (COL2) is the most abundant collagen within cartilage and provides the structural framework of the cartilage extracellular matrix. Type II collagen turnover is directly correlated with cartilage homeostasis and joint health. Interleukin 1β (IL1β), interleukin 8 (IL8), and interleukin-6 (IL6) are proinflammatory cytokines present in most arthritic conditions and key mediators of tissue inflammation and destruction.[18]

The upregulation of inflammatory cytokines has been linked to poorer OA prognoses. For instance, elevated levels of IL6 were associated with radiographic knee OA and cartilage loss in older adults.[19] P21, a cyclin-dependent kinase inhibitor, has multiple functions in the cell cycle, including mediation of cellular apoptosis.[20] Catabolic factors play a role in triggering inflammation and cartilage destruction through the production of matrix metalloproteinase-13 (MMP13) and a disintegrin-like and metalloproteinase with thrombospondin (ADAMTS), enzymes that cleave structural proteins within the cartilage matrix.[21] MMP13 also plays an active role in type II collagen degradation. A better understanding of the molecular environment within the hip will advance the ability to diagnose, predict, and treat FAIS.

The purpose of our study was to identify key molecular components within the FAI hip and compare the findings between male and female patients across varying age groups. We hypothesized that patients with FAIS would exhibit different molecular profiles across age and sex groups.

Table 1. Biomarkers and Their Functions

Biomarker	Function in Osteoarthritis Development
Aggrecan (ACAN)	Cartilage degradation
Interleukin-1ß (IL1B)	Inflammation, activation of protein degradation; suppresses cartilage synthesis
Interleukin-8 (IL8)	Inflammation
Interleukin-6 (IL6)	Inflammation
Matrix metalloproteinase-13 (MMP13)	Degradation of the extracellular matrix
Collagen Type II (COL2A1)	Key component of cartilage
*ADAMSTS1	Degradation of the extracellular matrix
*ADAMSTS5	Degradation of the extracellular matrix
Cartilage oligomeric matrix protein (COMP)	Cartilage metabolism; associated with OA progression
P21	Cell-cycle arrest

ADAMSTS, a disintegrin-like and metalloproteinase with thrombospondin; OA, osteoarthritis.

Methods

Patient Selection

All research was approved by the institutional review board at our institution and informed consent was obtained from all patients. Inclusion criteria were patients undergoing hip arthroscopy at University of Wisconsin Health with a diagnosis of FAIS, without dysplasia, on the basis of clinical and radiographic findings with magnetic resonance imaging confirmation of a labral tear. All patients who were ultimately offered hip arthroscopy had not responded to conservative treatment, including activity modification, oral nonsteroidal anti-inflammatory drugs, physical therapy, and in some cases intra-articular cortisone injections. Institutional review board approval for this work was limited to patient identification by age and sex and intraoperative tissue collection and did not permit data analysis of clinical characteristics.

Tissue Sample Collection

Once hip arthroscopy had been indicated, the procedure ensued per current standard of care. This included hip distraction via post-less traction, and the use of an interportal capsulotomy to access the central compartment of the hip joint (Fig 1 A and B). During the procedure, loose articular cartilage, excess synovium, damaged labral tissue, and minimal adipose tissue were debrided only as needed for visualization and tissue repair purposes and collected via a filter positioned between the suction tubing from the arthroscopic shaver and the suction tubing which was passed off the table to the nonsterile field (Fig 2). Tissue collection was standardized as it was collected by a single surgeon (A.M.S.). Each patient received treatment according to the standard of care. This tissue debris was only collected from the central compartment of the hip joint, and the collection device was detached

