

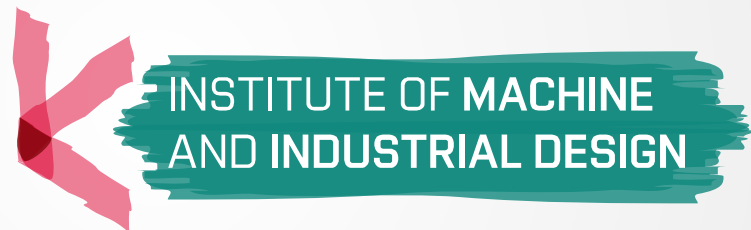
THE EFFECT OF SYNOVIAL FLUID CONSTITUENTS ON FRICTION AND LUBRICATION OF ARTICULAR CARTILAGE

Ing. Pavel Čípek

Supervisor: doc. Ing. Martin Vrbka, Ph.D.

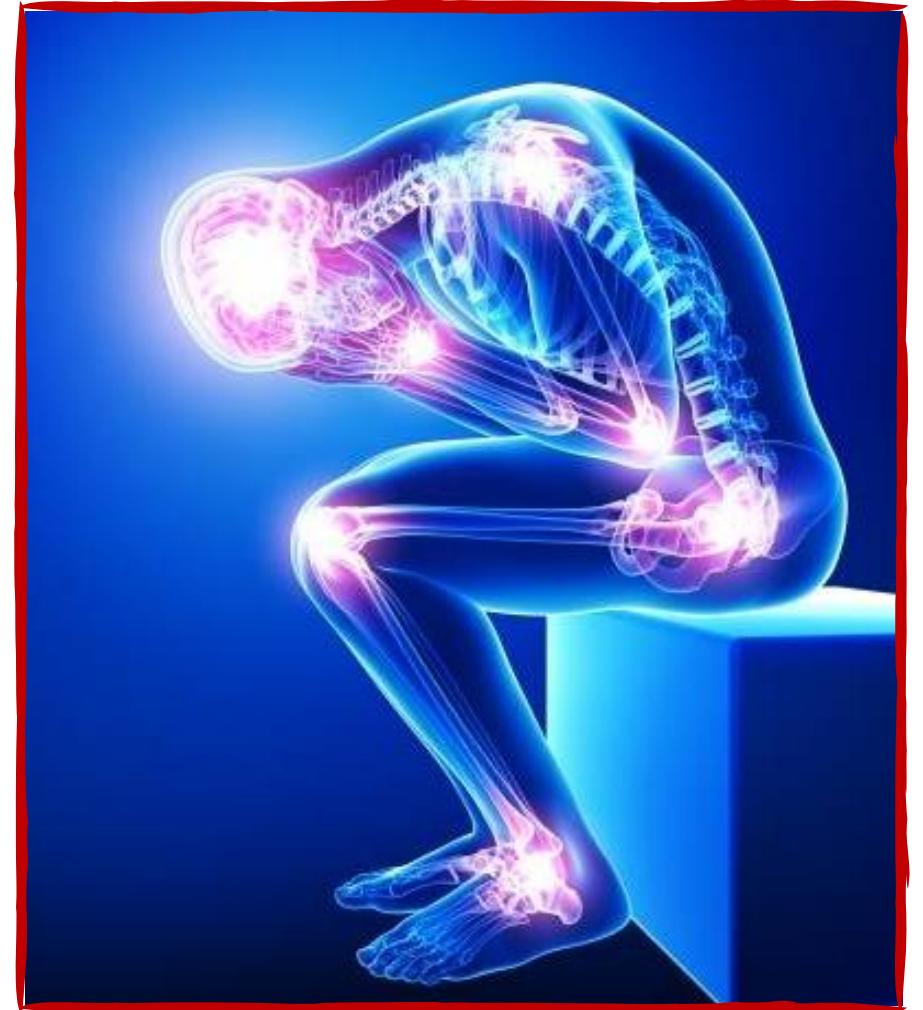
Institute of Machine and Industrial Design
Faculty of Mechanical Engineering
Brno University of Technology

Brno, 31th March 2022



Content of presentation

- Introduction and Motivation
- State of the Art
 - Friction, Lubrication, Visualization
- Summary of Literature Review
- Aim of PhD Thesis
- Scientific Questions, Hypotheses
- Materials and Methods
 - Reciprocating Tribometer and Experimental Methodology
- Results and Discussion
- Conclusions, List of Publications



Osteoarthritis

Wear of cartilage tissue

Distraction of lubrication mechanism

Increasing of friction coefficient

Changes in synovial fluid composition

Reduction of hyaluronic acid concentration



Painful movement

Movement limitations

Decline in living standard

Osteoarthritis

Total endoprosthesis

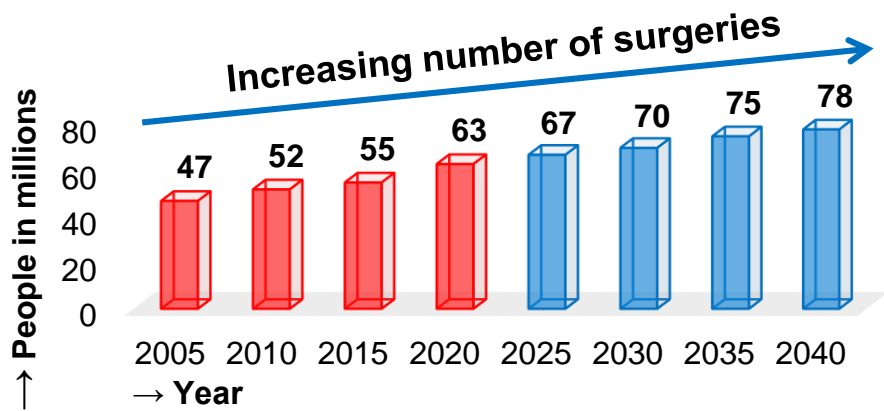


Limited longevity

Revision operations

Wear debris

Aseptic loosening



Osteoarthritis

Total endoprosthesis



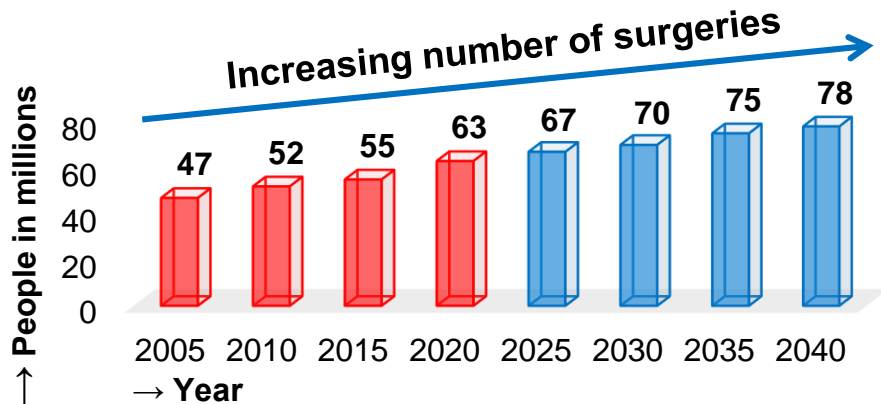
Viscosupplementation



Non-invasive treatment

Intra-articular injection with hyaluronic acid

Increasing number of surgeries



Osteoarthritis

Reduction of inflammation

Friction reduction

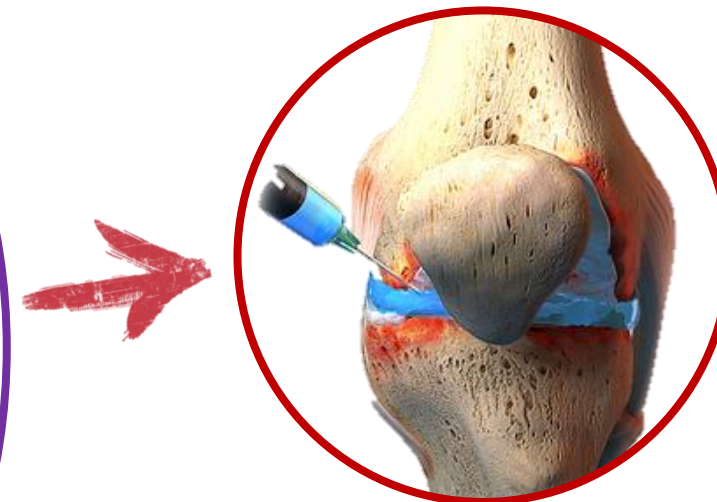
Improved lubrication processes

Hyaluronic acid supplementation

Improving the ability to absorb shocks



Viscosupplementation



Non-invasive treatment

Intra-articular injection with hyaluronic acid

Total endoprosthesis



Viscosupplementation



Tribology of endoprosthesis

- Many publications – well explored area
- ✓ Friction coefficient (CoF)
- ✓ Lubricating film formation

Tribology of synovial cartilage

- Limited number of publications
- Non-verified lubrication theories
- Not completely understood

Tribology of synovial joint

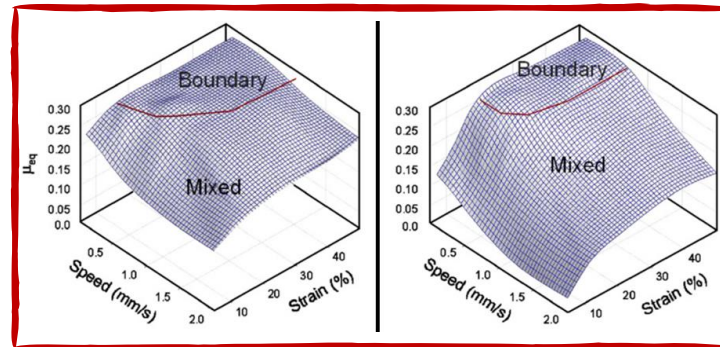
Friction in synovial joint

- Operating conditions impact
- Many publications
- Relatively well described problematics

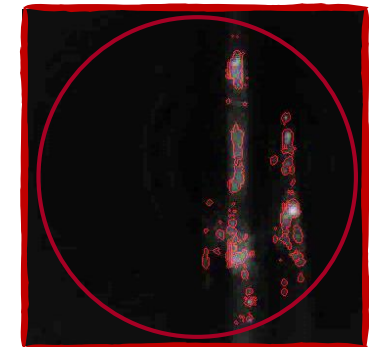


Lubricating processes in synovial joint

- Role of SF components
- Role of cartilage poreous structure
- Lubricating regimes



- Visualization of the cartilage contact
- Fluorescence microscopy



Friction in synovial joint

STACHOWIAK, G.

1994

Friction and wear changes in synovial joints

- Cartilage vs stainless steel
- Lubricant – synovial fluid

MERKHER, Y. et al.

2006

A rational human joint friction test using a human cartilage

- Tests with human cartilage
- Lubricant – synovial fluid

CHAN, S. et al.

2011

The role of lubricant entrapment at biological interfaces ...

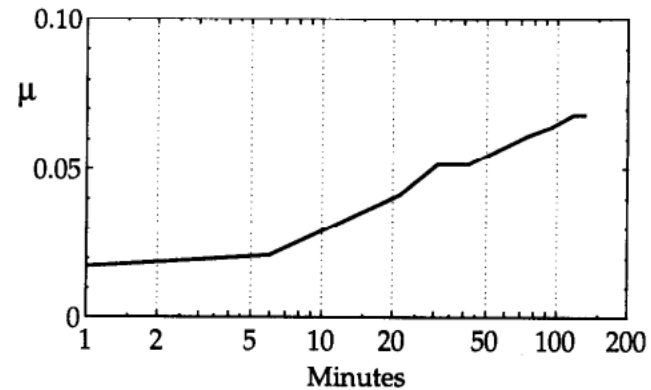
- Cartilage sample from high (M1) / low (M4) contact pressure

MOORE, A. et al.

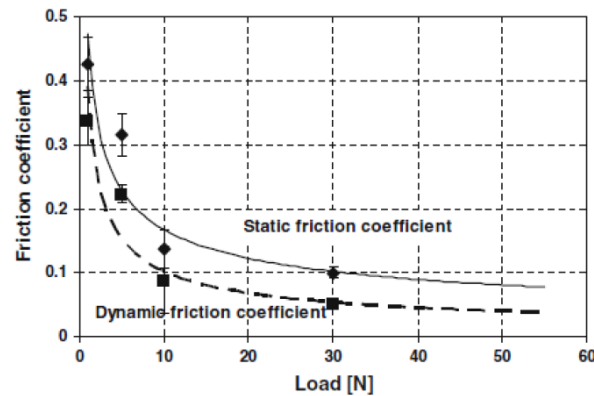
2016

New insights into joint lubrication.

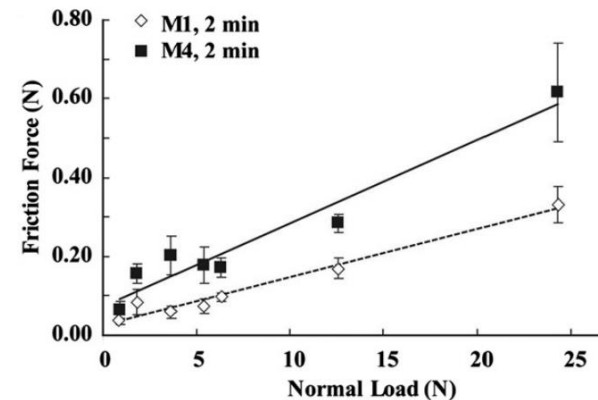
- Rehydration impact
- Cartilage vs glass plate



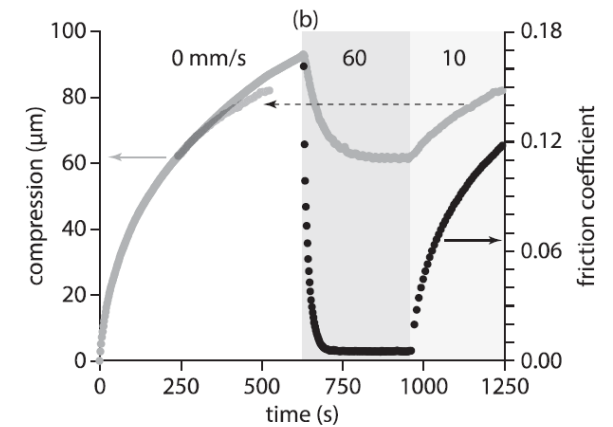
CoF between cartilage and stainless steel.



