# *Mechanical properties of human bone-tendon-bone grafts and human pins and screws grafts preserved by different methods and radiation-sterilized used for ligament reconstruction*

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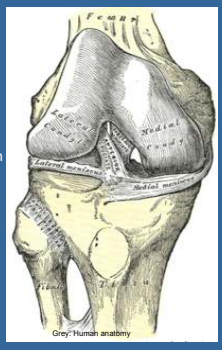
## Anterior crucial ligament (ACL)

Function:
- stabilisation of knee joint:
  - restriction of anterior displacement of tibia in relation to femur,
  - restriction of over flexion and over extention in a knee joint,
  - restriction of loopsiding or club-footing in flextion and extention,
- proprioreception,
- join femur and tibia bones.



Grey: Human anatomy

## ACL damage

Sport disciplines:
soccer,
skiing,
basketball,
rugby.

Predominate in females

## ACL damage

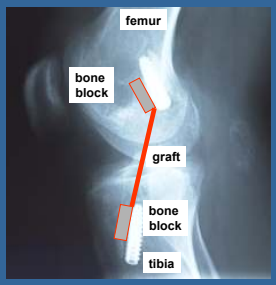
diagnostics

- Interview
- Clinical examination
- USG
- MRI
- RTG comparative of both knees

## ACL Reconstruction

- **During last 30 years**
  - over 400 surgical techniques
  - 5 conservative methods
- **During 1970-1980**
  - surgical complications - over 50%
- **During 1981-1990** (introduction of arthroscopic techniques)
  - surgical complications - 10% to 52%
- **Nowadays:**
  - indications for revision surgery - 5% to 25%

## ACL Reconstruction



## ACL Reconstruction

**Autografts**


Patellar tendon


Quadriceps tendon


Hamstring tendon

**Allografts**


Patellar tendon


Achilles tendon


A. tibialis tendon


Fascia lata

**Synthetic material**

Gore-Tex,
Dacron,
Carbon fibers,
Artelon

LAD Ligament Augmentation Device

**Tissue engineered products**

- Mesenchymal stem cells (Cooper et al., 2002)
- scafold: silk, umbilical vein, chitosan, nanofibers, collagen, PLA

## ACL Reconstruction

Patellar tendon auto- and allografts are commonly used in orthopaedic surgery for reconstruction of the anterior crucial ligaments (ACL).

Autografts are mainly used for primary reconstruction, while allografts are useful for revision surgery.

## Bone transplants

- **Biocompatible**
- **Rebuild by own patient's tissue**
- **The absence of the need for implant removal**
- **Non inflammatory response**
- **Compatible with MRI and CT**

## Tissue banking

- I - donor qualification,
- II - procurement of tissues,
- III - testing of material,
- IV - release from quarantine or disqualification,
- V - processing (sterilization),
- IV - storage,
- V - distribution,
- VI - traceability.

The risk of infectious disease transmission (donor to recipient) limits the use of fresh, frozen human tissue grafts. To avoid this risk allografts should be sterilized.

As radiation-sterilization is supposed to decrease the mechanical strength of tendon and bone tissue, it is important to establish methods of allografts preservation and sterilization resulting in their best quality and safety.

## Objective

The aim of the study was to compare the tensile strength of the central one third of human patellar tendon (as used for ACL reconstruction), preserved by different methods (deep freezing, glicerolisation or lyophilisation) and subsequently radiation-sterilized with doses of 0 (control), 25, 35, 50 or 100 kGy.

All conducted experiments were approved by the Local Ethical Committee.

## Material and methods

- 25 male cadaveric donors,
- 17-84 year old,
- Both (left and right) patella tendons were procured up to 48 hours after death,
- Bone-tendon-bone (BTB) allografts were formed and processed at the Central Tissue Bank in Warsaw according to standard procedure.

## Material and methods

- Bone-tendon-bone grafts were prepared from cadaveric human patella tendon with both patellar and tibial attachments.

## Material and methods

- BTB grafts were preserved by deep freezing, glicerolisation or lyophilisation
- From every one pair first graft was radiation-sterilized, and the second was not (control).
- 7 groups

| Conservation | Sterilization dose and condition |
|---|---|
| Freezing | 25 kGy on dry ice |
| Freezing | 35 kGy on dry ice |
| Freezing | 50 kGy on dry ice |
| Freezing | 100 kGy on dry ice |
| Freezing | 35 kGy at room temp. |
| Glicerol | 35 kGy on dry ice |
| Lyophilisation | 35 kGy at room temp. |

## Material and methods

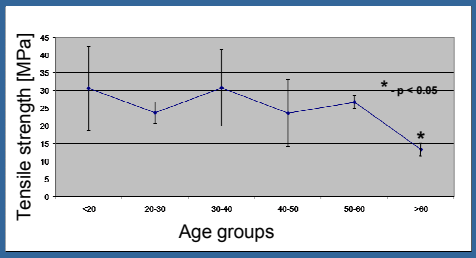
- The sterilization by electron beam accelerator was provided at the Institute of Nuclear Research at Warsaw.


## Material and methods

- To estimate mechanical properties all samples were subjected to tensile tests to failure using INSTRON system.
- Before these tests all lyophilised grafts were rehydrated.