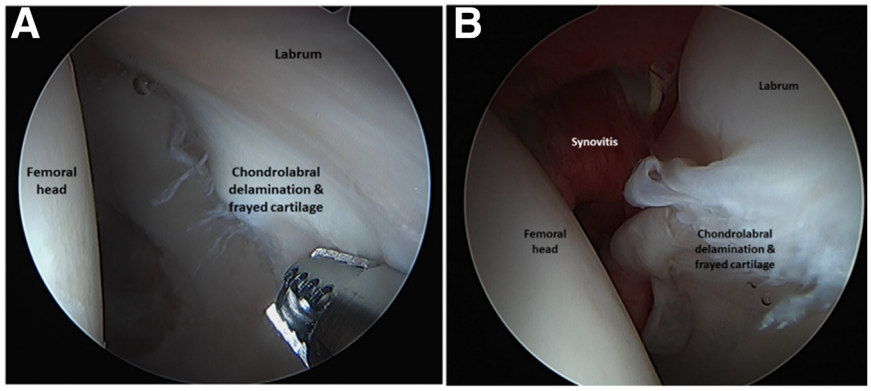

Fig 1. Representative intraoperative images of a patient with femoroacetabular impingement syndrome. Image depicts a left hip in an 18-year-old male patient. This image demonstrates the frayed and damaged cartilage at the chondrolabral junction will be debrided by the shaver as viewed through the accessory medial portal (A) and as viewed through the anterolateral portal (B).

once labral repair and unstable cartilage debridement were complete, prior to entering the peripheral compartment. Samples were immediately placed in DNA/RNA shield (Zymo Research, Irvine CA) and transported to the laboratory while on ice. Upon arrival, samples were washed with phosphate-buffered saline, weighed, and placed at −80°C until further use.

Quantitative Reverse Transcription Polymerase Chain Reaction (PCR)

Total RNA was isolated from tissue samples combining the TRIzol (Invitrogen, Carlsbad, CA) method with column fractionalization steps of the RNA midiprep kits (Zymo Research, Tustin, CA), according the manufacturers' protocols. Tissue was homogenized using a PowerGen 500 homogenizer (Fisher Scientific, Pittsburgh, PA). Subsequent to RNA isolation, yield and purity of RNA was quantified by nanodrop spectrophotometric measurement at 260 nm (Nanodrop Technologies, Wilmington, DE). Total RNA was reverse transcribed into cDNA using the High Capacity Reverse Transcription kit (Applied Biosystems, Foster City, CA). Quantitative PCR (qPCR) was performed using a Bio-Rad thermocycler (Bio-Rad, Hercules, CA). All reactions were carried out using Bio-Rad Sybr Green Supermix (Bio-Rad). Samples were standardized by mass of coding DNA (cDNA) including 2 ng of cDNA/reaction. Genes examined included *ACAN*, *IL1β*, *IL8*, *IL6*, *MMP13*, *COL2A1*, a disintegrin and metalloproteinase with thrombospondin motifs-1 (*ADAMTS1*), a disintegrin and metalloproteinase with thrombospondin motifs-1 (*ADAMTS5*), cartilage oligomeric matrix protein (*COMP*), glyceraldehyde 3-phosphate dehydrogenase (*GAPDH*), and *P21* (Table 1). Primer sets were obtained from published reports (Table 2). Quality control of qPCR experiments was performed to ensure specificity and validity of the primers for target genes. Assessment of genes was

