Collagen Biomarker Response to Acute Joint Injury in a Non-terminal Animal Model of Osteoarthritis

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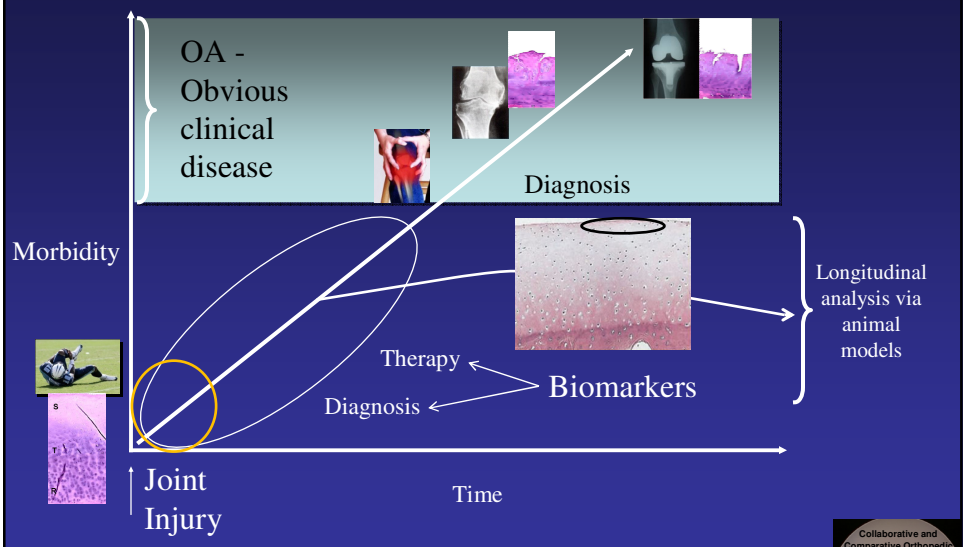
Disclosure

- The authors have nothing to disclose

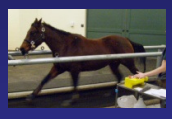
Post-traumatic osteoarthritis development



Adapted from Lohmander LS, et al. 2004

Post-traumatic osteoarthritis (PTOA)

- Translational models of PTOA
  - Biomechanical instability often worse than biochemical
  - Disease often advanced
- Current models are terminal








- Horse as a translational model
  - Athletes, naturally occurring PTOA
  - Can control exercise
  - Large size facilitates sample collection without euthanasia
    - Can easily collect and examine SF (>3 mL)

- Goal: Develop a model of early PTOA and determine whether biomarkers could be used to identify early subtle biochemical changes in the joint after injury and identify the onset and progression PTOA.

- Central Hypothesis: Biochemical changes resulting from an acute osteochondral (OC) injury will reflect the onset and progression of early PTOA.

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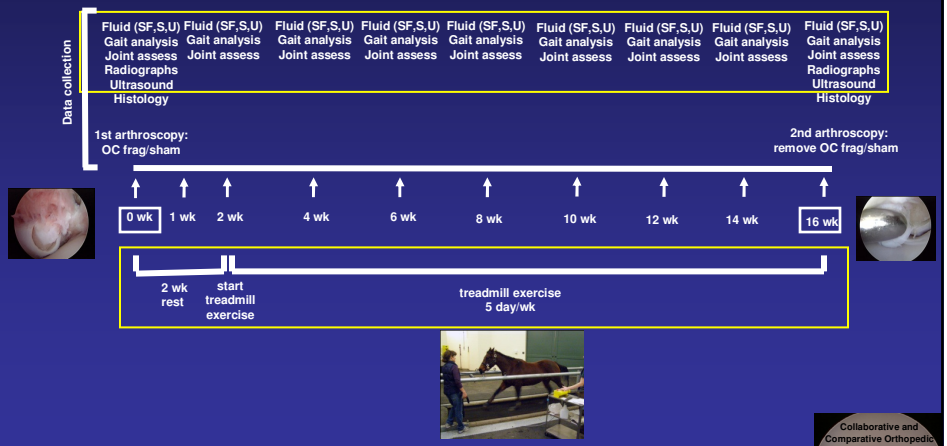
Study design