Dependency between load and CoF



Dependency between load and CoF (M1/M4)



Tribological rehydration and recovery

Lubricating of synovial joint

GREENE, G. et al.

2008

WU, T. et al.

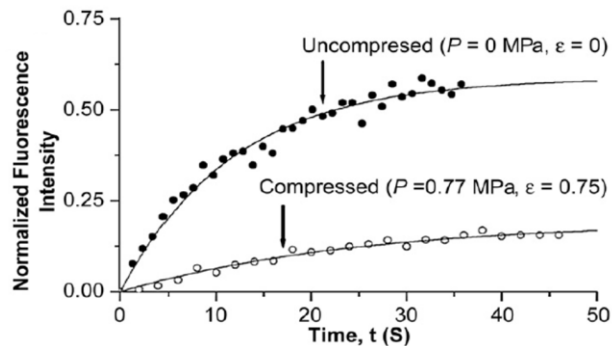
2015

Changes in pore morphology and fluid transport in compressed articular cartilage and the implications ...

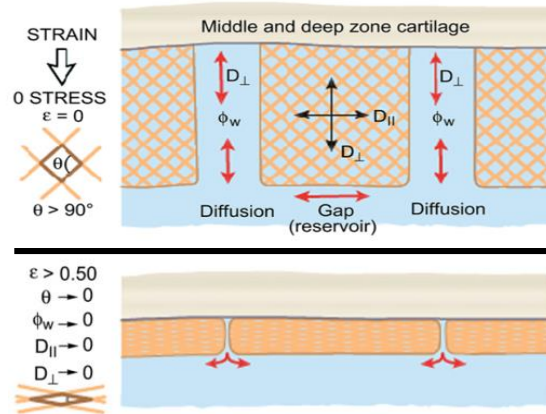
The lubrication effect of hyaluronic acid and chondroitin sulfate on the natural temporomandibular cartilage ...

- Flowing of lubricant through the cartilage porous structure
- Impact of deformation of porous structure to lubricant flowing
- Fluorescence microscopy

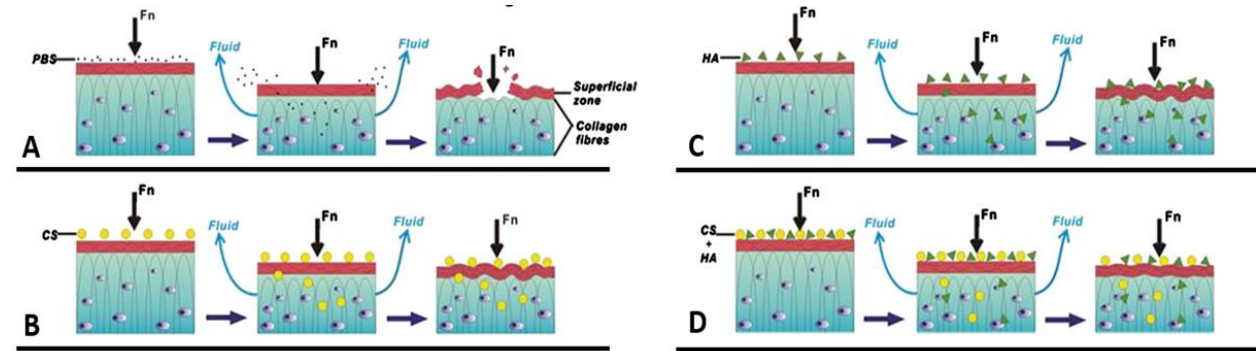
- HA Molecular weight impact
- Higher HA molecular weight = better surface protection
- Bigger particles adsorb on the cartilage surface



Recovery hydration ability of cartilage



Compression of porous structure



Physical lubrication mechanism model

Lubricating of synovial joint

JAHN, S. et al.

2016

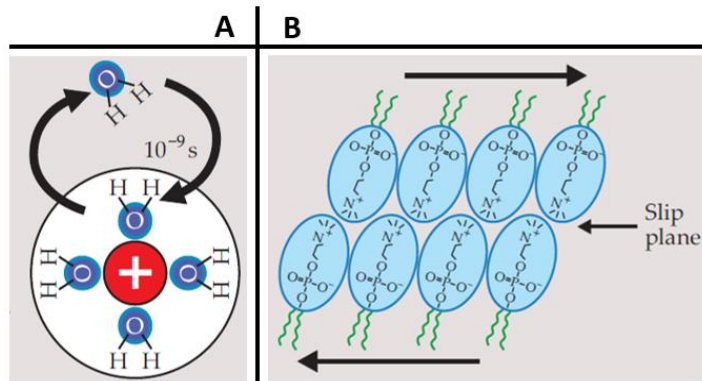
MURAKAMI, T. et al.

2017

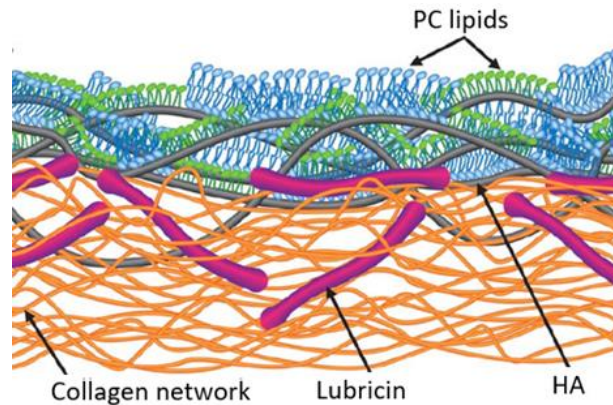
Lubrication of Articular Cartilage

Importance of adaptive multimode lubrication mechanism ...

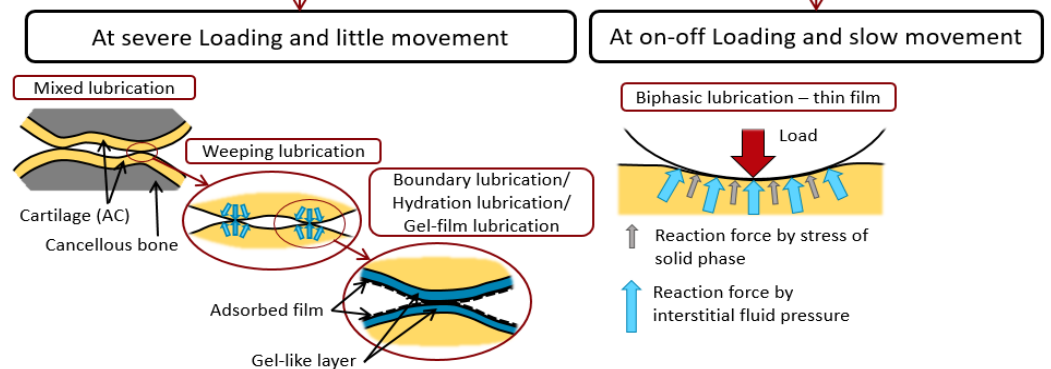
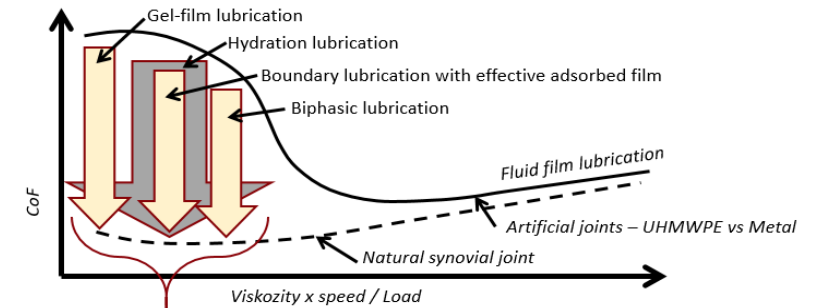
- Principle of hydration lubrication
- Mica plates – similar hydrophilicity as cartilage
- HA creates layer on the mica plate



Hydration shell – the principle of hydration lubrication



Phospholipidic bilayer



Adaptive multimode mechanism

Visualization of the cartilage contact

NAKASHIMA, K. et al.

2006

YARIMITSU, S. et al.

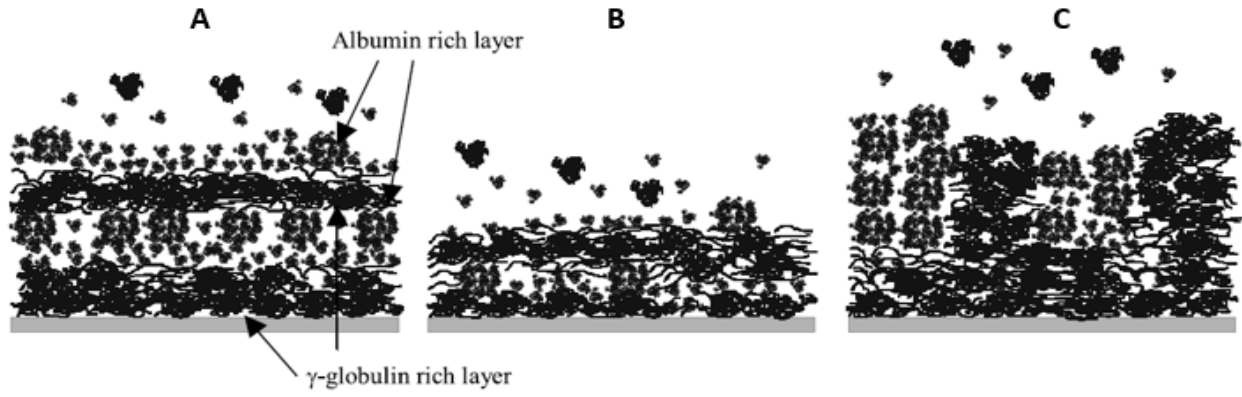
2008

Study on wear reduction mechanisms of artificial cartilage by synergistic protein boundary film formation

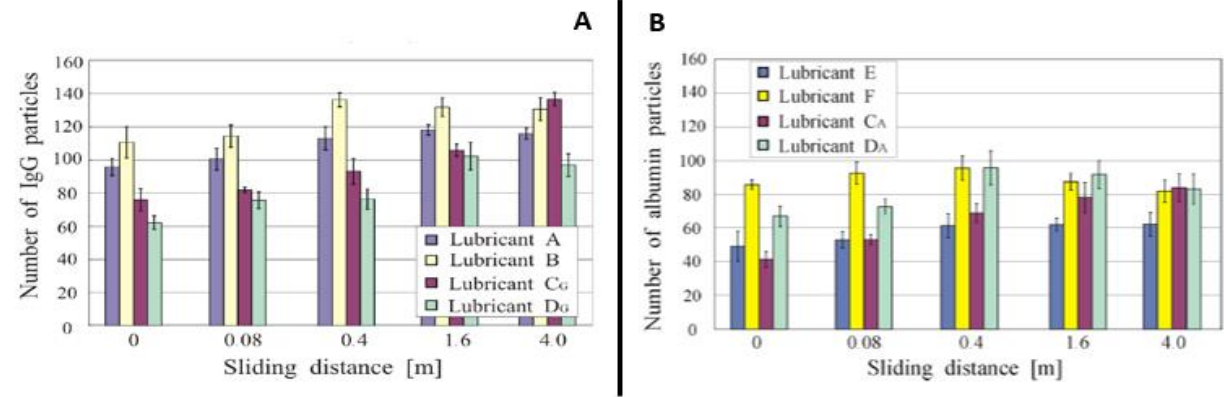
Effects of Lubricant Composition on Adsorption Behavior of Proteins on Rubbing Surface and Stability of Protein ...

- PVA hydrogel contact visualization by fluorescence microscopy
- Fluorescence analysis after performing the experiment

- PVA hydrogel contact visualization by fluorescence microscopy
- More complex lubricants show larger number of particles



Model of lubricating film formation.



Number of particles during friction reciprocating test.

Tribology of synovial joint

Friction in synovial joint

- Influence of operating conditions, lubricant composition, rehydration, contact pairs
- Many publications

✓ Comprehensive description of cartilage friction behaviour

Lubricating processes in synovial joint

- Limited number of studies
- Unproven theories
- Insufficiently described lubrication processes in cartilage contact

- Only post analysis of lubricating film
- Very limited number of studies – focused only on PVA hydrogel

- Comprehensive description of lubricating processes in cartilage contact
- ✗ Simultaneous friction analysis and cartilage contact visualization
- Influence of lubricant composition on lubricating film forming

Aim of PhD Thesis

The aim of this PhD Thesis is to describe the effect of individual components of synovial fluid on lubricating film formation in the model of synovial joint. The Thesis is focused on the experimental analysis of friction coefficient and observation of adsorbed lubricating film with the use of the principle of fluorescence microscopy.