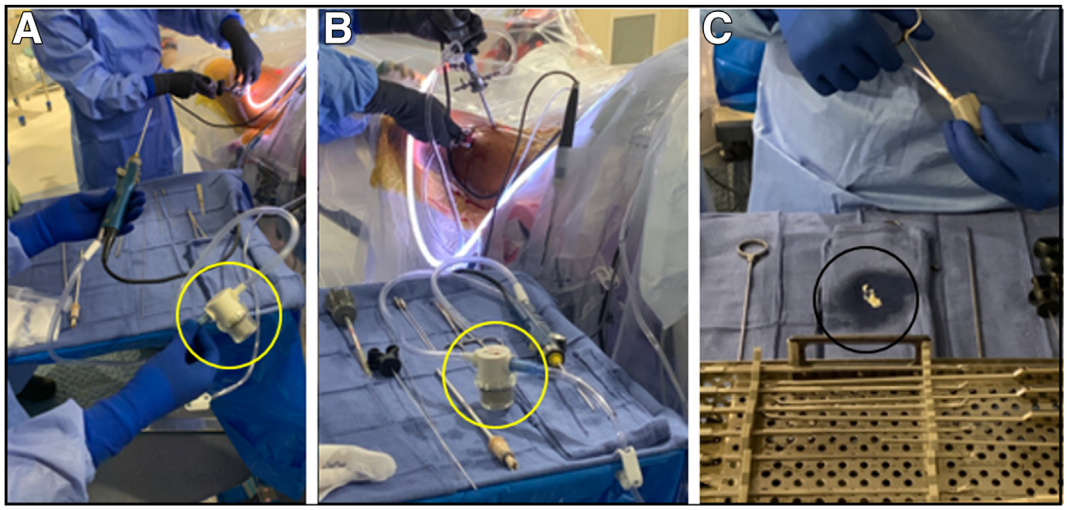

Fig 2. (A) and (B) show the intraoperative orientation of the filter device (circled in yellow) used to collect tissue debrided from the central compartment of the hip joint. (C) The filter device was detached after labral repair and necessary cartilage debridement were complete, and samples (circled in black) immediately transported to the laboratory via appropriate storage and transport protocols.

Table 2. Biomarkers and Gene Primers

Biomarker	Forward (5′-3′)	Reverse (5′-3′)
ACAN[22]	GTGCCTATCAGGACAAGGTCT	GATGCCTTTCACCACGACTTC
COL2A1[23]	GTGTCAGGGCCAGGATGT	TCCCAGTGTCACAGACACAGAT
COMP[24]	AACACGGTCACGGATGACGACTATG	CACAGAGCGTTCCGCAGCTGTT
MMP13[23]	TTTCCTCCTGGGCCAAAT	GCAACAAGAAACAAGTTGTAGCC
**ADAMTS1*[25]	GGACAGGTGCAAGCTCATCTG	TCTACAACCTTGGGCTGCAAA
**ADAMTS5*[25]	TATGACAAGTGCGGAGTATG	TTCAGGGCTAAATAGGCAGT
IL1β[26]	GTGCTGAATGTGGACTCAATCC	ACCCTAAGGCAGGCAGTTG
IL6[27]	CCGGGAACGAAAGAGAAGCT	GCGCTTGTGGAGAAGGAGTT
IL8[26]	GAGGGTTGTGGAGAAGTTTTTG	CTGGCATCTTCACTGATTCTTG
P21[27]	GACACCACTGGAGGGTGACT	CAGGTCCACATGGTCTTCCT
GAPDH[28]	GGAGCGAGATCCCTCCAAAAT	GGCTGTTGTCATACTTCTCATGG

ACAN, aggrecan; ADAMTS1, a disintegrin and metalloproteinase with thrombospondin motifs-1; ADAMTS5, a disintegrin and metalloproteinase with thrombospondin motifs-5; COL2A1, collagen type II; COMP, cartilage oligomeric matrix protein; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; IL1β, interleukin-1β; IL6, interleukin-6; IL8, interleukin-8; MMP13, matrix metalloproteinase-13.

performed by including negative controls in each qPCR experiment examining PCR melt-curves to ensure specificity and verifying the primer efficiency was within 90% to 110% on the basis of the standard curve. Negative controls validated that there was no contamination in the study. Negative controls were run using only PCR primers, polymerase, DNA binding dye, nucleotides, and buffer without DNA. PCR melting curves were performed by increasing temperature and measuring activity of the DNA binding dye. Identification of a single peak in the melting curves was used to validate the specificity of the assay without presence of gene amplification that was not specific to the chosen primers.

Statistical Analysis

qPCR data were normalized to the housekeeping gene, *GAPDH*, and data expressed as relative expression using the ΔCT method. Pairwise comparisons between male and female patients, <20-year-old female versus <20-year-old male patients, 21- to 40-year-old female versus 21- to 40-year-old male patients, and >40-year-old female versus >40-year-old male patients were performed using Student *t* tests. To compare differences between groups incorporating both age and sex, one-way analysis of variance with Tukey honest significant difference was included for multiple comparisons between groups.