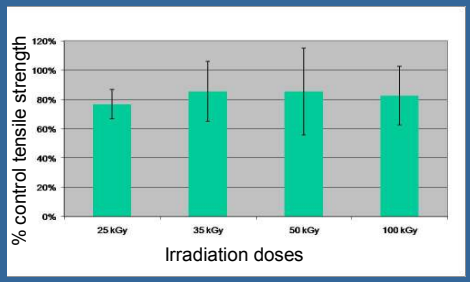
## Results

Tensile strength in MPa of frozen non-irradiated controls related to the age



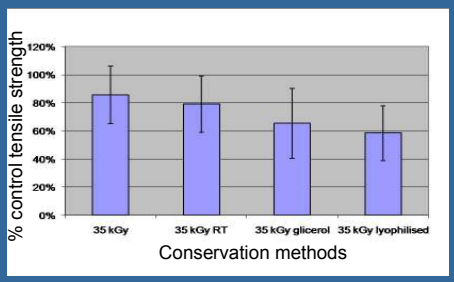
## Results

Tensile strength of frozen grafts related to non-irradiated controls




## Results

Tensile strength of grafts related to non-irradiated controls




## Allogenic fixation device

The major goal of the second study was to create a novel, based on the human cortical bone transplants, resorbable internal fixation device, shaped as screws and pins for the use in orthopaedic surgery.

## Graft fixation

**Nonresorbable (steel)**

- foreign body
- need for implant removal

**Resorbable (PLA)**

- slowly absorption without osteoinduction
- inflammatory response
- local acidification

## ACL reconstruction

**Fixation of reconstructed ligament**

Steel screws

Titanium screws

Bovine bone screws

PLA/PGA screws

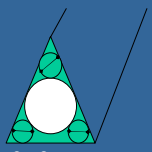
HA coated screws

## Materials and methods

- 5 male cadaveric donors,
- 18 - 46 years old (med. 28.6),
- The lower limb bones (tibia and femur) were procured up to 48 hours after death,
- and stored frozen.

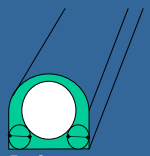
## Anatomy

After confirmation of negative serological results, cortical bone tissue used to form pins and screws.



6 - 8 mm

TIBIA



5 - 6 mm

FEMUR

## Machining processing

The physical properties of the cortical bone are enough to process mechanical treatment.

- Small pins D = 3,5 mm L = 40 mm
- Large pins D = 5-7 mm L = 40 mm
- Small screws D = 5-6 mm L = 40 mm
- Large screws D = 6-7 mm L = 40 mm

## Processing

Bone transplants were processed at the Central Tissue Bank in Warsaw according to standard procedure:

- delipidation,
- radiation-sterilisation with 35 kGy.

## Mechanical properties

All samples were subjected to mechanical tests using INTRON© system.

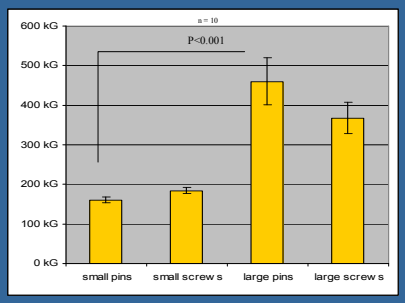
- Bending test (3 points test; small pins),
- Shear test (2 points test; screws and large pins).

Parameters assessed:

- Breaking force [kG],
- Tensile strength [MPa].

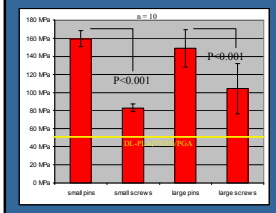
## Results

Breaking force

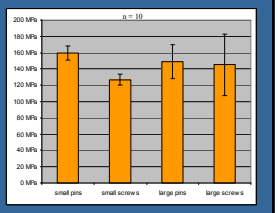



## Results

Critical tension



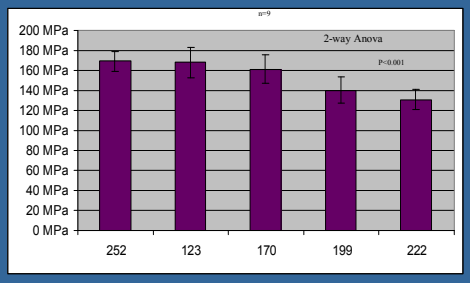

TOTAL DIAMETER



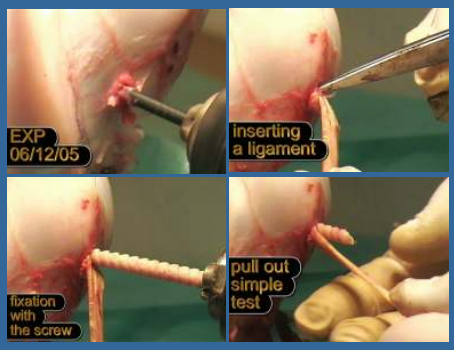

INNER CORE

## Results

Tissue quality (donor variability)




## Pull-out test




## Conclusions

- Tensile strength of frozen BTB grafts decreses in the case were tissues are retrieved from donors in the aged over 60.
- Tensile strength of radiation-sterilized grafts decreses in the group of frozen BTB grafts by approx. 20% as compared to non-irradiated controls.
  There were no signifficant dose-related differences in a range of investigated doses (25-100 kGy).
- Deep freezing seems to be the most efficient method for conservation of BTB allografts if subsequently radiation-sterilized.

## Conclusions

- The mechanical strength of pins and screws prepared from bone material is similar to the original bone material.
- Radiation dose of 35 kGy might be used for the sterilization of bone pins and screws.
- It is possible to obtain successful fixation of a ligament into bone using allogenic screws.
- The mechanical quality of bone should be checked for each donor, because it's donor depended.

## Conclusions

- Obtained results indicate that radiation-sterilized pins and screws formed from allogenic bone can be used for clinical applications offering:
  - high mechanical strength of bone fixation,
  - regenerative properties,
  - safety and superior biocompatibility.