Horses

Age, breed, gender matched; clinically, radiographically normal

11 experimental: Osteochondral (OC) fragment created in 1 metacarpophalangeal (MCP) joint
- sham operation in contralateral joint

11 exercise control



Fluids collected between 9-11 am; SF collected without lavage

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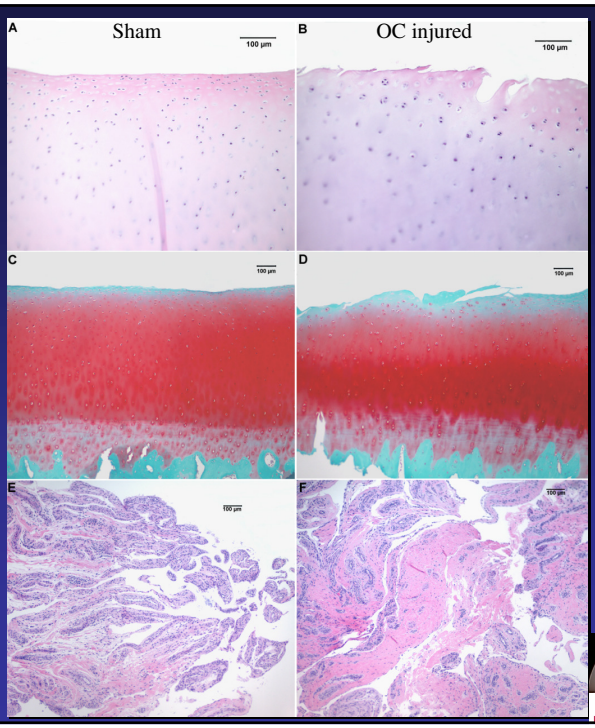
Histology

Cartilage superficial layers (H&E)

Cartilage full thickness (Safranin-O)

Synovial membrane (H&E)



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Histology: Cartilage

Poster P-17

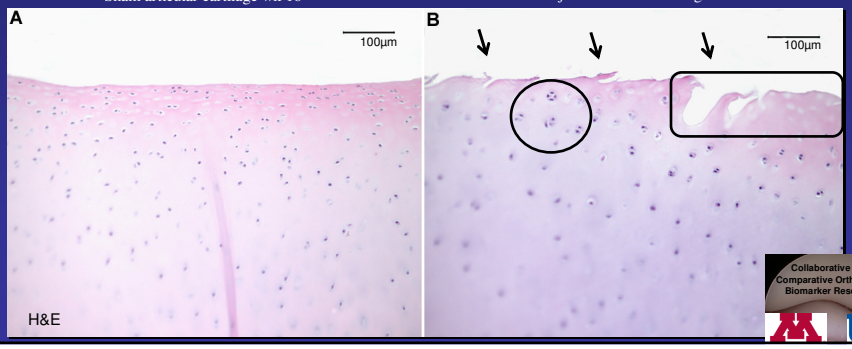
Summary of **cartilage** histologic data obtained from OC injured and sham MCP joints.

	Week 16 (mean ±SD)	
	OC injured (11/11)	Sham (6/11)
Fibrillation/fissuring (0-4)	1.36 ±1.29*	0.33 ±0.52
Cluster formation (number/area)	8.14 ±6.01**	1.33 ±2.11
Chondrocyte death (% area)	6.0 ±2.0**	2.0 ±1.0
Decreased matrix PG (% area)	15.0 ±12.0	14.0 ±12.0

*represents a significant increase in OC injured values compared to sham values at week 16 (*P<0.05, **P<0.01, ***P<0.001).