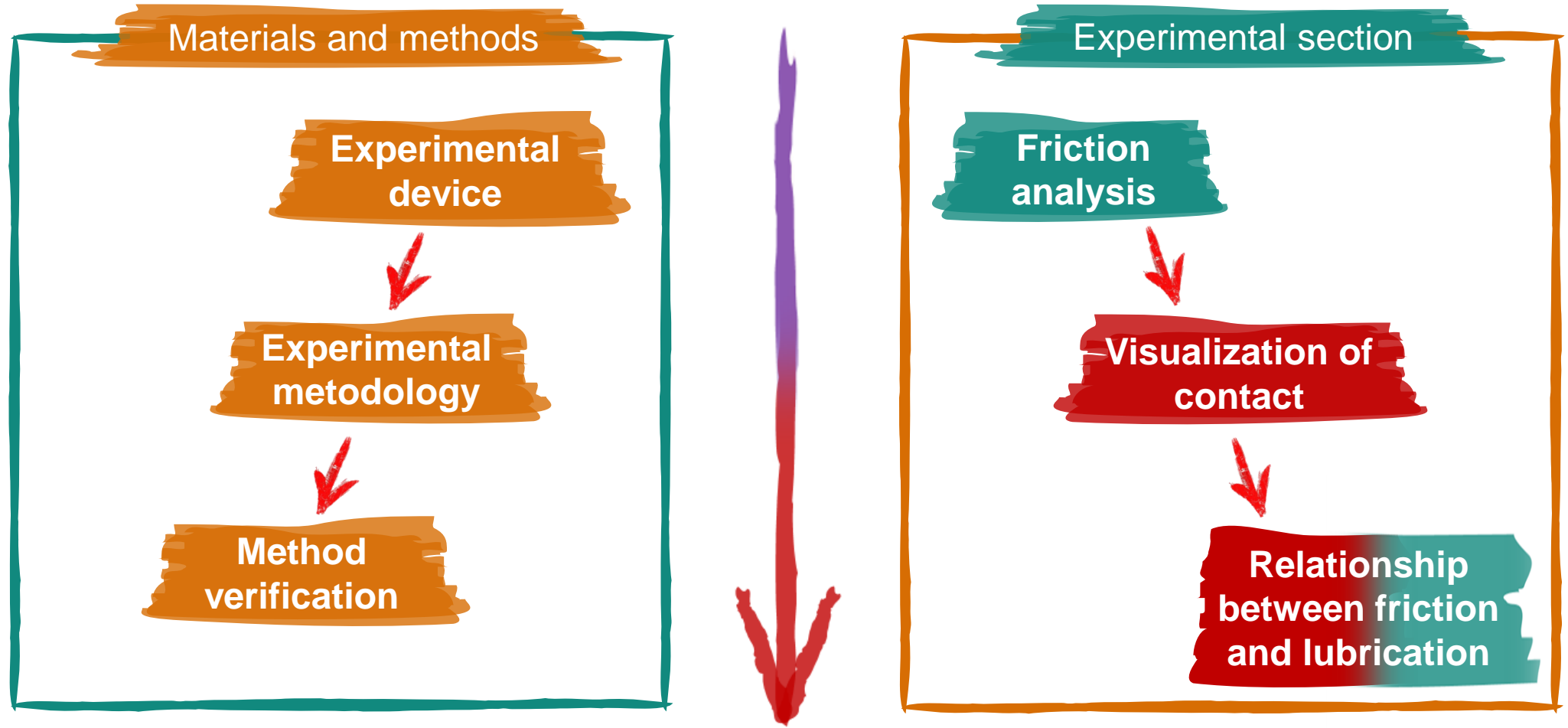
Scientific questions

- Q1:** What is the influence of the individual components contained in the model synovial fluid on the lubricating film formation in the model of synovial joint?
- Q2:** How is the friction coefficient affected by the number of dominant protein particles in the model of synovial joint?

Hypotheses

- H1:** A simple protein solution does not create a stable lubricating film with high friction coefficient. A combination of simple proteins causes the proteins to bind to each other. Forming of the lubricating film is mostly affected by hyaluronic acid and phospholipids; these components contribute to the stability of lubricating film and increase its thickness.
- H2:** It is expected that a higher particle count of dominant proteins component adsorbed in the contact causes a higher friction coefficient whereas the thickness and area of lubricating film increases.

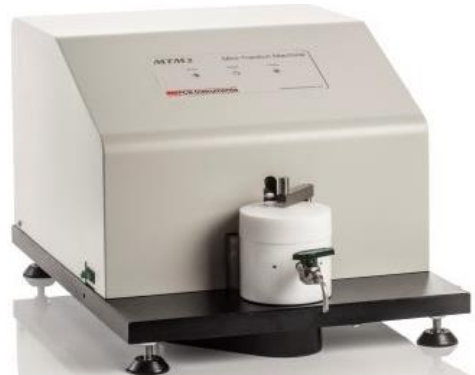
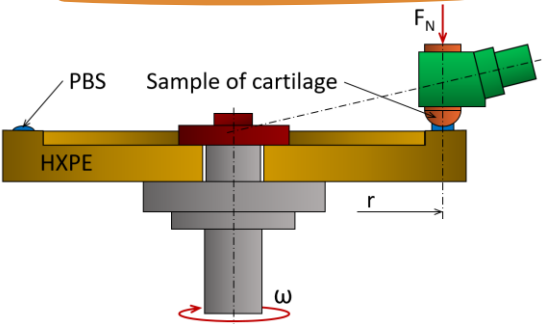
State of art



Aim of PhD Thesis

Experimental devices

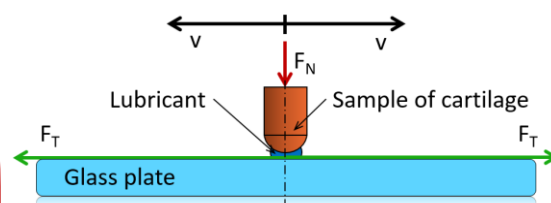
Mini Traction Machine (MTM)



Verification of cartilage sample storing method

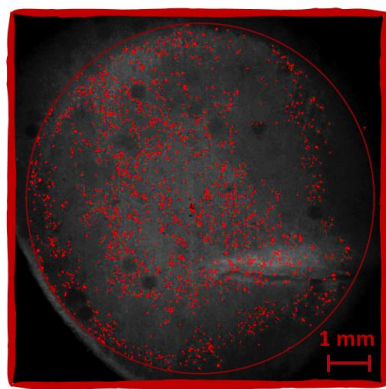
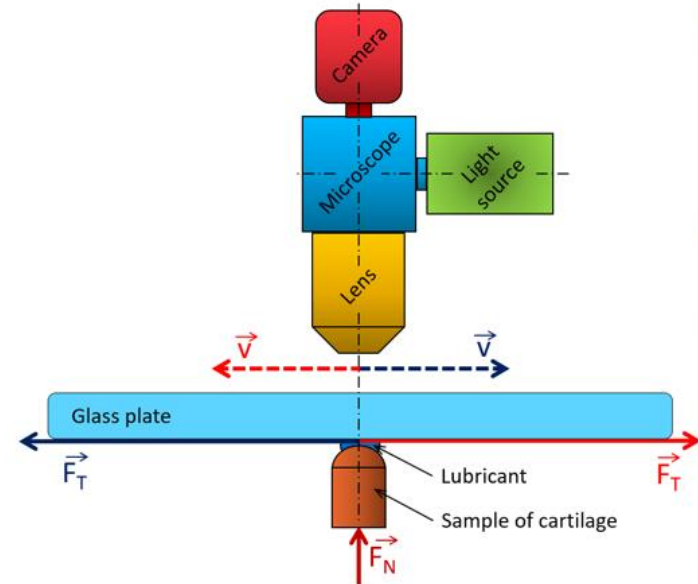
Defence of the PhD Thesis