Results

Samples were obtained from a total of 91 patients 15 to 58 years old undergoing hip arthroscopy for the diagnosis of FAIS (Table 3). Of the included patients, 54 (59.3%) were female with a mean age of 31.4 (± 1.46) years. A total of 37 (40.7%) males were included, with a mean age of 31.3 (± 1.85) years.

qPCR results indicated that the relative expression of the inflammatory cytokines *IL6* ($P < .05$, 95% confidence interval [CI] 0.047-0.083 female [F], 0.070-0.12 male [M]) and *IL8* ($P = .04$, 95% CI 0.059-0.10 F, 0.082-0.18 M) was significantly greater in male subjects compared with female subjects regardless of age (Fig 3 A and B, Table 4). *IL1β* was greater in male than female subjects ($P = .09$) (Fig 3C, Table 4). Pairwise comparisons of specific age groups indicated that *IL1β* ($P < .01$ 95% CI 0.013-0.063 F, 0.073-0.25 M) and *IL6* ($P = .02$, 95% CI 0.026-0.070 F, 0.067-0.17 M), but not *IL8*, were significantly greater in male compared with female subjects younger than 20 years old (Fig 3 D-F, Table 4). The cell senescent factor, *P21*, was not significantly different between sexes (Fig 3G, Table 4). However, *P21* expression was greater ($P = .06$) in male versus female patients younger than the age of 20 years (Fig 3H, Table 4). The anabolic factors *ACAN*, *COL2A1*, and *COMP* also were tested (Fig 4 A-F, Table 4). No significant age or sex differences were noted, but *COL2A1* was greater ($P = .07$) in >40-year-old male

Table 3. Demographics of Included Patients

	Female		Male		Total	
	Mean age, yr ± SE	N	Mean age, yr ± SE	N	Mean age, yr ± SE	N
<20 yr	16.9 (± 0.46)	11	16.8 (± 0.75)	8	16.8 (± 0.40)	19
21-40 yr	31.6 (± 1.22)	32	30.3 (± 1.12)	20	31.1 (± 0.86)	52
>40 yr	45.3 (± 1.22)	11	46.6 (± 1.80)	9	45.9 (± 1.06)	20
Total	31.4 (± 1.46)	54	31.3 (± 1.85)	37	31.3 (± 1.14)	91

SE, standard error.