Sham articular cartilage wk 16

OC injured articular cartilage wk 16



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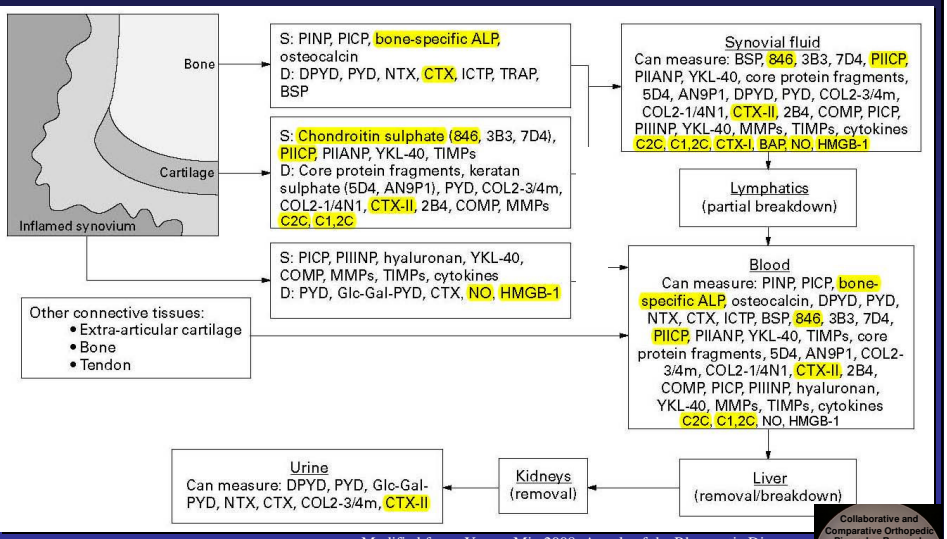
OA biomarkers examined



Modified from: Young-Min 2008, Annals of the Rheumatic Diseases

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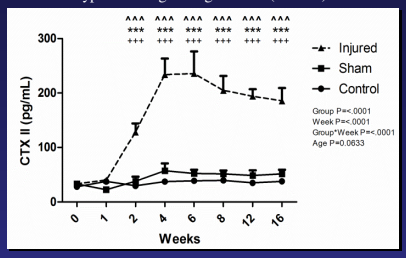
SF biomarkers – Cartilage degradation

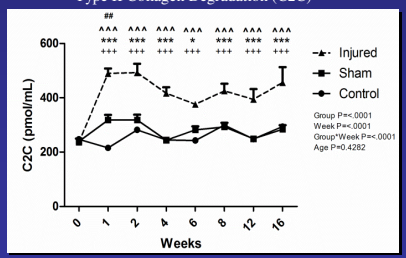
\+ = OC injured to baseline
* = OC injured to sham
^ = OC injured to control

Type II Collagen Degradation (CTX II)

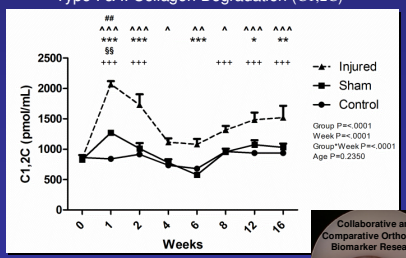


- Clear differentiation of injured joints
- Injury caused cartilage damage
- Minimal response to arthroscopy and arthrocentesis
- Biphasic response
- Triple helix proteolysis (C2C & C12C) before cross-linked telopeptide breakdown (CTX II)

Type II Collagen Degradation (C2C)



Type I & II Collagen Degradation (C1,2C)



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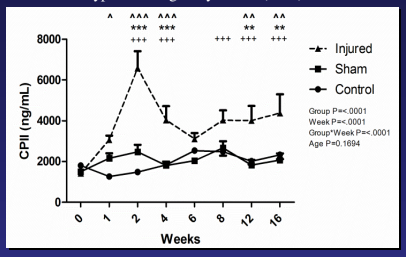
SF biomarkers – Cartilage synthesis

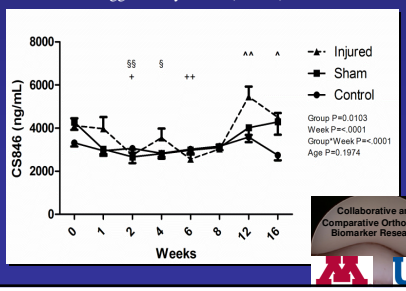
+ = OC injured to baseline
* = OC injured to sham
^ = OC injured to control