Bruker UMT TriboLAB

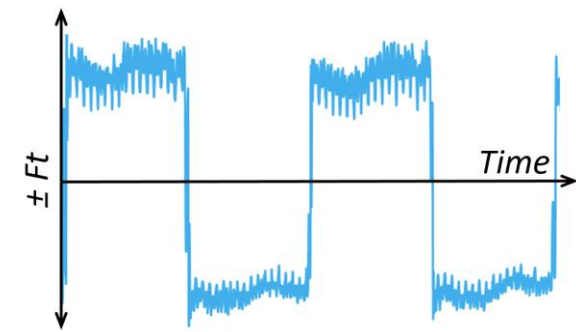


Verification of the new designed tribometer

New reciprocating tribometer



Simultaneous friction + visualization

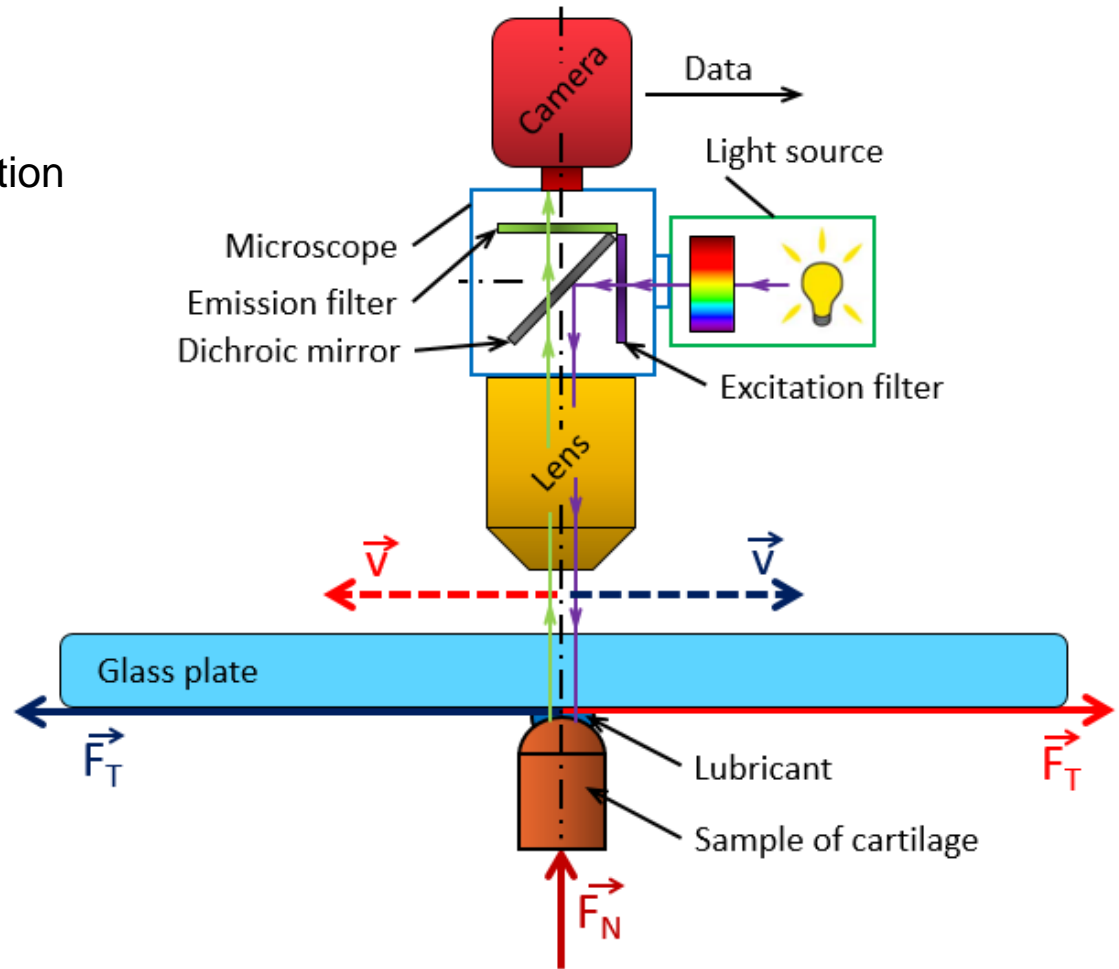


New reciprocating tribometer

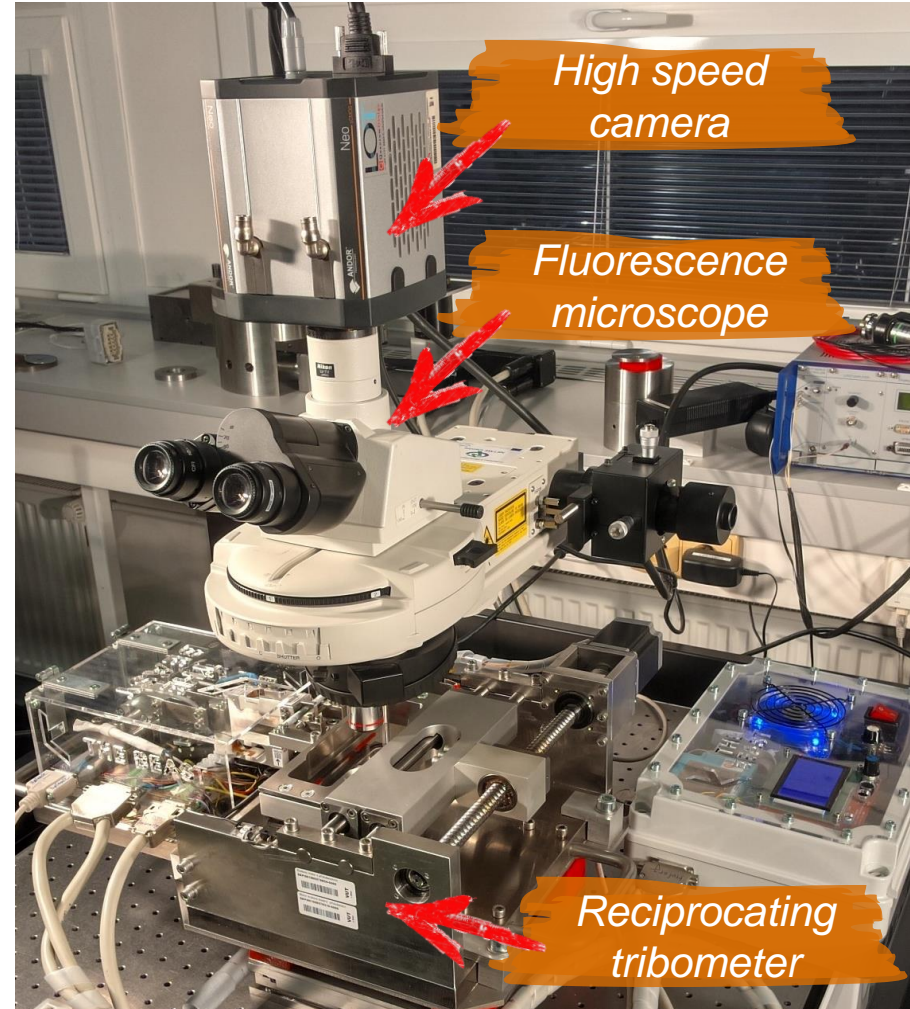
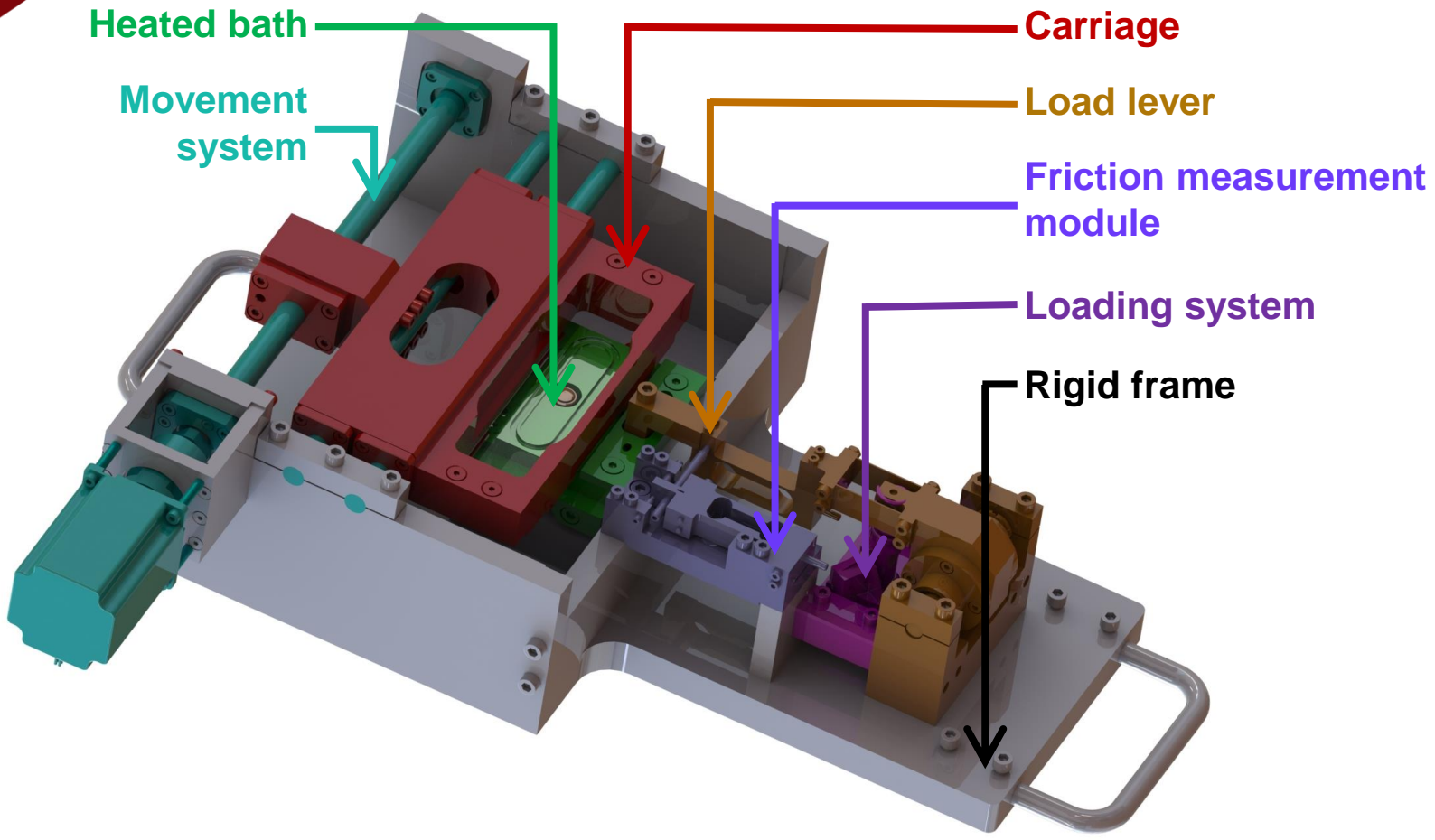
- Reciprocating pin-on-plate tribometer
- Simultaneous friction measurement and contact visualization
- Visualization method – fluorescence microscopy

Experimental conditions

- Glass plate – optical glass B270
- Pin – cartilage sample \varnothing 9,7 mm
- Load 10 N (approximately 0,8 MPa)
- Speed 10 mm/s
- Stroke length 20 mm
- Fluorescently marked lubricants



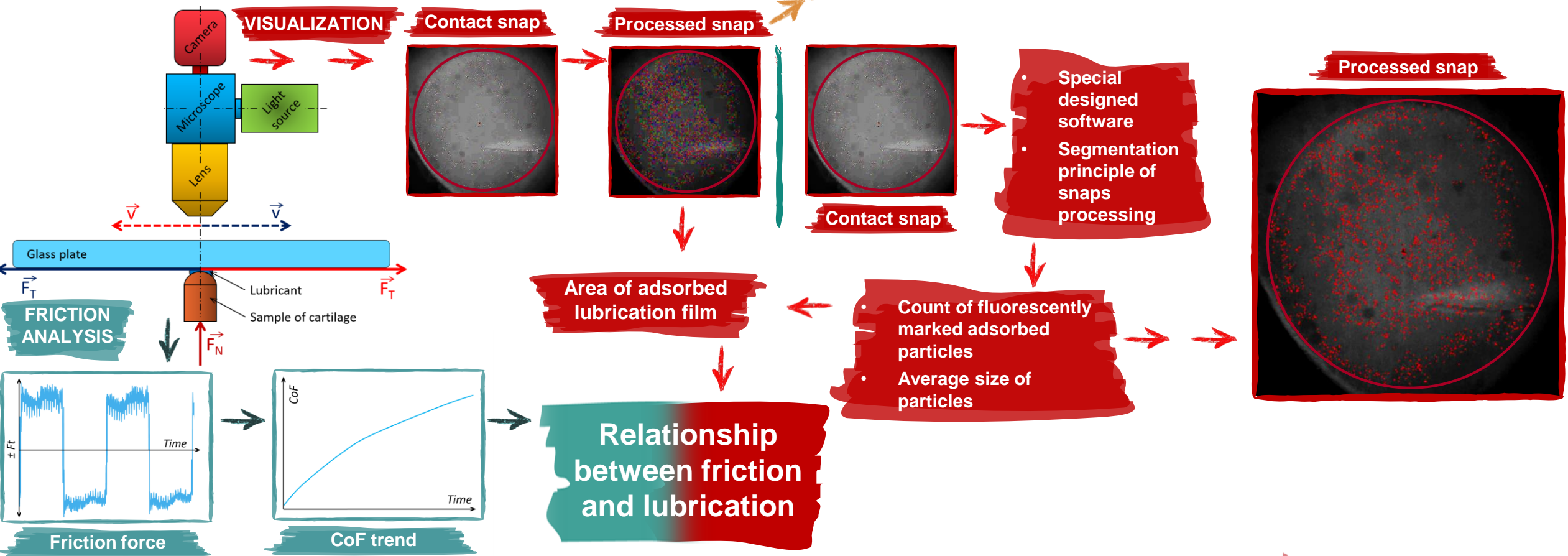
New reciprocating tribometer



Data processing

Methodology of data acquisition from the experiments

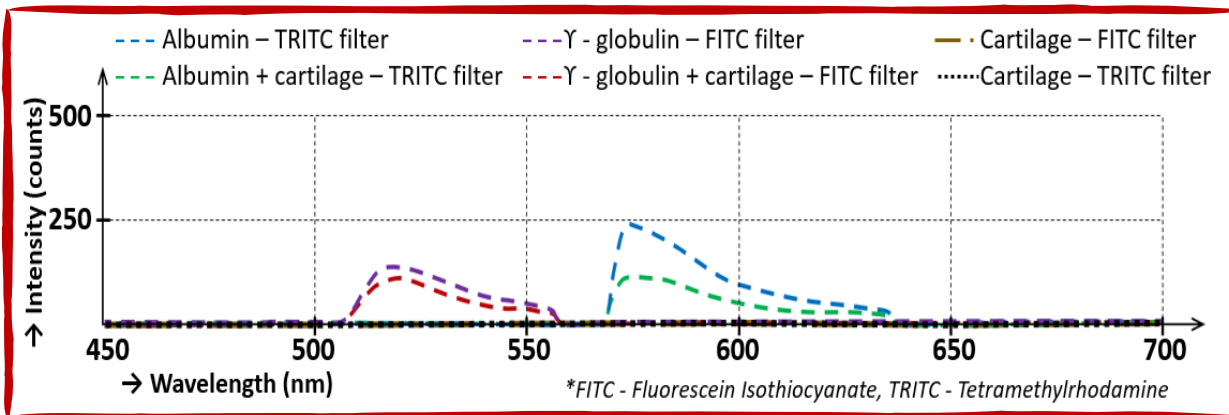
Processing – special designed SW



Verification of method

Verification of the cartilage contact

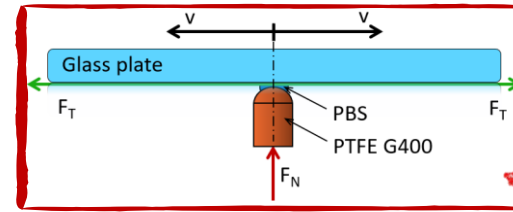
- Spectral analysis of contact with fluorescently marked lubricant
- The cartilage within fluorescently marked lubricant does not emit any light
- Only the fluorescently marked lubricants emit light



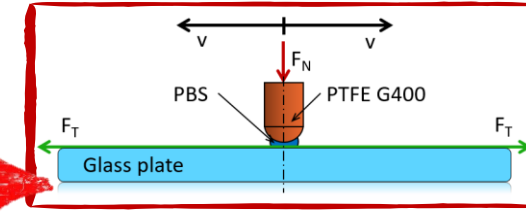
Verification and calibration of new tribometer

- The same configuration pin-on-plate
- Comparing of friction measurement (CoF)

New reciprocating tribometer



Bruker UMT TriboLAB



Verification the of data process

- Calibrating of new processing software (calibrating of segmentation process, threshold settings)
- Setting (calibrating) of software based on fluorescence intensity
- New Calibrating process to each cartilage sample

Experimental design and conditions

Cartilage sampling



- Femoral hip head of mature pigs
- Removing by ejector
- Sample s Ø 9.7 mm
- Storing in deep frozen state -20°C

Experimental design

- 9 consecutive experiments
- Rehydration performed between each experiments
- 1 experiment → 2 min → 1 rehydration 2 min

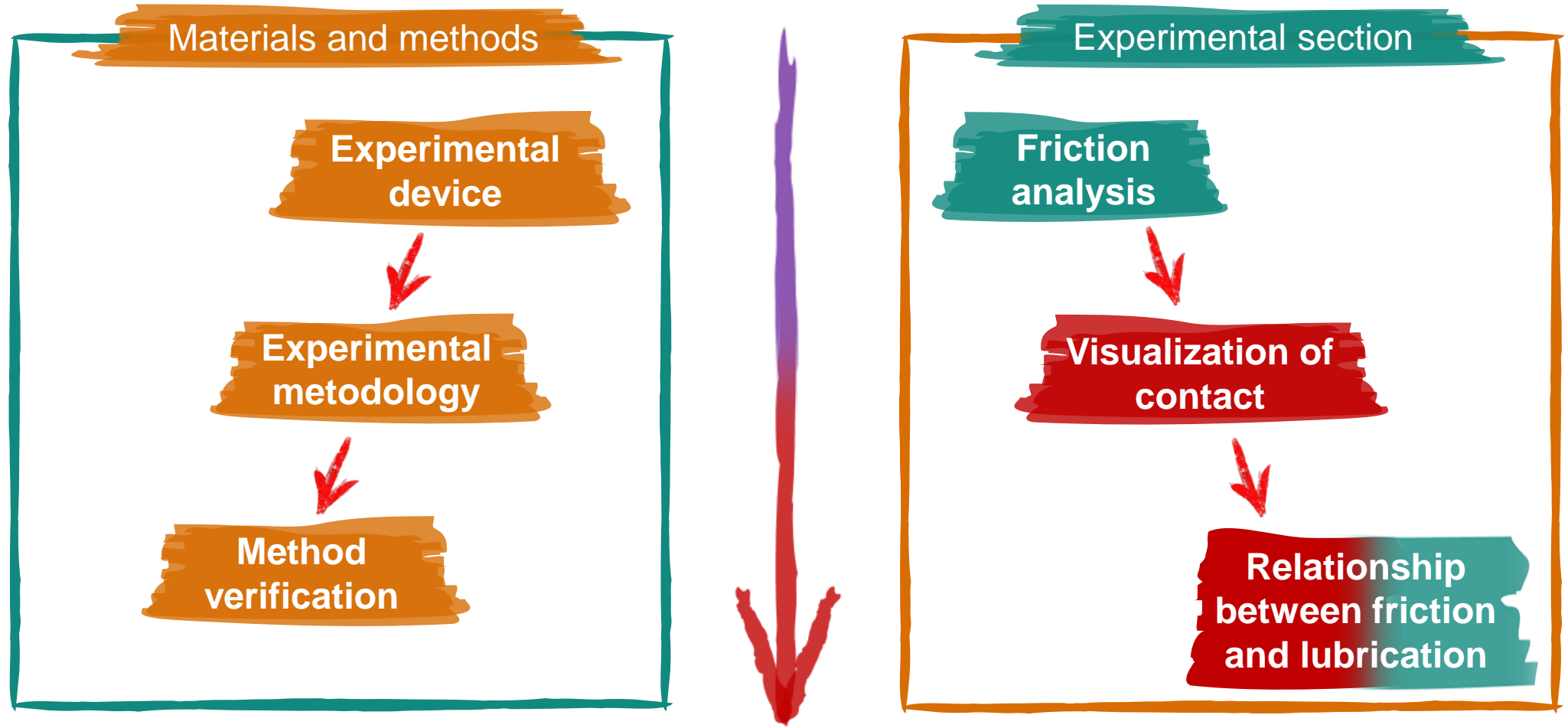
Lubricants

Lubricant label	Composition, concentration (mg/ml)				Labelled component
	Albumin	γ-globulin	HA	Phospholipids	
Lubricant 1	20	-	-	-	Albumin
Lubricant 2	20	3.6	-	-	Albumin
Lubricant 3	20	3.6	2.5	-	Albumin
Lubricant 4	20	3.6	2.5	0.15	Albumin