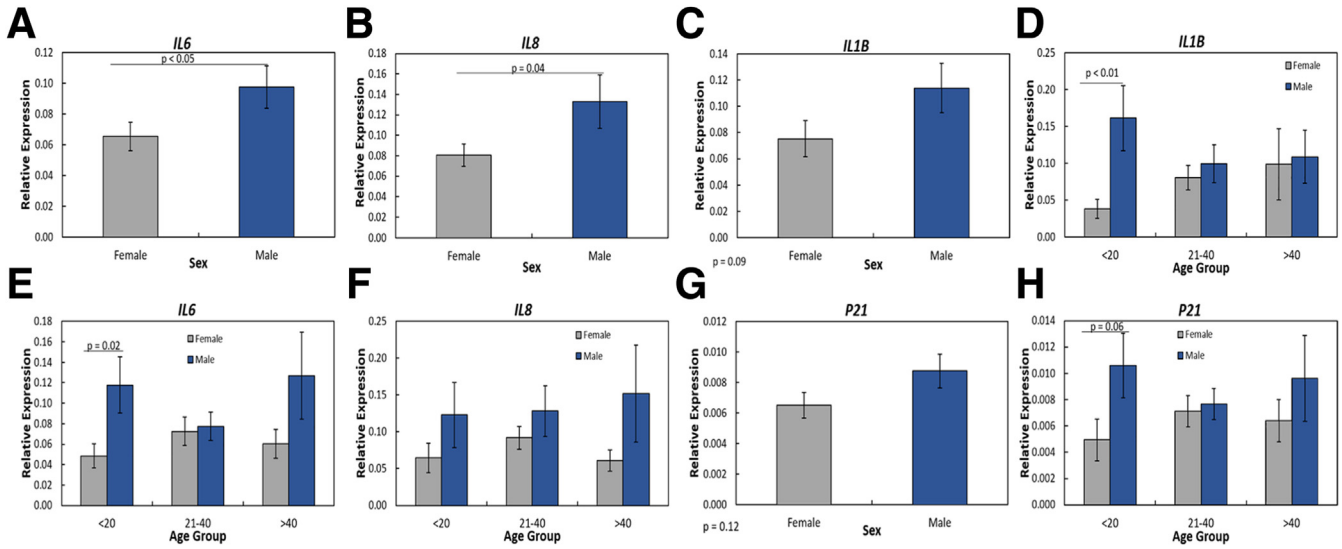

Fig 3. Inflammatory biomarker results comparing sexes and age groups in a cohort of patients with femoroacetabular impingement syndrome (FAIS). Quantitative polymerase chain reaction results of inflammatory and cell senescent factors of tissue remnants obtained during hip arthroscopy to correct for FAIS. Results indicate that *IL6* (A) and *IL8* (B) expression levels were significantly greater in male than female subjects. No significance was noted in *IL1β* (C). Analysis of specific age groups further indicated that expression levels of *IL1β* (D) and *IL6* (E) were greater in male patients <20 years old compared with <20-year-old female subjects. (F) No differences between age groups were noted with *IL8* expression. (G and H) The cell senescent factor *P21* was not significantly different between sexes. (H) Although male subjects <20 years old expressed greater levels of *P21* compared with female subjects <20 years old, only a trend was noted. There were no significant differences for data grouped by age and not sex. Data are expressed as mean ± standard error of the mean. Data are considered significant if $P \leq .05$. (*IL1β*, interleukin-1β; *IL6*, interleukin-6; *IL8*, interleukin-8.)

patients compared with >40-year-old female patients (Fig 4D; Table 4). Testing of catabolic factors *MMP13*, *ADAMTS1*, and *ADAMTS5* indicated that although no significant differences in sex were noted, there was a significant increase in *MMP13* ($P = .047$, 95% CI 0.0051-0.017 F, 0.0084-0.052 M) by <20-year-old male patients compared with <20-year-old female patients (Fig 4 G-L; Table 4). There were no significant differences for data grouped by age and not sex by one-way analysis of variance beyond those aforementioned results.

Discussion

The most important finding of this study was that the intra-articular inflammatory factors *IL6* and *IL8* differed in male and female subjects with FAIS. FAIS alters hip contact mechanics and, hence, provides a macroscopic explanation and premise for subsequent hip pain and eventual OA.[29] However, a corresponding knowledge of the development of OA from FAIS-induced pathology at a molecular level has yet to be fully elucidated. An improved understanding of the molecular environment of the FAIS hip within a varying patient population (i.e., age and sex) can facilitate improved, more targeted, surgical and nonsurgical treatments in the future. Therefore, the goal of this study was to compare molecular differences between male and female patients of varying age groups in tissue remnants obtained during hip arthroscopy for FAIS.

The inflammatory factors *IL6* and *IL8* were significantly greater in male patients compared with female patients with FAIS. *IL6* and *IL8* have been associated with OA.[30] Serum and synovial fluid *IL6* has been associated with incidence and severity of OA in humans.[31] Similarly, *IL8* has been shown to be increased in patients with OA compared with control patients with osteoporosis.[32] *IL6* increases matrix degrading enzymes and suppresses matrix producing genes.[33] Similarly, *IL8* promotes the production of chondral matrix metalloproteinases and enhances leukocyte migration into the joint.[32]

Although these inflammatory cytokines have been less studied in hip OA, preclinical and clinical studies in knee OA could share some parallels in OA pathogenesis with FAIS. In a mouse knee model of post-traumatic OA, loss of *IL6* in male mice suppresses cartilage degradation and reduces pain, but neither of these effects are recapitulated in female mice.[33] These data imply that *IL6* has more specific effects on cartilage loss and pain in male but not in female subjects.[33] Sex-specific differences in *IL6* may relate to estrogen and pain receptors. Estrogen and its receptor have been shown to negatively regulate *IL6* production in vitro.[34,35] In knee OA, several studies have shown

Table 4. qPCR Results of the Femoroacetabular Impingement Syndrome Samples Compared Between Age and Sex Groups