- Immediate collagen synthesis response
- Biphasic
- No proteoglycan repair in acute phase

Type II Collagen Synthesis (CPII)



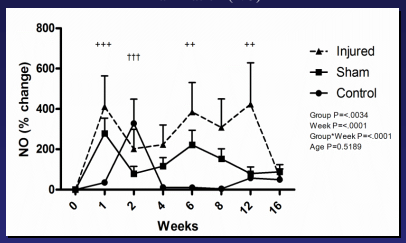
Aggrecan Synthesis (CS846)



SF biomarkers - Inflammation

+ = OC injured to baseline
* = OC injured to sham
^ = OC injured to control

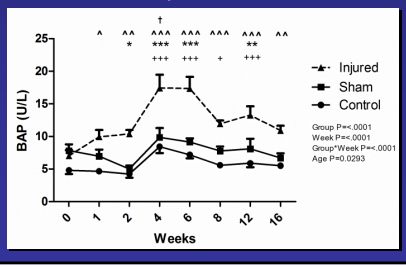
Inflammation (NO)



- Nitric oxide
  - Spike immediately after injury, weeks 6 and 12
  - Affect of arthroscopy
  - Affect of arthrocentesis

SF biomarkers - Bone

Bone Synthesis (BAP)

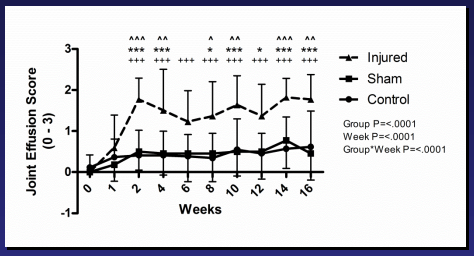


- BAP
  - Delayed response
  - Stimulation of OC fragment healing?

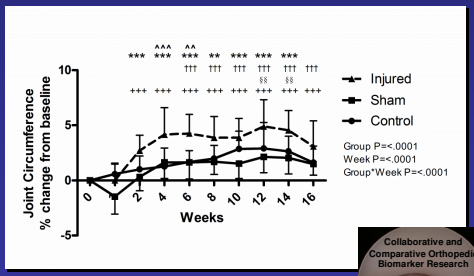
Clinical assessment of the MCP joint

+ = OC injured to baseline
* = OC injured to sham
^ = OC injured to control

- Joint effusion: OC injured joints
  - ↑ from baseline
  - ↑ compared to sham, control joints



- Joint circumference: OC injured
  - ↑ from baseline
  - Variable ↑ compared to sham and control joints



- Decreased ROM in OC injured joints

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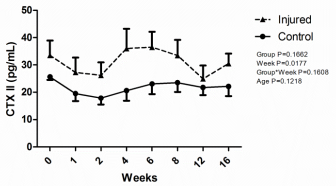
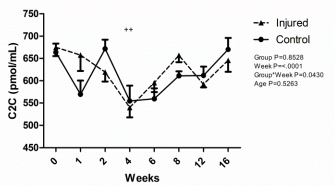
Serum biomarkers

- Changes similar to control horses

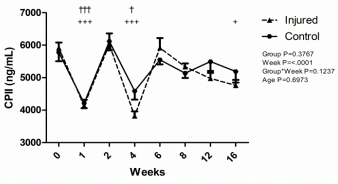
Serum: Type II Collagen Degradation (CTX II)



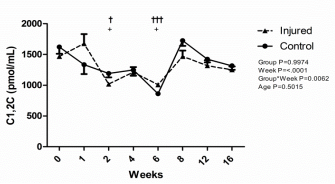
Serum: Type II Collagen Degradation (C2C)