Experimental conditions

Load	Contact pressure	Velocity	Temperature
N	MPa	mm/s	°C
10	0.8	10	37

State of art



Aim of PhD Thesis

Lubricating film formation - FRICTION ANALYSIS

Experimental task

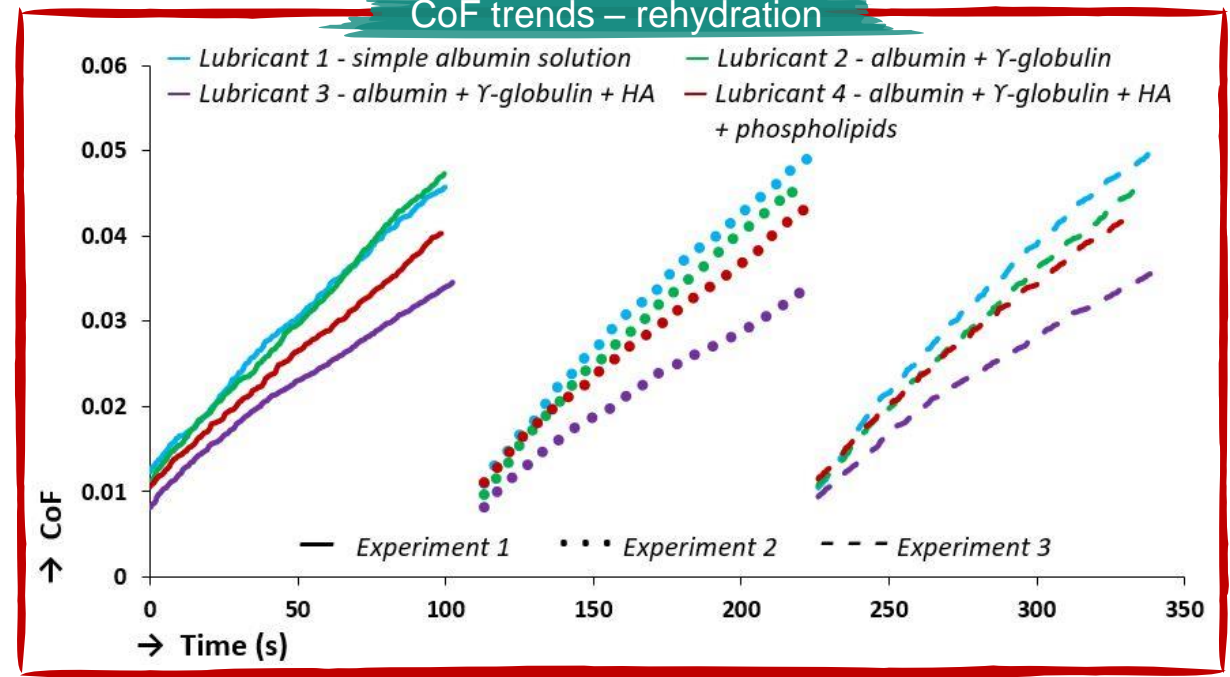
Visualization

Friction analysis

Relationship between friction and lubrication

- Simultaneous visualization and friction analysis
- Albumin protein was fluorescently marked through all experiments
- Focusing of experiments - observation of albumin proteins

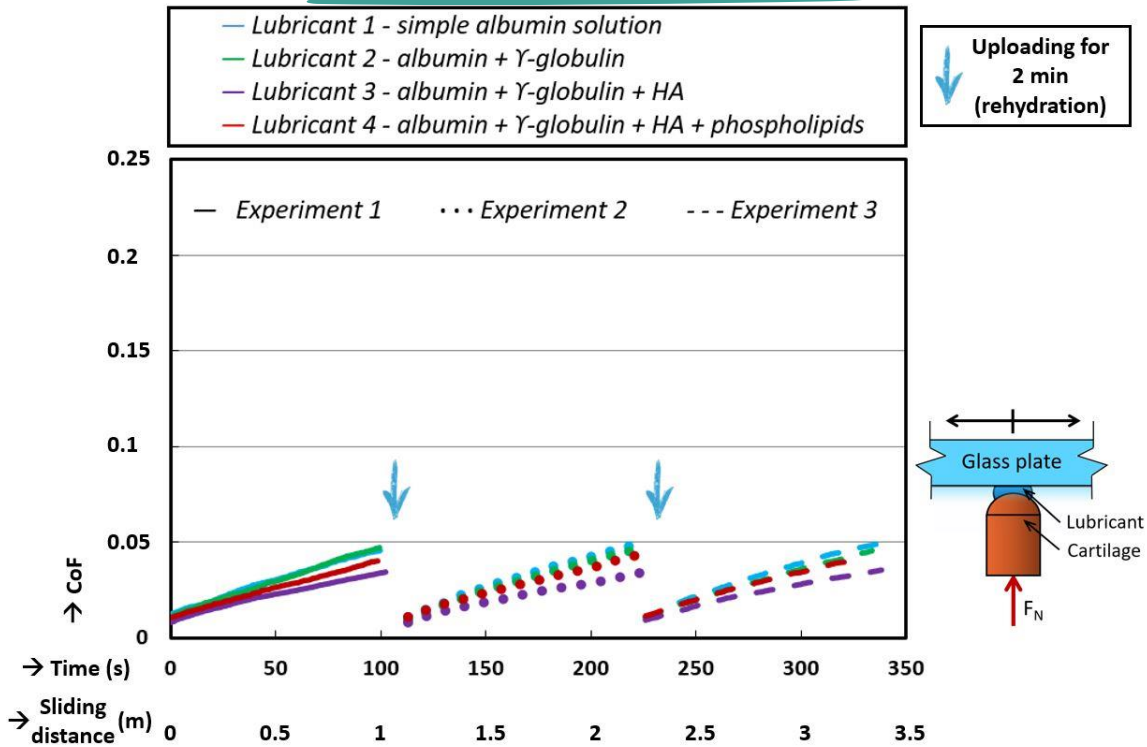
CoF trends – rehydration



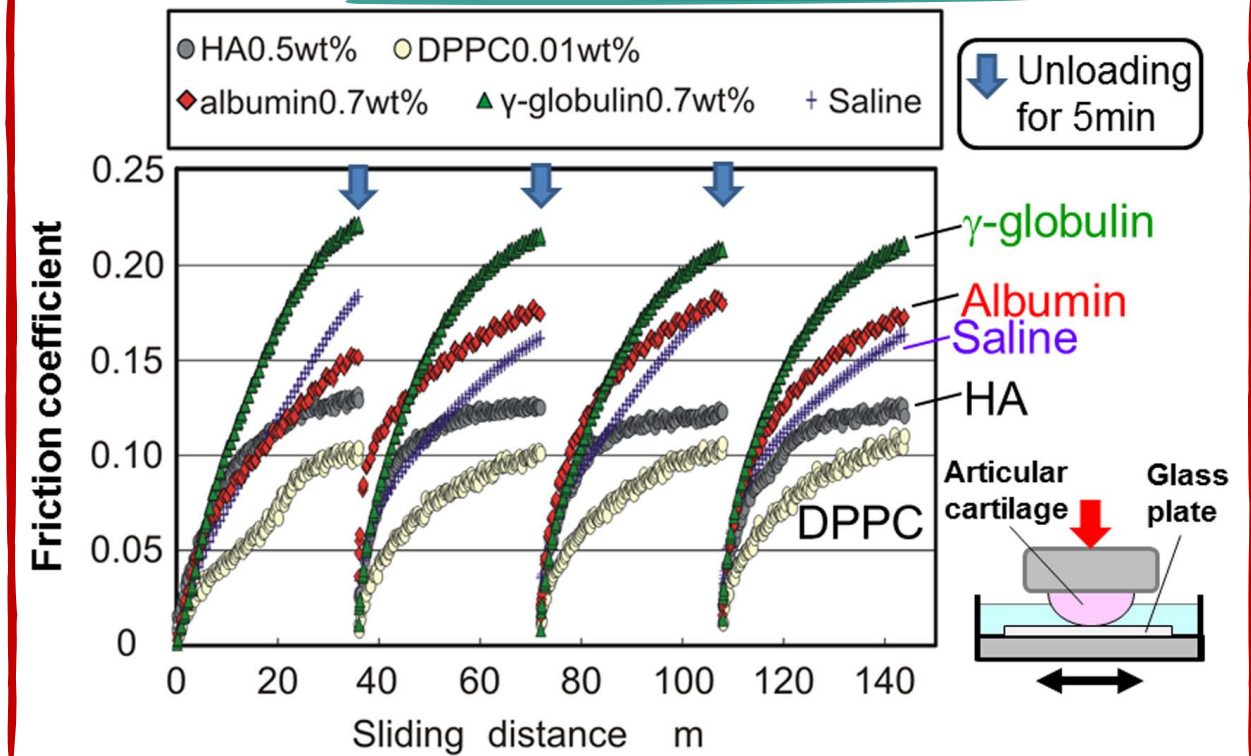
Lubricating film formation - FRICTION ANALYSIS

Friction analysis - comparison with other works

CoF trends



CoF trends – Murakami, T. et al.