		Overall Male vs Female		<20 Years Old, Male vs Female		21-40 Years Old, Male vs Female		>40 Years Old, Male vs Female	
Gene	Sex	Relative Expression (± SEM)	*P* Value	Relative Expression (± SEM)	*P* Value	Relative Expression (± SEM)	*P* Value	Relative Expression (± SEM)	*P* Value
IL6	Female	0.065 (± 0.009)	<.05*	0.048 (± 0.012)	.02*	0.072 (± 0.014)	.81	0.060 (± 0.014)	.12
	Male	0.097 (± 0.014)		0.118 (± 0.027)		0.077 (± 0.014)		0.127 (± 0.043)	
IL8	Female	0.081 (± 0.011)	.04*	0.064 (± 0.020)	.19	0.092 (± 0.016)	.28	0.061 (± 0.014)	.18
	Male	0.133 (± 0.026)		0.123 (± 0.044)		0.128 (± 0.034)		0.152 (± 0.066)	
IL1B	Female	0.075 (± 0.014)	.09†	0.038 (± 0.013)	<.01*	0.081 (± 0.017)	.52	0.099 (± 0.048)	.87
	Male	0.114 (± 0.019)		0.161 (± 0.045)		0.100 (± 0.026)		0.109 (± 0.036)	
P21	Female	0.007 (± 0.001)	.11	0.005 (± 0.002)	.06†	0.007 (± 0.001)	.77	0.006 (± 0.002)	.37
	Male	0.009 (± 0.001)		0.011 (± 0.002)		0.008 (± 0.001)		0.010 (± 0.003)	
ACAN	Female	0.081 (± 0.011)	.55	0.067 (± 0.021)	.28	0.087 (± 0.016)	.57	0.075 (± 0.014)	.21
	Male	0.091 (± 0.012)		0.103 (± 0.023)		0.074 (± 0.013)		0.119 (± 0.033)	
COL2A1	Female	0.001 (± 0.0001)	.16	0.001 (± 0.0002)	.79	0.001 (± 0.0002)	.93	0.001 (± 0.0001)	.07†
	Male	0.001 (± 0.0003)		0.001 (± 0.0001)		0.001 (± 0.0002)		0.003 (± 0.0011)	
COMP	Female	0.151 (± 0.043)	.50	0.090 (± 0.039)	.10	0.169 (± 0.066)	.82	0.154 (± 0.061)	.22
	Male	0.200 (± 0.062)		0.379 (± 0.203)		0.194 (± 0.080)		0.060 (± 0.029)	
MMP13	Female	0.016 (± 0.003)	.24	0.011 (± 0.003)	.047*	0.018 (± 0.004)	.89	0.015 (± 0.006)	.78
	Male	0.021 (± 0.003)		0.030 (± 0.011)		0.018 (± 0.004)		0.018 (± 0.006)	
ADAMTS1	Female	0.212 (± 0.030)	.25	0.194 (± 0.058)	.10	0.243 (± 0.045)	.96	0.131 (± 0.027)	.20
	Male	0.265 (± 0.034)		0.350 (± 0.063)		0.246 (± 0.043)		0.243 (± 0.083)	
ADAMTS5	Female	0.012 (± 0.004)	.57	0.002 (± 0.001)	.08†	0.018 (± 0.006)	.41	0.005 (± 0.001)	.35
	Male	0.016 (± 0.007)		0.046 (± 0.030)		0.010 (± 0.004)		0.003 (± 0.001)	

ACAN, aggrecan; *ADAMTS1*, a disintegrin and metalloproteinase with thrombospondin motifs-1; *ADAMTS5*, a disintegrin and metalloproteinase with thrombospondin motifs-5; *COL2A1*, collagen type II; *COMP*, cartilage oligomeric matrix protein; *IL1β*, interleukin-1β; *IL6*, interleukin-6; *IL8*, interleukin-8; *MMP13*, matrix metalloproteinase-13; qPCR, quantitative polymerase chain reaction; SEM, standard error of the mean.

*$P \leq .05$.

†$P < .1$.