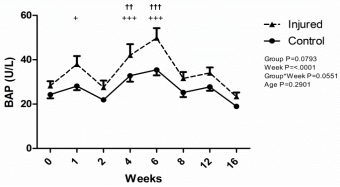
Serum: Type II Collagen Synthesis (CPII)



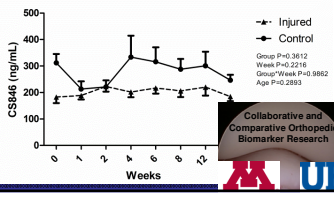
Serum: Type I & II Collagen Degradation (C1,2C)



Serum: Bone Synthesis (BAP)



Serum: Aggrecan Synthesis (CS846)



+ = OC injured to baseline
† = Control to baseline

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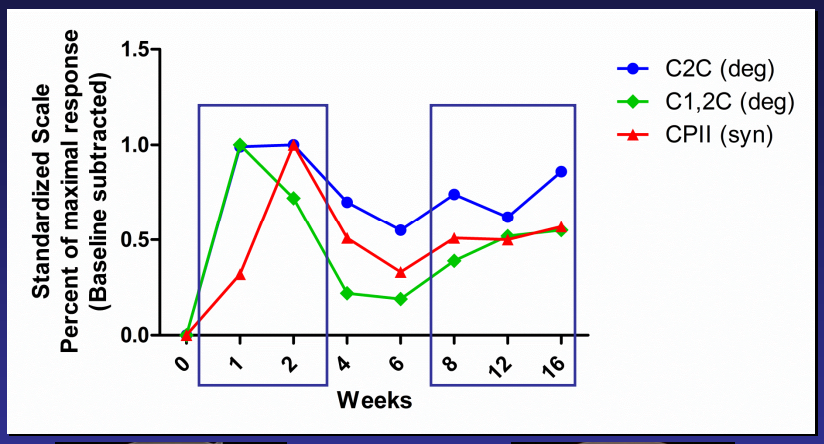
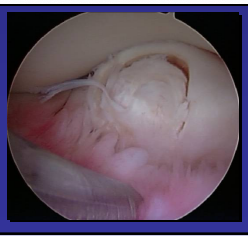
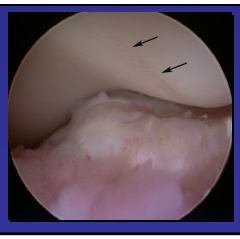
SF biomarkers: Standardized scale (OC injured joints)







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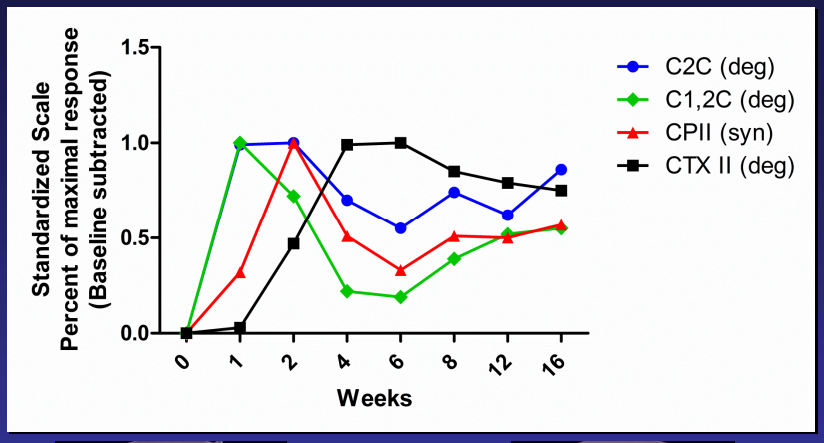