Lubricating film formation - VISUALIZATION

Visualization

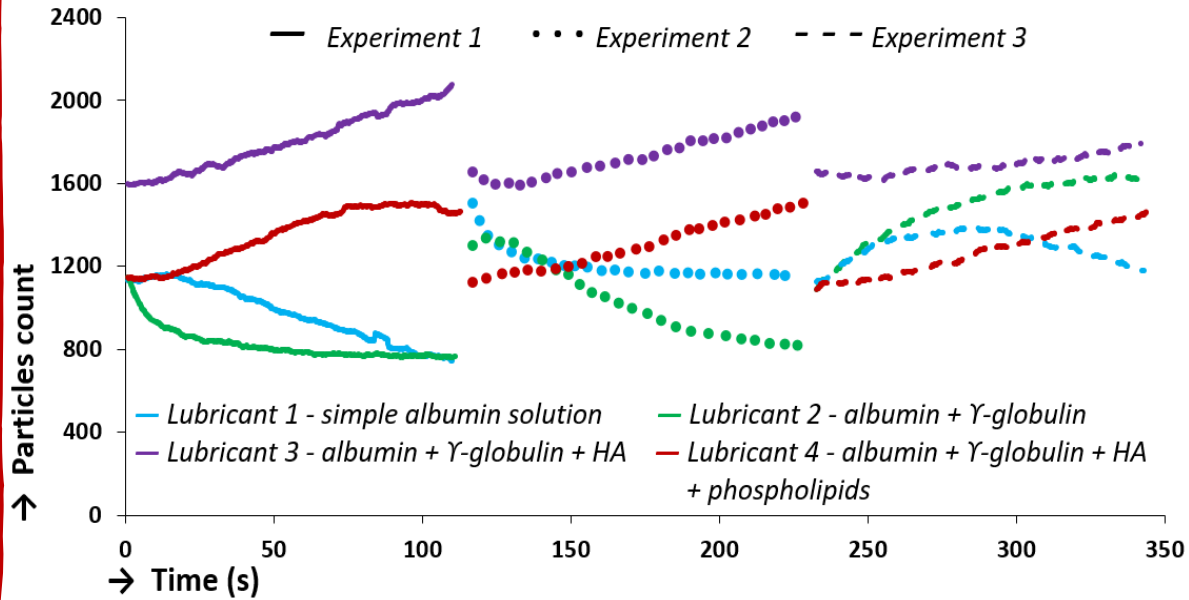
Record of contact area

Particles count

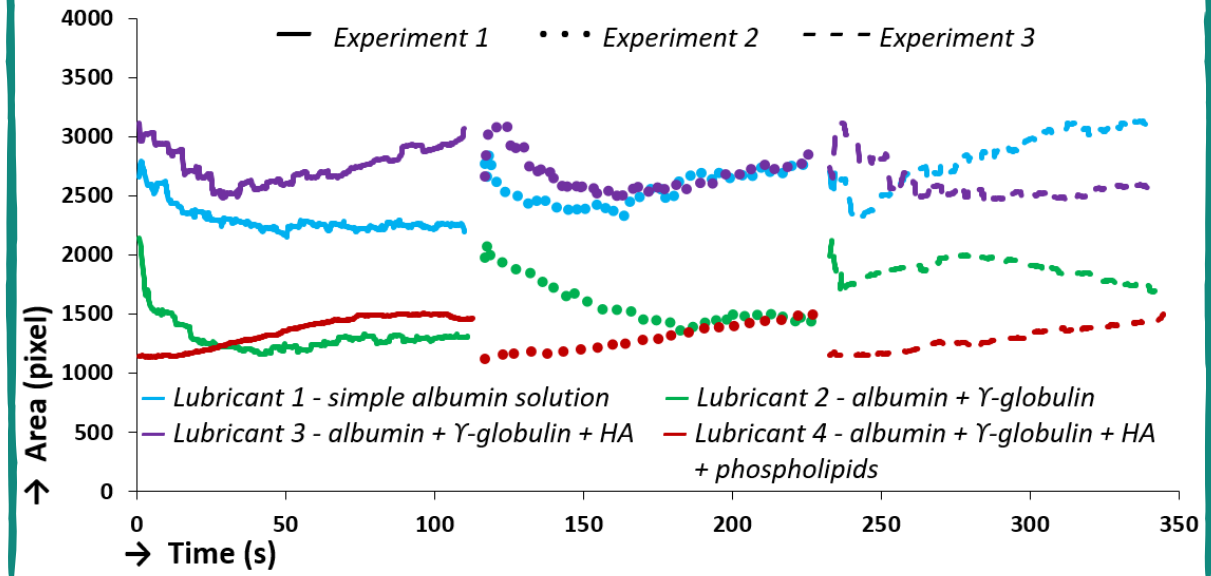
Average size of particles

Area of adsorbed lubrication film

Particles count in the contact



Area of adsorbed lubrication film created by albumin



Lubricating film formation - VISUALIZATION

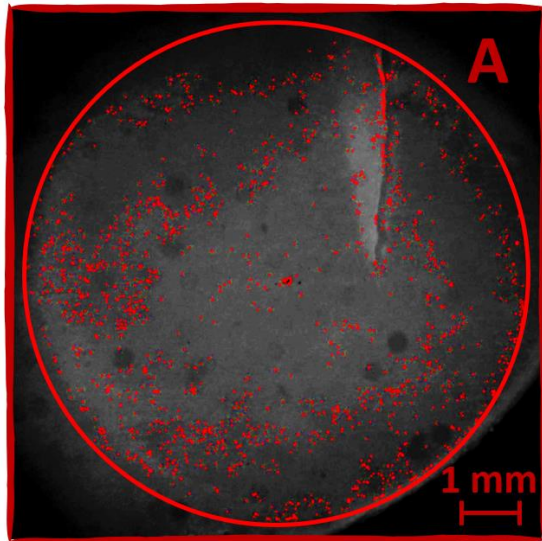
Visualization

Record of contact area

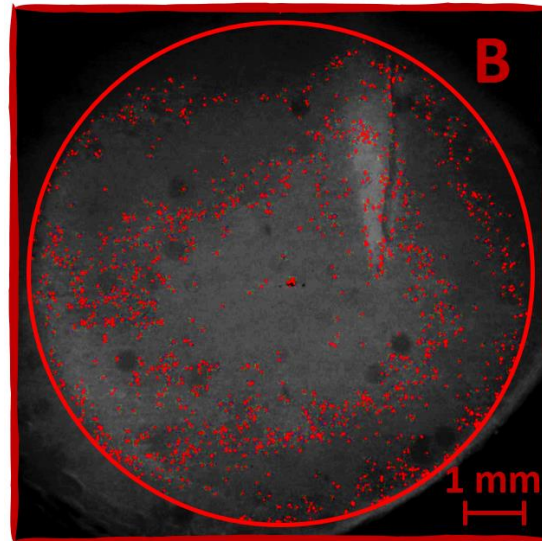
Particles count

Average size of particles

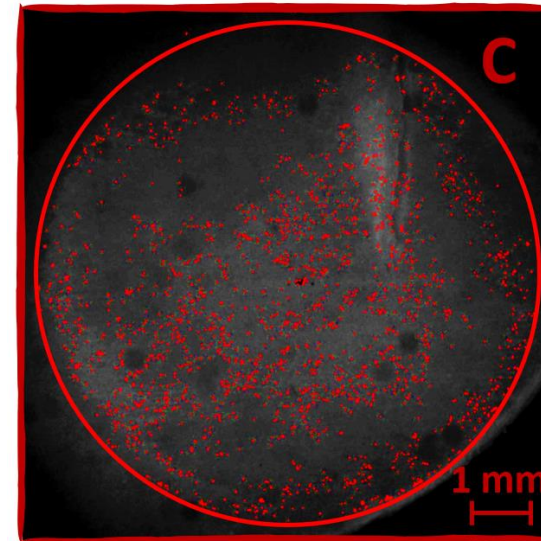
Area of adsorbed lubrication film



t = 0 s
Particles count: 1579
Average size of particles: 1

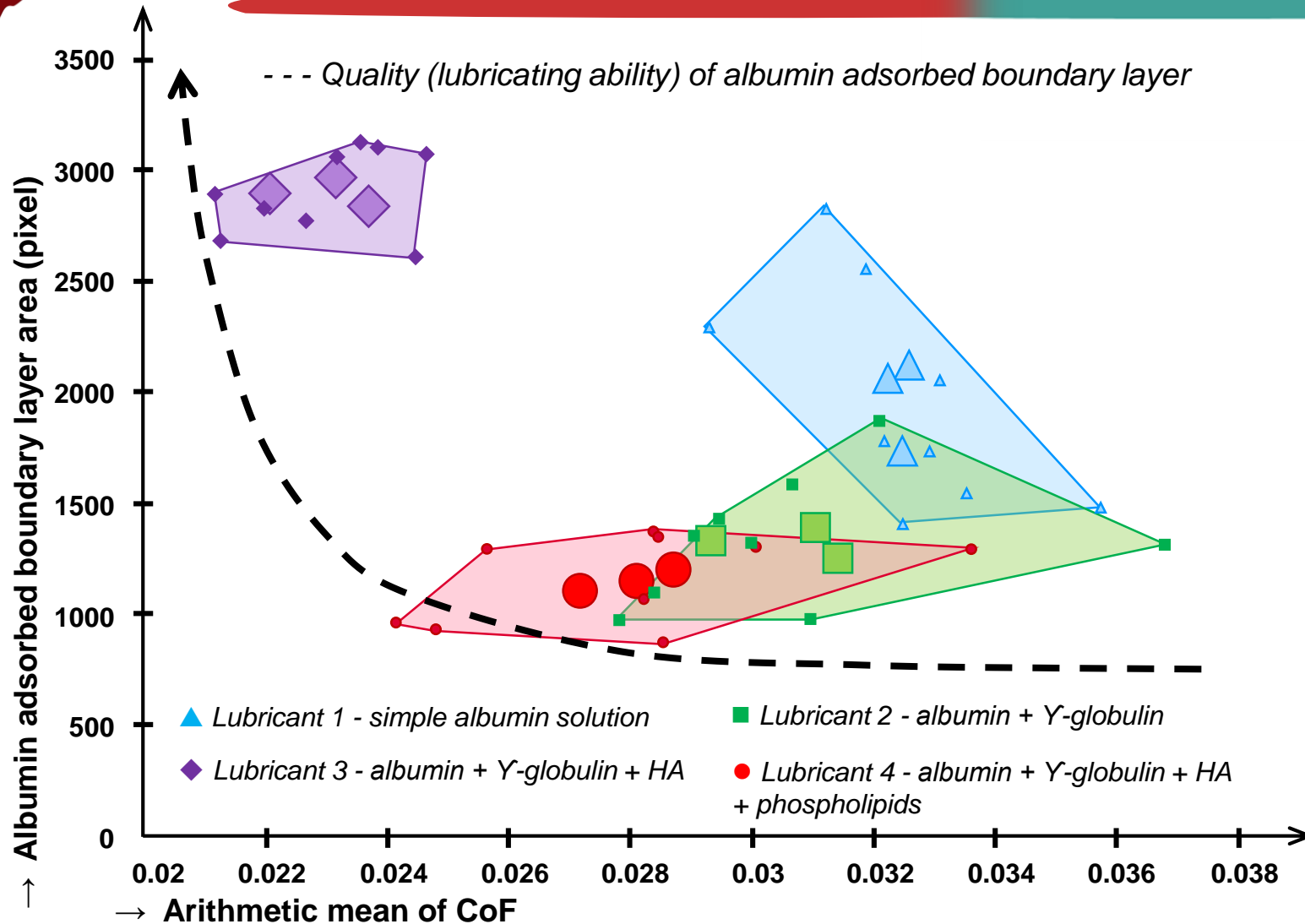


t = 25 s
Particles count: 1686
Average size of particles: 1



t = 100 s
Particles count: 2164
Average size of particles: 1

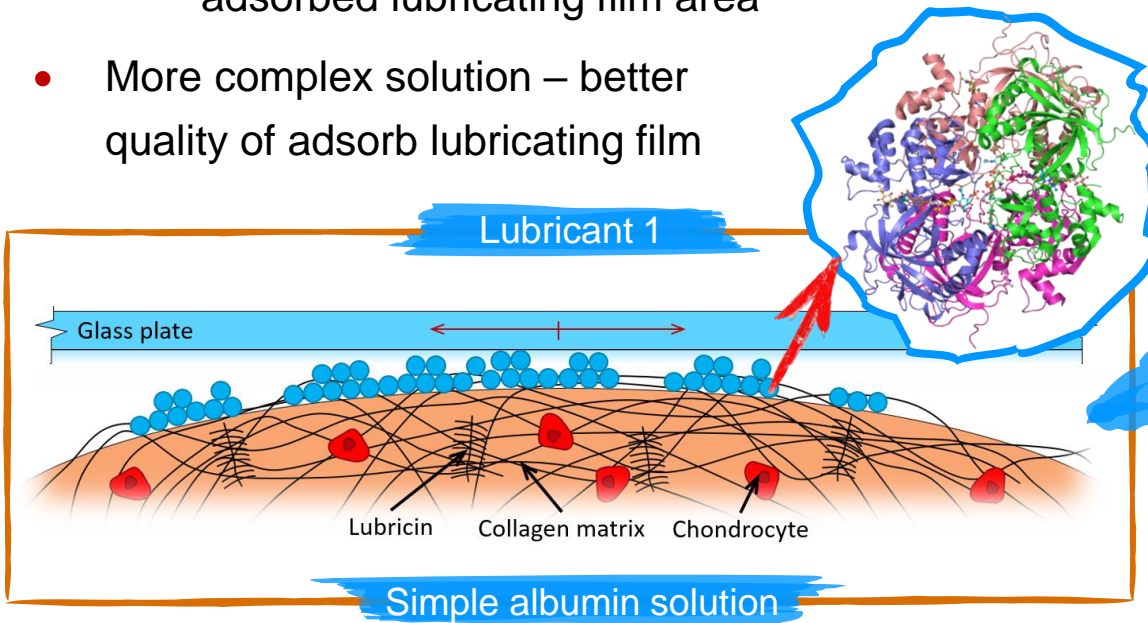
Lubricating film formation – RELATIONSHIP BETWEEN FRICTION AND LUBRICATION



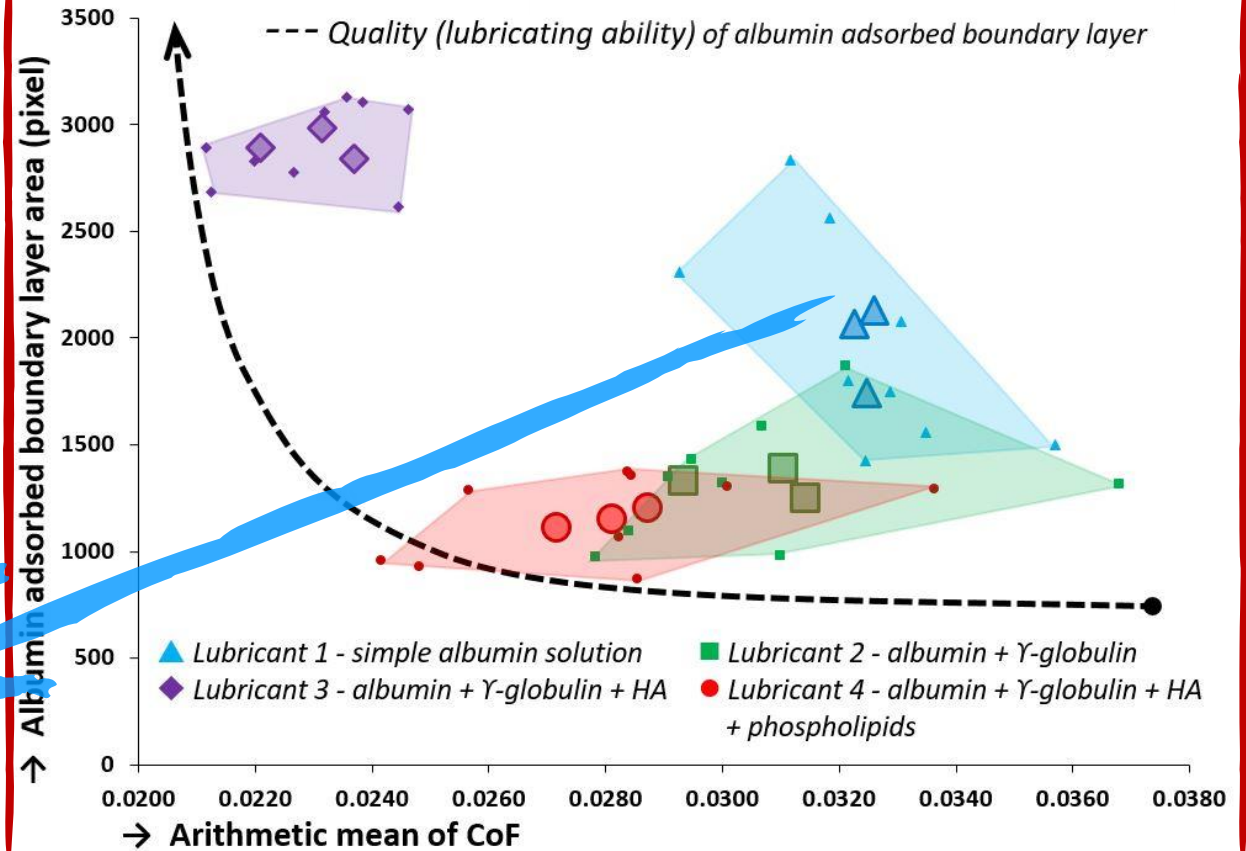
- Relationship between
 - FRICTION
 - LUBRICATION
- Global goal
 - the smallest possible CoF
 - the largest adsorbed lubricating film area
- Smaller points – each experiment
- Bigger points – average of the experimental set
- Regions – limit the extreme points of the area
- Best lubricating ability – Lubricant 3
- The only lubricant 4 provides long-term protection of cartilage surface

Lubricating film formation – RELATIONSHIP BETWEEN FRICTION AND LUBRICATION

- Dependence of CoF on adsorbed film area
- Quantification of quality of adsorbed lubricating film
- Global goal
 - The smallest possible CoF along with the largest adsorbed lubricating film area
- More complex solution – better quality of adsorb lubricating film