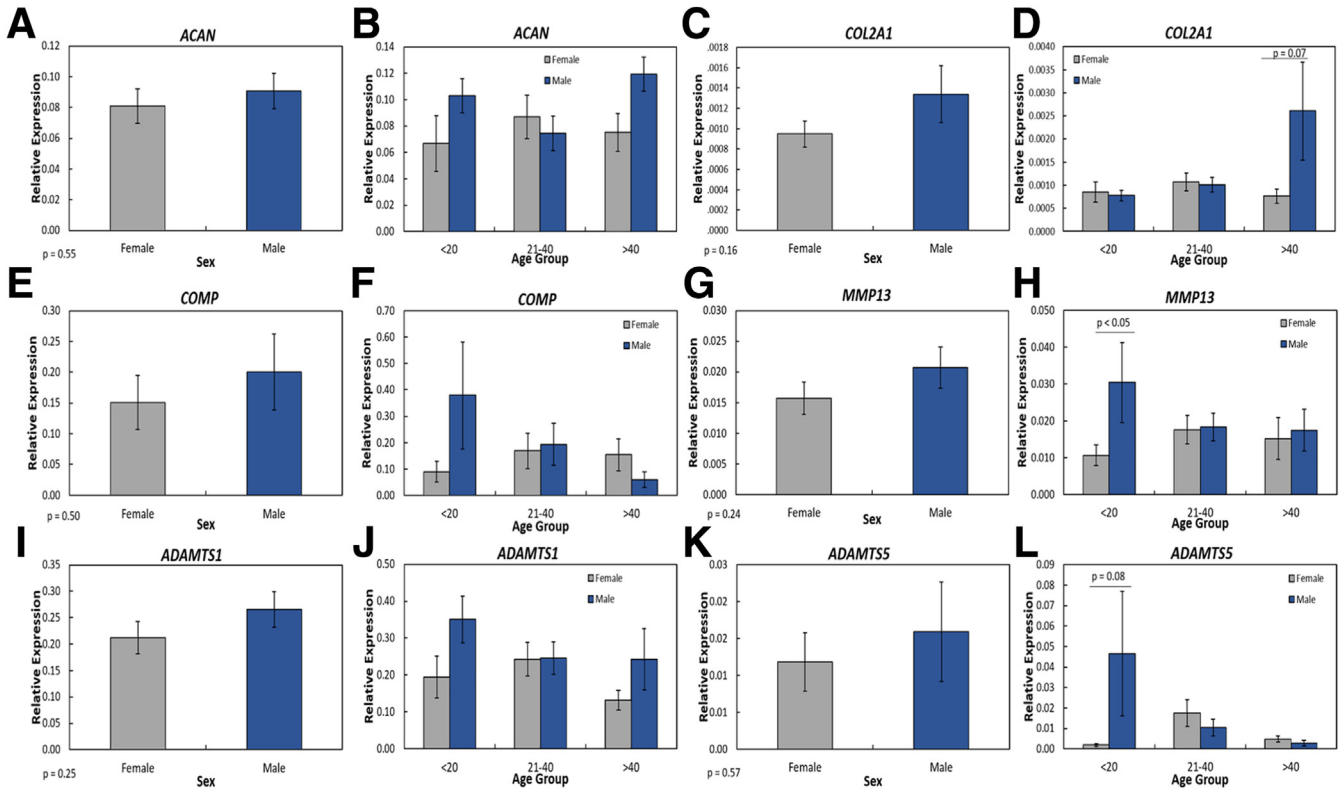

Fig 4. Anabolic and catabolic biomarker results compared between sexes and age groups in a cohort of patients with femoroacetabular impingement syndrome (FAIS). Quantitative polymerase chain reaction results of anabolic and catabolic factors expressed in tissue remnants obtained during hip arthroscopy to correct for FAIS. Results indicate that *ACAN* (A and B), *COL2A1* (C and D), and *COMP* (E and F) were not significantly different between samples. *COL2A1* was greater in >40-year-old male patients compared with age-matched female patients ($P = .07$). (G-L) No differences were noted with the catabolic factors, *MMP13*, *ADAMTS1*, or *ADAMSTS5*, although an increase in *ADAMTS5* was noted between male subjects <20 years of age compared with their aged female counterparts. There were no significant differences for data grouped by age and not sex. Data are expressed as mean ± standard error of the mean. Data are considered significant if $P \leq .05$. (*ACAN*, aggrecan; *ADAMTS1*, a disintegrin and metalloproteinase with thrombospondin motifs-1; *ADAMTS5*, a disintegrin and metalloproteinase with thrombospondin motifs-5; *COL2A1*, collagen type II; *COMP*, cartilage oligomeric matrix protein; *MMP13*, matrix metalloproteinase-13.)