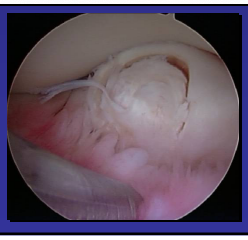

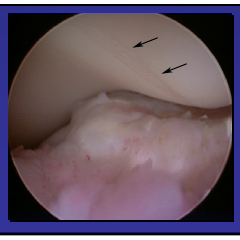

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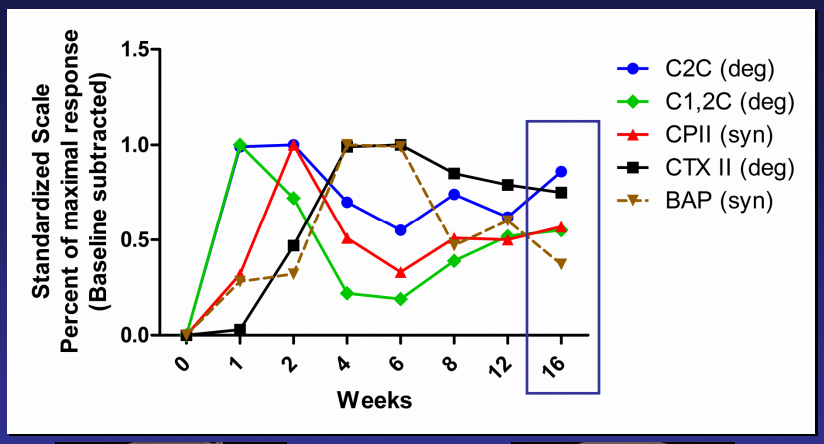
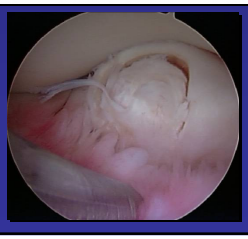
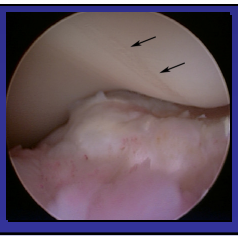
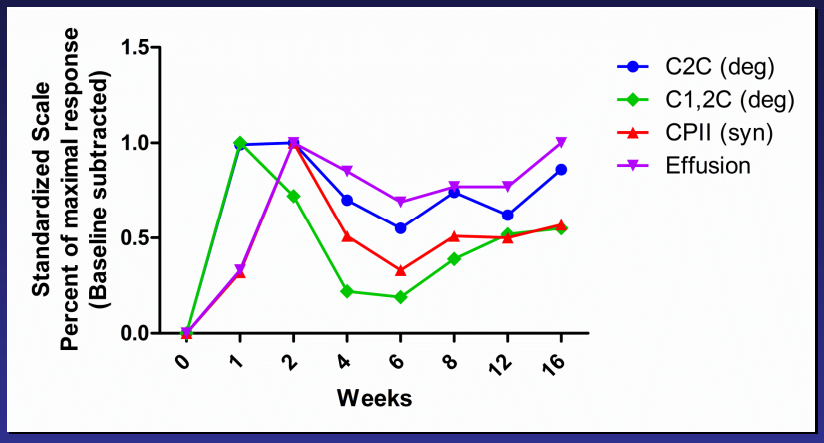
SF biomarkers: Standardized scale (OC injured joints)







SF biomarkers: Standardized scale (OC injured joints)



SF biomarker correlations with PTOA model

Mean (±SD) rho correlations (*P* values) of SF biomarkers with clinical, arthroscopic and histologic scores in OC injured horses at week 16.