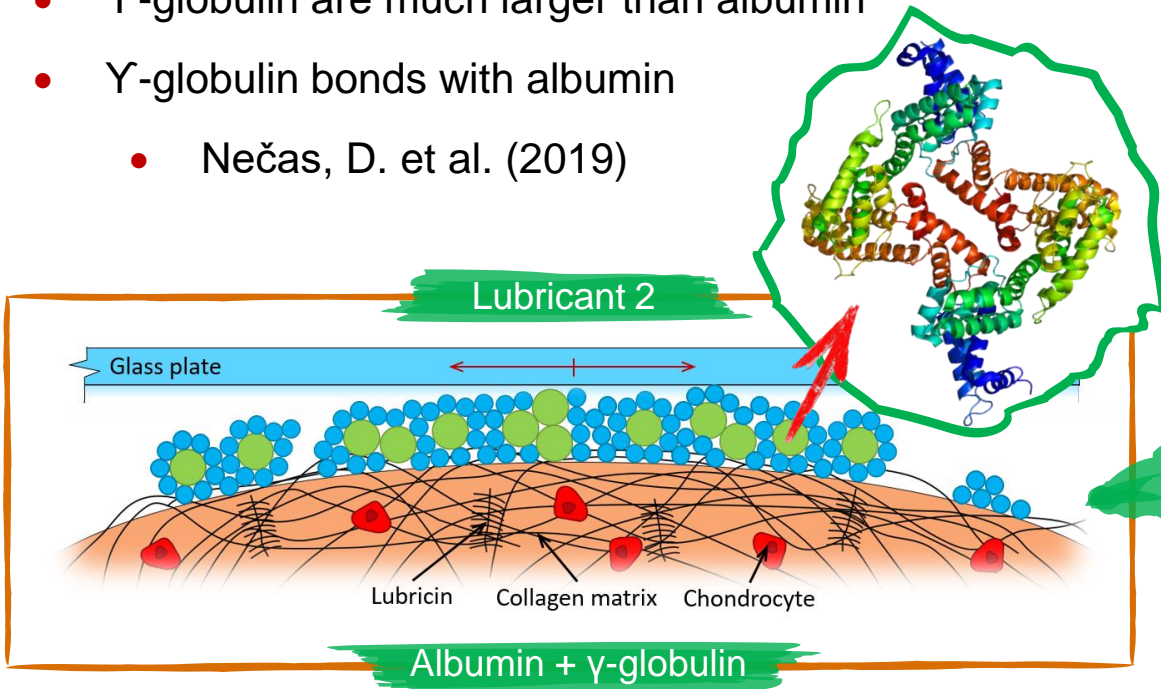


Relationship between friction and lubrication

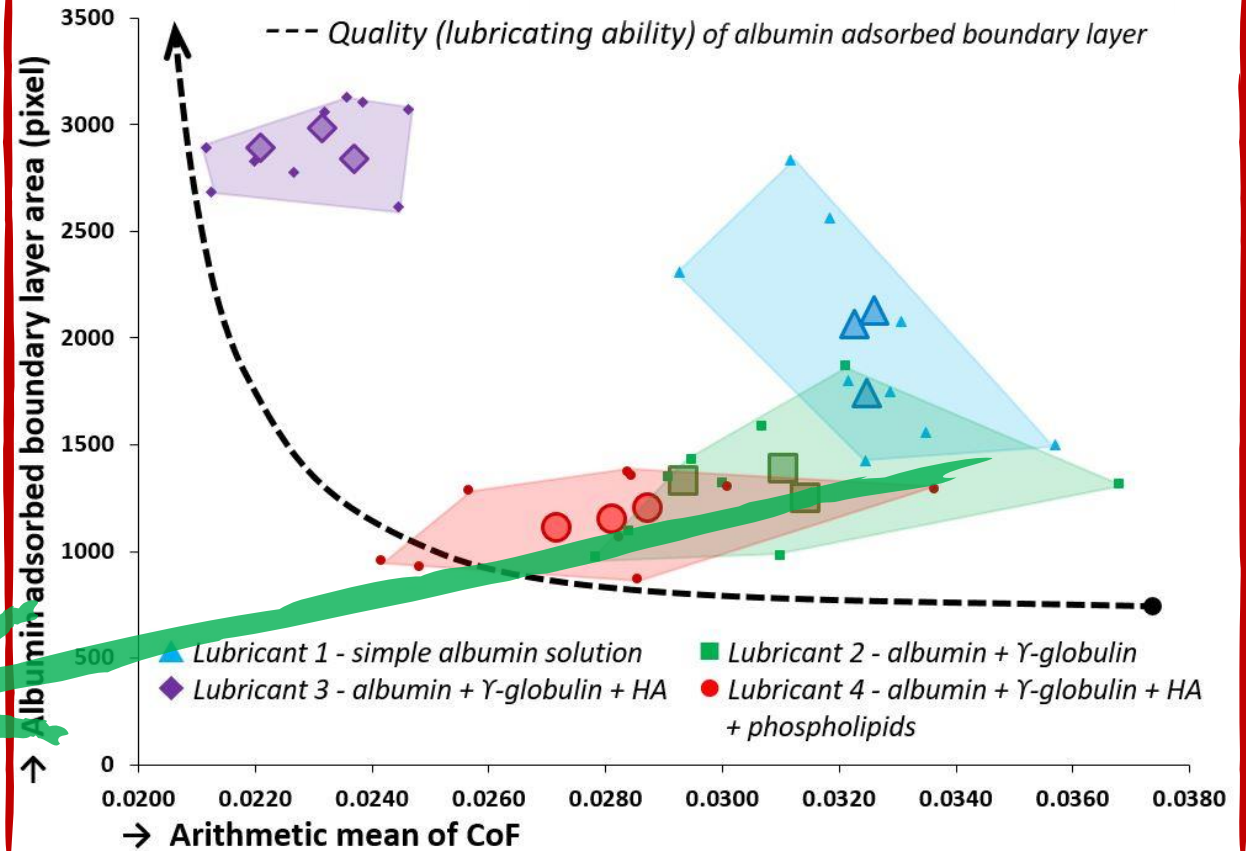


Lubricating film formation – RELATIONSHIP BETWEEN FRICTION AND LUBRICATION

- Dev, S. B. et al. (1988), Howard, M. J. et al. (2005)
 - Albumin - α -helix structure
 - γ -Globulin - β -sheet structure
- γ -globulin are much larger than albumin
- γ -globulin bonds with albumin
 - Nečas, D. et al. (2019)

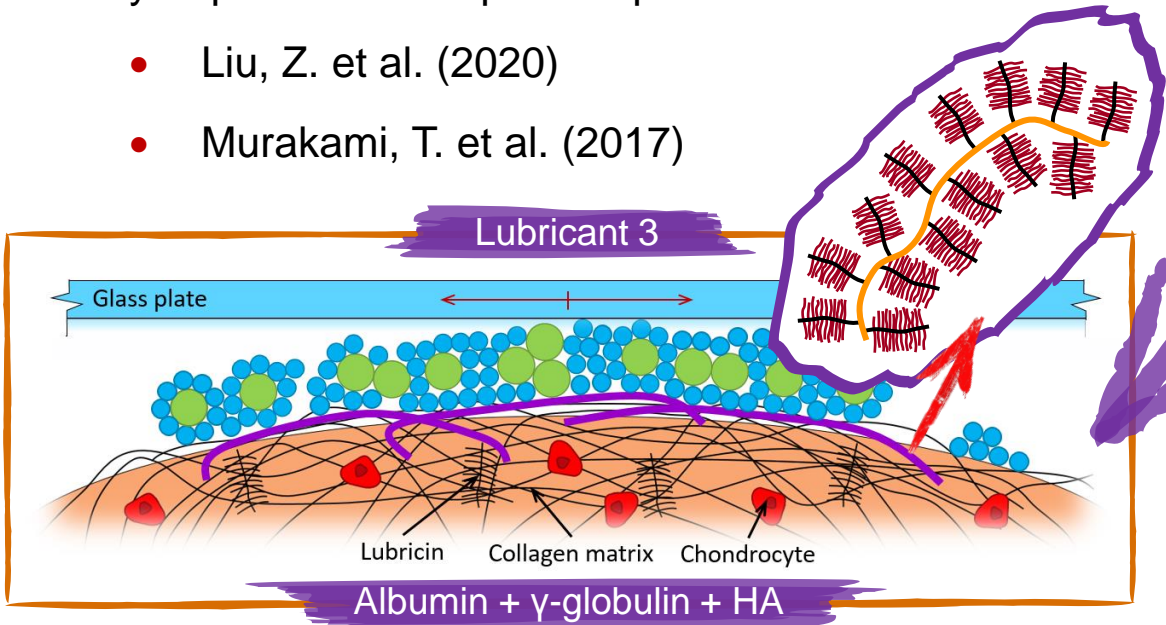


Relationship between friction and lubrication

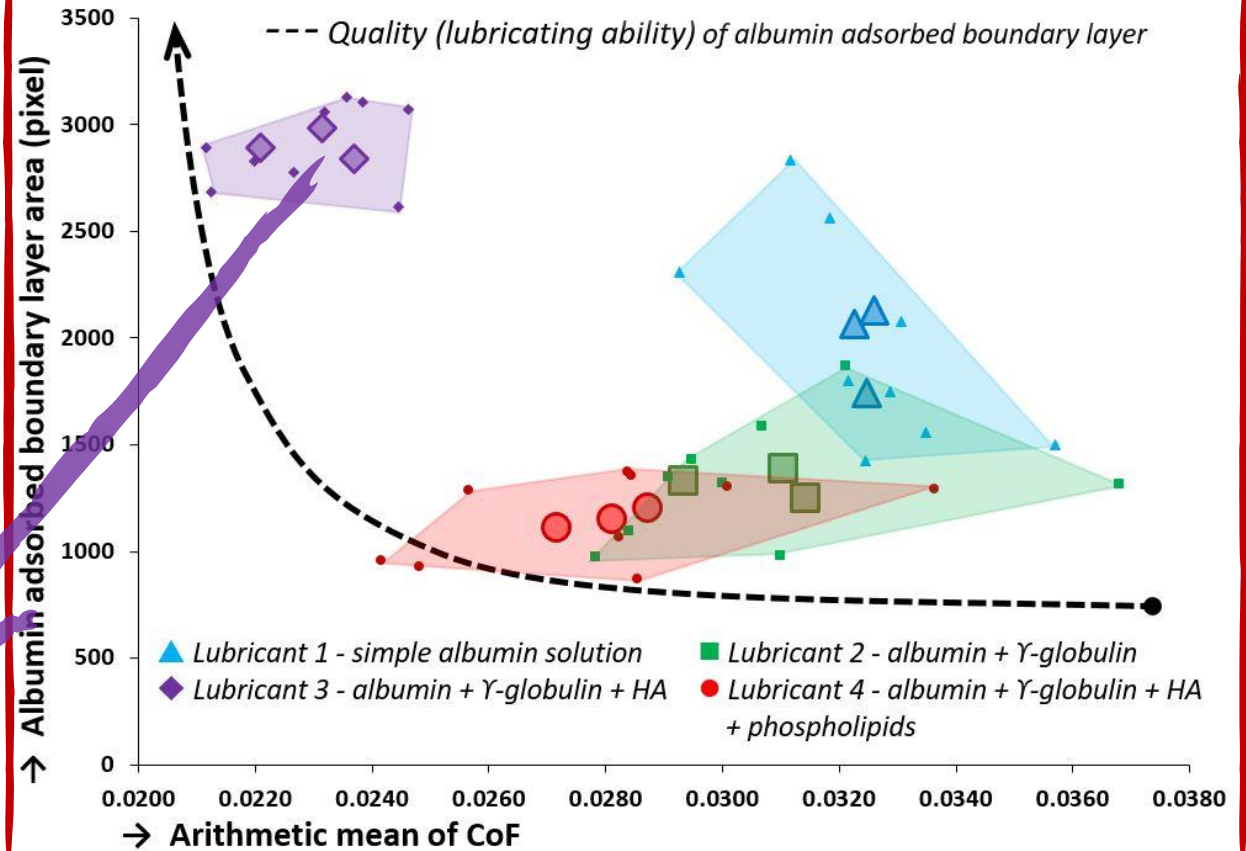


Lubricating film formation – RELATIONSHIP BETWEEN FRICTION AND LUBRICATION

- Larger molecules of HA create gel-like layer on the cartilage surface
 - Wu, T. et al. (2015), Forsey, R. et al. (2006)
- HA gel-like layer on cartilage surface is strongly hydrophilic → absorption of proteins increases
 - Liu, Z. et al. (2020)
 - Murakami, T. et al. (2017)