that older male and female patients do not differ in IL6 serum levels; however, increased *IL6* is associated with worse pain in female patients.[32,36,37] Conversely, male patients with knee OA have greater levels of IL8, which is associated with increased pain but is less associated with pain in female patients.[37,38]

Further investigation of sex differences in inflammatory factors is needed to better define the pathogenesis of hip OA after FAIS. Although inflammatory and catabolic factors in the present study were found to be greater in male than female subjects, meta-analysis of FAIS outcomes demonstrate that female subjects have worse postoperative outcomes.[39] These data may be skewed in part by age, as some previous publications have found that female patients older 45 years of age have worse postoperative outcomes than male patients of similar age and younger patients of both sexes.[40,41] Changes in sex hormones with age may have differential effects in male and female subjects and may be a focus of future investigation.[16]

The present study examined *ACAN*, *COMP*, *ADAMTS1*, *ADAMTS5*, *IL6*, *IL8*, *IL1β*, *MMP13*, *P21*, and *COL2* (Table 1). We chose to focus on these biomarkers, as they are indicative of greater levels of inflammation, cell senescence, and/or anabolic or catabolic factors with known implications in OA. Hashimoto et al.[21] examined similar biomarkers and found that articular cartilage from the impingement zone of affected hips with FAIS expressed greater levels of *IL8*, *ACAN*, and *ADAMTS4*. Haneda et al.[42] in their study of 45 hips, with 7 control hips found that those with cam FAIS and advanced OA secondary to FAIS expressed greater levels of *IL1β* and *COL2* compared with control hips. Similarly, we found that inflammatory cytokines including *IL1β* are upregulated in younger males with FAIS. Comparison with sexes and inclusion of control hips would further

validate these findings. Unlike the previous reports, our study examined the damaged tissue from the acetabular zone of impaction (chondrolabral junction and labrum as well as the overlying synovitis) rather than the area of femoral head-neck junction impaction, which can often be worn away from impingement and may not yield much tissue for analysis. Tissue from this area of the femoral neck also excludes analysis of torn acetabular articular cartilage or torn labral tissue.

As noted, previous literature has found that articular cartilage obtained at the head-neck junction of hips with FAIS expressed markedly elevated levels of select cytokines and catabolic factors, compared with control hips.[29,42] Our results expand upon those data further and demonstrate that young males with FAIS have greater levels of *IL1β*, *IL6*, and *MMP13* expressed when compared with young female patients. Given the known differences between male and female patients related to impingement type (cam type more common in males and pincer type more common in female patients), as well as the understanding that larger cam lesions are correlated with greater articular cartilage damage, this correlates to previous clinical evidence regarding sex differences in FAIS.[43,44]

Literature suggests that those with FAIS who undergo arthroscopy within 6 months of symptom onset have better outcomes than those whose symptoms persist for a longer duration prior to surgical intervention.[16,45,46] Better characterization of the sex-specific molecular pathogenesis of OA after FAIS will allow for further refinement of treatment timelines and parameters as well as the development of biomarkers to track treatment severity and recovery.

Limitations

This study is not without limitations. A limitation of this study is the relatively small sample size of 91 total patients when making both age and sex comparisons. This experimental design may have limited the number of significantly different comparisons that would appear if the sample size was larger. However, we found sufficient group consistency in expression patterns to report significant differences in multiple genes. Furthermore, control patients without FAIS were not included in this study, so there is no indication that FAIS hips are more metabolically active. Another limitation is that we did not include clinical and biomechanical characteristics that could have affected the biological milieu of the hip joint, such as boney morphology or magnetic resonance imaging findings. The inclusion of both patients who did or did not have a steroid injection will have a substantial effect on outcomes, especially inflammatory cytokine levels. Finally, data collection was limited by the parameters of our institutional review board approval, so some important variables were not included in this study.

Conclusions

In patients with FAIS, there are significant differences between male and female patients in the biomarkers present in the affected hip at the time of surgery. Male patients have greater levels of *IL6* and *IL8* and male patients younger than 20 years of age have greater levels of *IL1β*, *IL6*, and *MMP13* compared with age-matched female patients.

Disclosures

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