	CTX II	C2C	C1,2C	CPII	CTX I	BAP	CS846	NO %change	HMGB-1
Clinical									
Effusion	**0.548 (0.010)**	**0.529 (0.016)**	**0.480 (0.032)**	0.420 (0.065)	-0.147 (0.524)	**0.573 (0.005)**	0.459 (0.056)	0.092 (0.726)	0.025 (0.912)
JC %change	0.148 (0.522)	0.367 (0.112)	**0.582 (0.007)**	**0.550 (0.012)**	**-0.510 (0.018)**	0.162 (0.471)	0.158 (0.531)	-0.272 (0.291)	-0.335 (0.128)
R-Total	**0.696 (<.001)**	**0.625 (0.003)**	**0.475 (0.035)**	0.438 (0.054)	-0.090 (0.700)	**0.603 (0.003)**	0.418 (0.084)	-0.290 (0.258)	0.190 (0.397)
Arthroscopy									
Synovium	**0.490 (0.024)**	**0.585 (0.007)**	**0.500 (0.025)**	**0.593 (0.006)**	-0.245 (0.285)	0.287 (0.195)	0.152 (0.546)	-0.215 (0.406)	0.200 (0.371)
Cartilage	**0.692 (0.001)**	**0.507 (0.022)**	**0.450 (0.046)**	0.428 (0.060)	-0.107 (0.644)	**0.652 (0.001)**	0.322 (0.192)	-0.080 (0.759)	0.054 (0.811)
CDK	**0.786 (<.001)**	**0.581 (0.007)**	**0.489 (0.029)**	**0.493 (0.027)**	0.005 (0.982)	**0.701 (<.001)**	0.198 (0.431)	0.052 (0.843)	0.049 (0.830)
Total	**0.679 (0.001)**	**0.559 (0.010)**	**0.458 (0.042)**	**0.462 (0.040)**	-0.214 (0.353)	**0.588 (0.004)**	0.309 (0.213)	-0.179 (0.491)	0.124 (0.583)
Histology Cartilage									
Fib/fis	**0.503 (0.047)**	0.392 (0.133)	0.271 (0.309)	0.182 (0.501)	-0.031 (0.910)	0.359 (0.157)	**-0.630 (0.016)**	-0.172 (0.592)	-0.142 (0.586)
Clusters	**0.606 (0.013)**	0.088 (0.747)	0.184 (0.494)	0.132 (0.625)	0.168 (0.535)	**0.514 (0.035)**	0.280 (0.332)	0.541 (0.069)	-0.130 (0.619)
Chond. death	**0.601 (0.014)**	**0.561 (0.024)**	**0.589 (0.016)**	**0.531 (0.034)**	-0.117 (0.665)	**0.580 (0.015)**	0.097 (0.739)	0.425 (0.169)	-0.101 (0.699)
PG loss	0.047 (0.862)	0.052 (0.849)	0.340 (0.198)	0.371 (0.157)	-0.266 (0.319)	-0.017 (0.948)	0.152 (0.603)	0.184 (0.568)	**-0.487 (0.047)**
Histology Synovium									
Vascularity	0.331 (0.142)	**0.521 (0.018)**	**0.475 (0.034)**	**0.526 (0.017)**	-0.235 (0.306)	0.406 (0.061)	-0.085 (0.739)	0.191 (0.463)	0.138 (0.539)
Hyperplasia	**0.508 (0.019)**	0.046 (0.847)	-0.115 (0.630)	-0.016 (0.946)	-0.065 (0.780)	0.241 (0.280)	0.027 (0.915)	**-0.566 (0.018)**	0.133 (0.554)
Fibrosis	**0.531 (0.013)**	**0.673 (0.001)**	**0.568 (0.009)**	**0.532 (0.016)**	-0.079 (0.734)	**0.566 (0.006)**	0.034 (0.894)	0.146 (0.576)	0.259 (0.244)

Bolded ρ values represent statistically significant correlations (P<0.05).

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Conclusions: Biomarkers

- SF biomarkers clearly distinguished injured joints from sham and control joints after acute injury
  - No exercise affect
- Biphasic response
  - Initial metabolic degradation and repair response to acute injury
  - Second stage of cartilage degradation/repair started around week 8 presumably indicating onset and progression of OA
- Demonstrated the early phases of progression to PTOA
- Biomarker profile at week 16 clearly reflected the early osteoarthritic lesions produced by the model
- Joint effusion paralleled collagen biomarkers
  - Effusion was the best clinical correlate to what occurred histologically (Poster P-17)
- SF was best fluid for analysis of a single joint after injury
  - No obvious correlation to serum or urine concentrations

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Limitations

- Additional biomarker analysis of inflammation
  - Indirect biomarkers on same samples (multiplex analysis)
    - Cytokines, MMPs, etc.
- Imaging – MRI
- Long-term monitoring
  - Identify progressors

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