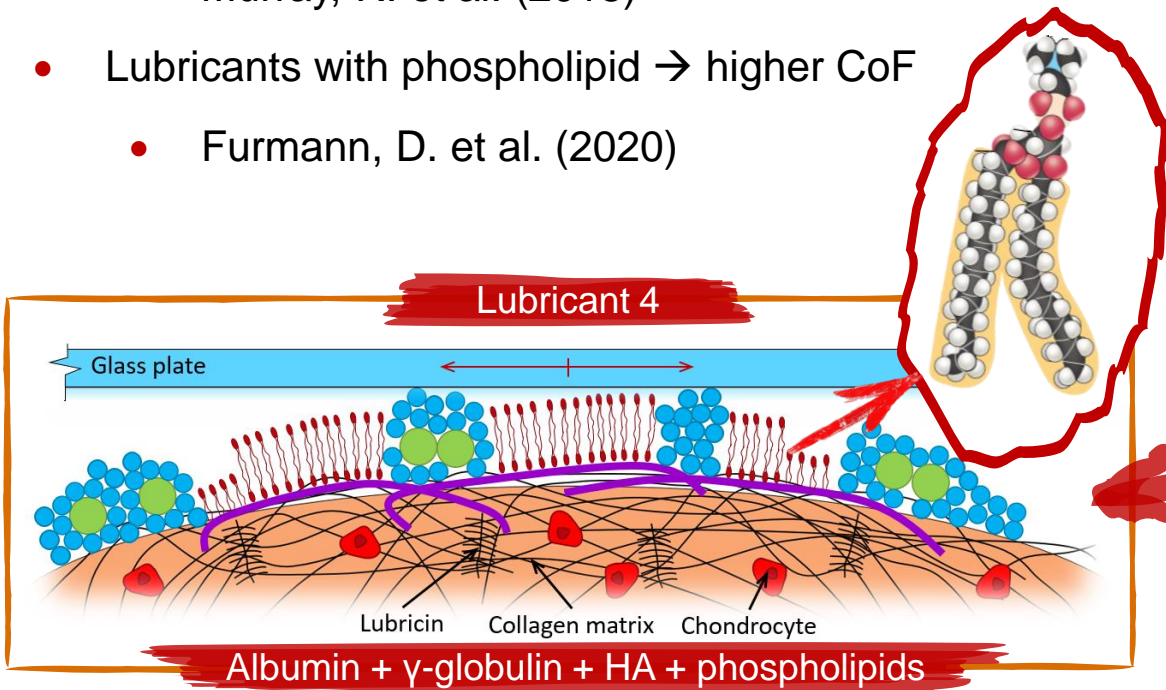


Relationship between friction and lubrication

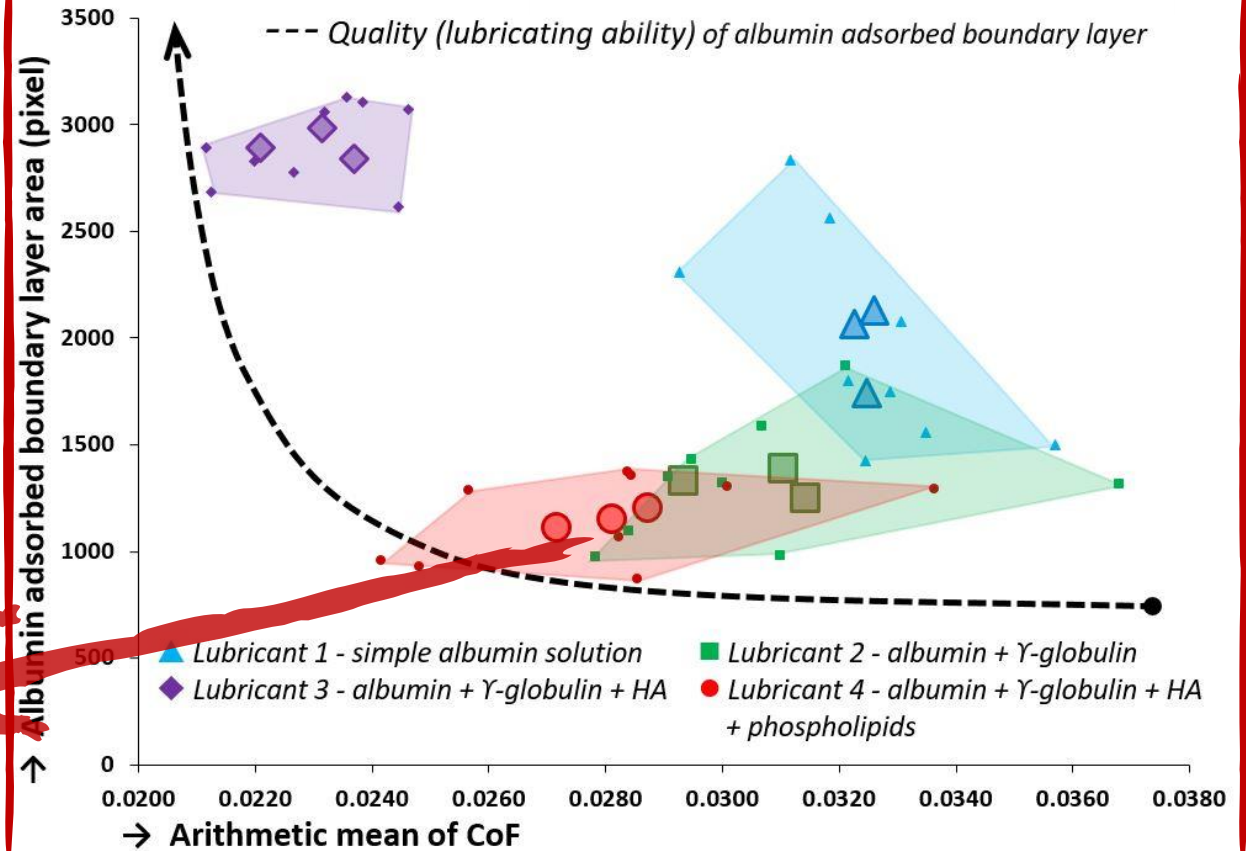


Lubricating film formation – RELATIONSHIP BETWEEN FRICTION AND LUBRICATION

- Phospholipids bond HA layer by phosphate core
- Phospholipids interbond by lipid tail → bilayer
 - Jahn, S. et al. (2016), Klein, J. et al. (2013), Murray, R. et al. (2018)
- Lubricants with phospholipid → higher CoF
 - Furmann, D. et al. (2020)



Relationship between friction and lubrication



Summarization of scientific questions and hypotheses

Hypothesis 1

A simple protein solution does not create a stable lubricating film with high friction coefficient. A combination of simple proteins causes the proteins to bind to each other. Forming of the lubricating film is mostly affected by hyaluronic acid and phospholipids; these components contribute to the stability of lubricating film and increase its thickness.

- The simple albumin solution does not create a stable lubrication film
- The mixture of proteins causes the binding of proteins to each other
- The forming of the lubricating film is mostly affected by hyaluronic acid
- The HA causes larger area of adsorbed film and noticeable reduction of friction
- Only the complex lubricants create stable adsorbed film - increasing of adsorbed film area during the experiment

**Hypothesis WAS
NOT FALSIFIED
WHITIN THIS Thesis**

Summarization of scientific questions and hypotheses

Hypothesis 2

It is expected that a higher particle count of dominant proteins component adsorbed in the contact causes a higher friction coefficient whereas the thickness and area of lubricating film increases.

- The best frictional properties were observed for the lubricant consists proteins and HA
- The largest area of adsorbed film was observed for the lubricant consists proteins and HA
- The simple albumin solution showed a larger adsorbed film than the mixture of proteins
- The mixture of proteins gives better values of friction coefficient

**Hypothesis WAS
FALSIFIED WITHIN
THIS Thesis**

NEW Hypothesis 2

The larger adsorbed protein film area causes higher CoF, however only for simple protein solutions. HA increases the adsorption of proteins in the contact and significantly reduces the CoF. Phospholipids cause the proteins to capture in the contact.

Thesis layout

Introducing the newly developed tribometer

Introducing the experimental methodology and data processing

Role of albumin in adsorbed film formation in cartilage contact

Tribology in Industry

2019

Materials

2020

JESTECH

2022

Visualization of Lubrication Film in Model of Synovial Joint

Biotribology of Synovial Cartilage: A New Method for Visualization ...

Biotribology of synovial cartilage: Role of albumin in adsorbed film ...

CiteScore = 1.09

Author's contribution: 70%

Journal impact factor = 3.06

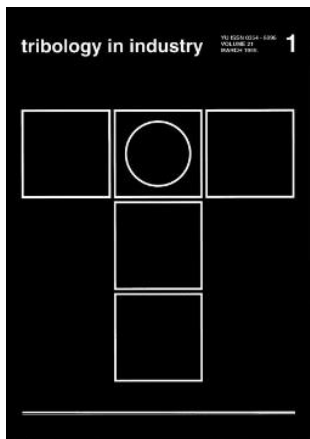
CiteScore = 3.26, Quartile Q2

Author's contribution: 50%

Journal impact factor = 4.36

CiteScore = 9.00, Quartile Q2

Author's contribution: 70%



materials



List of publication

Journals with impact factor

ČÍPEK, P.; VRBKA, M.; REBENDA, D.; NEČAS, D.; KŘUPKA, I. Biotribology of Synovial Cartilage: A New Method for Visualization of Lubricating Film and Simultaneous Measurement of the Friction Coefficient. *Materials*, 2020, **13**(9), 1-20. ISSN: 1996-1944.

REBENDA, D.; VRBKA, M.; ČÍPEK, P.; TOROPITSYN, E.; NEČAS, D.; PRAVDA, M.; HARTL, M. On the Dependence of Rheology of Hyaluronic Acid Solutions and Frictional Behaviour of Articular Cartilage. *Materials*, 2020, **13**(11), 1-14. ISSN: 1996-1944.

FURMANN, D.; NEČAS, D.; REBENDA, D.; ČÍPEK, P.; VRBKA, M.; KŘUPKA, I.; HARTL, M. The effect of synovial fluid composition, speed and load on frictional behaviour of articular cartilage. *Materials*, 2020, **13**(6), 1-16. ISSN: 1996-1944.

REBENDA, D.; VRBKA, M.; NEČAS, D.; TOROPITSYN, E.; YARIMITSU, S.; ČÍPEK, P.; PRAVDA, M.; HARTL, M. Rheological and frictional analysis of viscosupplements towards improved lubrication of human joints. *Tribology International*, 2021, **160**: 107030. ISSN: 0301679X

ČÍPEK, P.; VRBKA, M.; REBENDA, D.; NEČAS, D.; KŘUPKA, I. Biotribology of Synovial Cartilage: Role of Albumin in Adsorbed Film Formation. *Engineering Science and Technology, an International Journal*. 2022, **34**, 101090. ISSN: 22150986

Peer-reviewed journals

ČÍPEK, P.; REBENDA, D.; NEČAS, D.; VRBKA, M.; KŘUPKA, I.; HARTL, M. Visualization of Lubrication Film in Model of Synovial Joint. *Tribology in Industry*, 2019, 41(3), 387-393. ISSN: 0354-8996.

List of publication

Conference proceedings

ČÍPEK, P.; REBENDA, D.; VRBKA, M.; HARTL, M. OBSERVATION OF LUBRICATION FILM IN SYNOVIAL JOINT. In Proceedings on Engineering Science. Proceedings on Engineering Sciences - 16th International Conference on Tribology. Kragujevac: University of Kragujevac, Faculty of Engineering, 2019, 687-692. ISSN: 2620-2832.

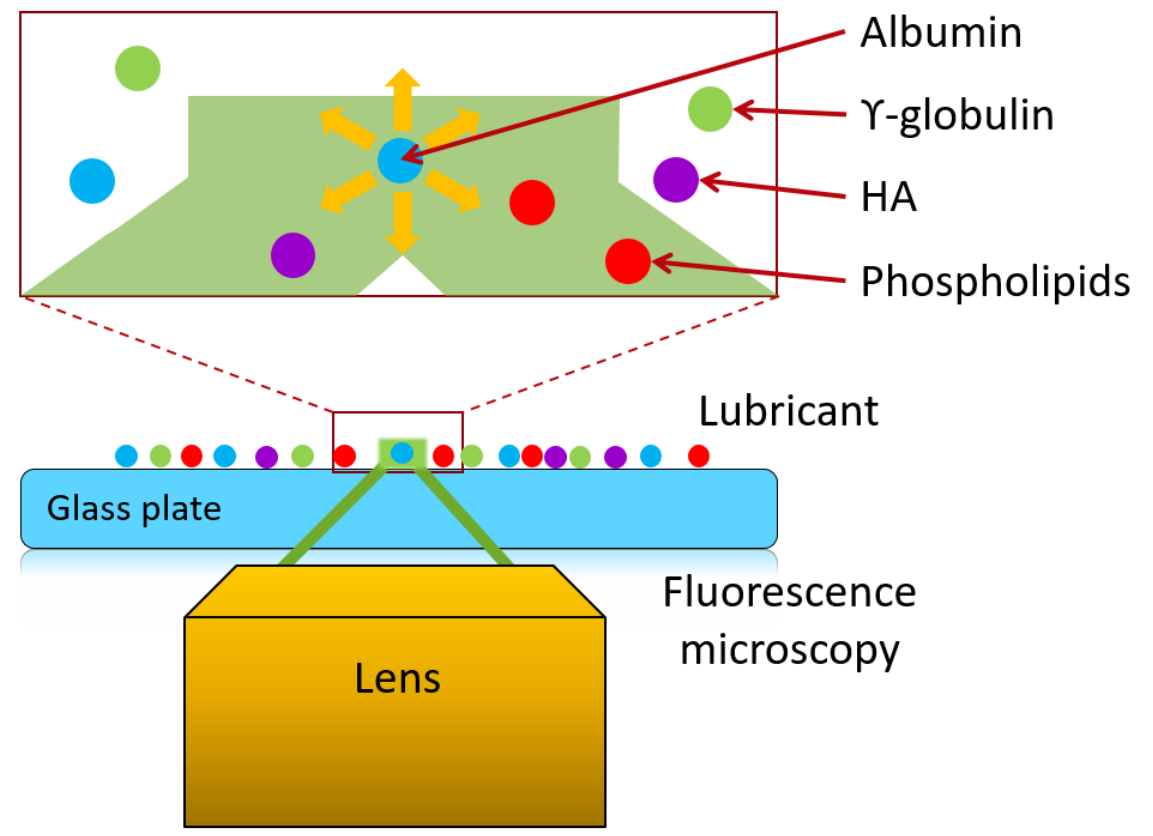
REBENDA, D.; ČÍPEK, P.; VRBKA, M.; KŘUPKA, I. Effect of Hyaluronic Acid Molecular Weight on Friction of Articular Cartilage. In Proceedings on Engineering Sciences - 16th International Conference on Tribology. Proceedings on Engineering Sciences - 16th International Conference on Tribology. Kragujevac: University of Kragujevac, Faculty of Engineering, 2019, 693-697. ISSN: 2620-2832.

ČÍPEK, P.; REBENDA, D.; NEČAS, D.; VRBKA, M.; KŘUPKA, I. Development of reciprocating tribometer for testing synovial joint. In Engineering Mechanics 2018. Engineering Mechanics 2018. First edition. Praha: Institute of Theoretical and Applied Mechanics of the Czech Academy of Sciences, Prague, 2018, 169-172. ISBN: 978-80-86246-88-8. ISSN: 1805-8256.

REBENDA, D.; ČÍPEK, P.; NEČAS, D.; VRBKA, M.; HARTL, M. Effect of Hyaluronic Acid on Friction of Articular Cartilage. In Engineering Mechanics 2018. First. Praha: Institute of Theretical and Applied Mechanics of the Czech Academy of Sciences, 2018, 709-712. ISBN: 978-80-86246-91-8.

Future works

- Observation of all components of synovial fluid
- Influence of viscosupplementation on friction and lubrication
- Influence of HA molecular weight on friction and lubrication



THANK YOU FOR YOUR ATTENTION

Pavel Čípek, Ing.

Pavel.Cipek@vut.cz

