

WORKSHOP

Tissue-based Biomarkers for Advancement of Personalized Cancer Treatment

Tissue Quality Control



Peter Riegman

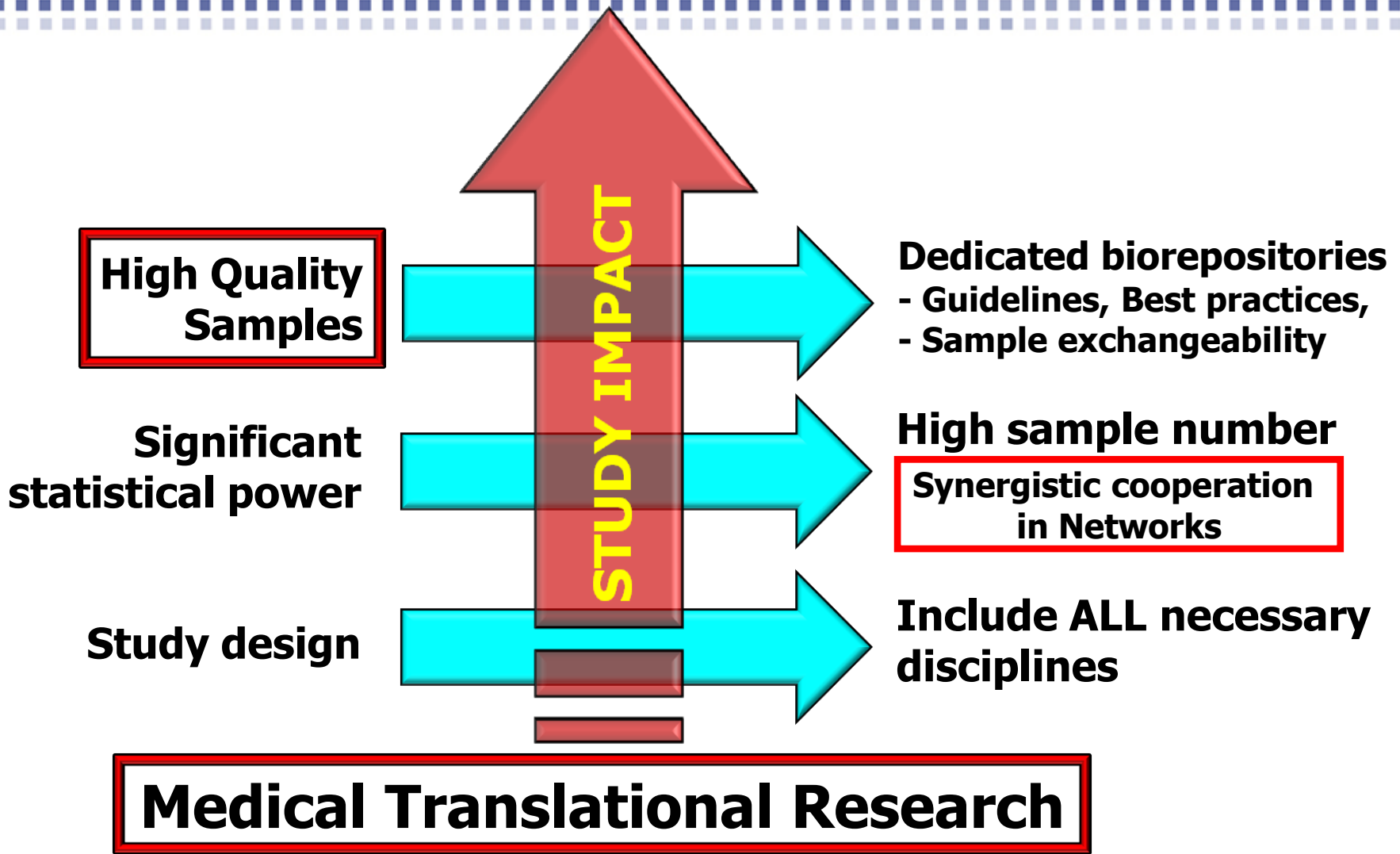


Medical University of Graz, Austria, 28th – 29th March 2014

Erasmus MC



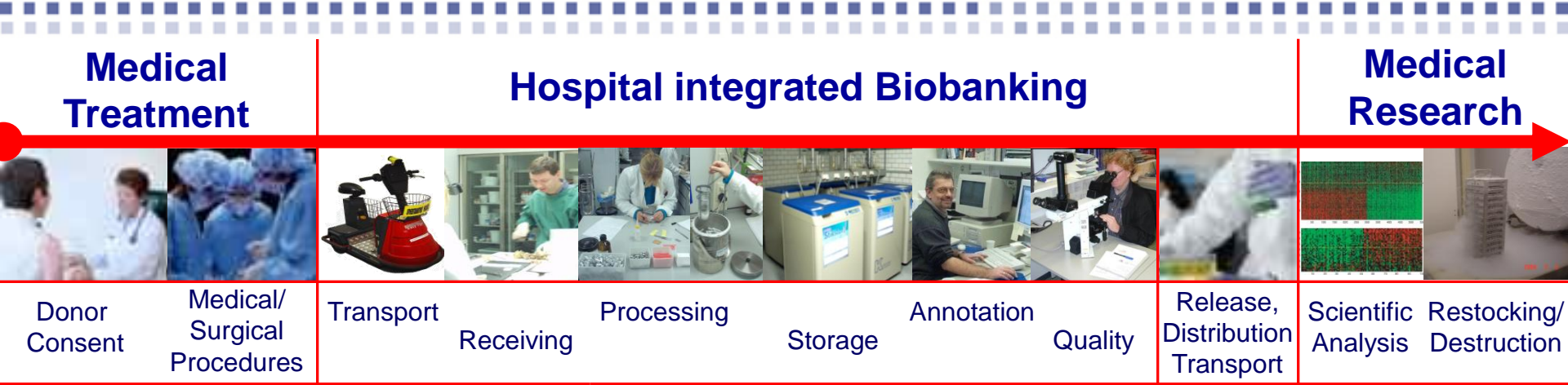
Innovation of Patient Care



Tissues used for plethora of techniques



Quality of tissue samples



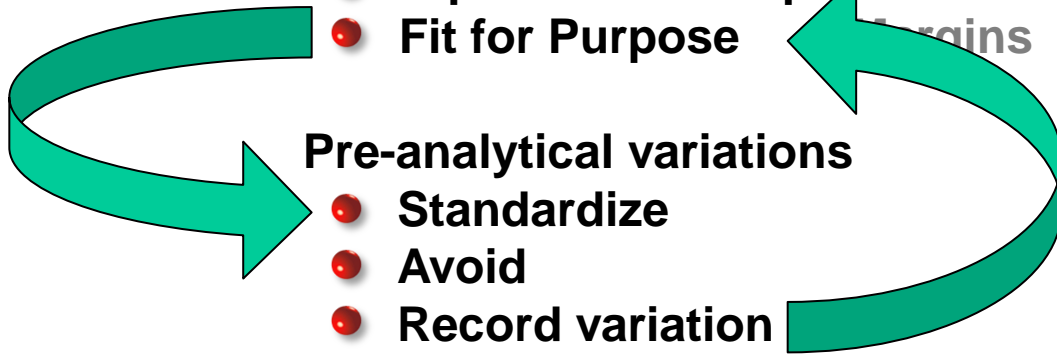
30 minutes

Pre-analytical phase

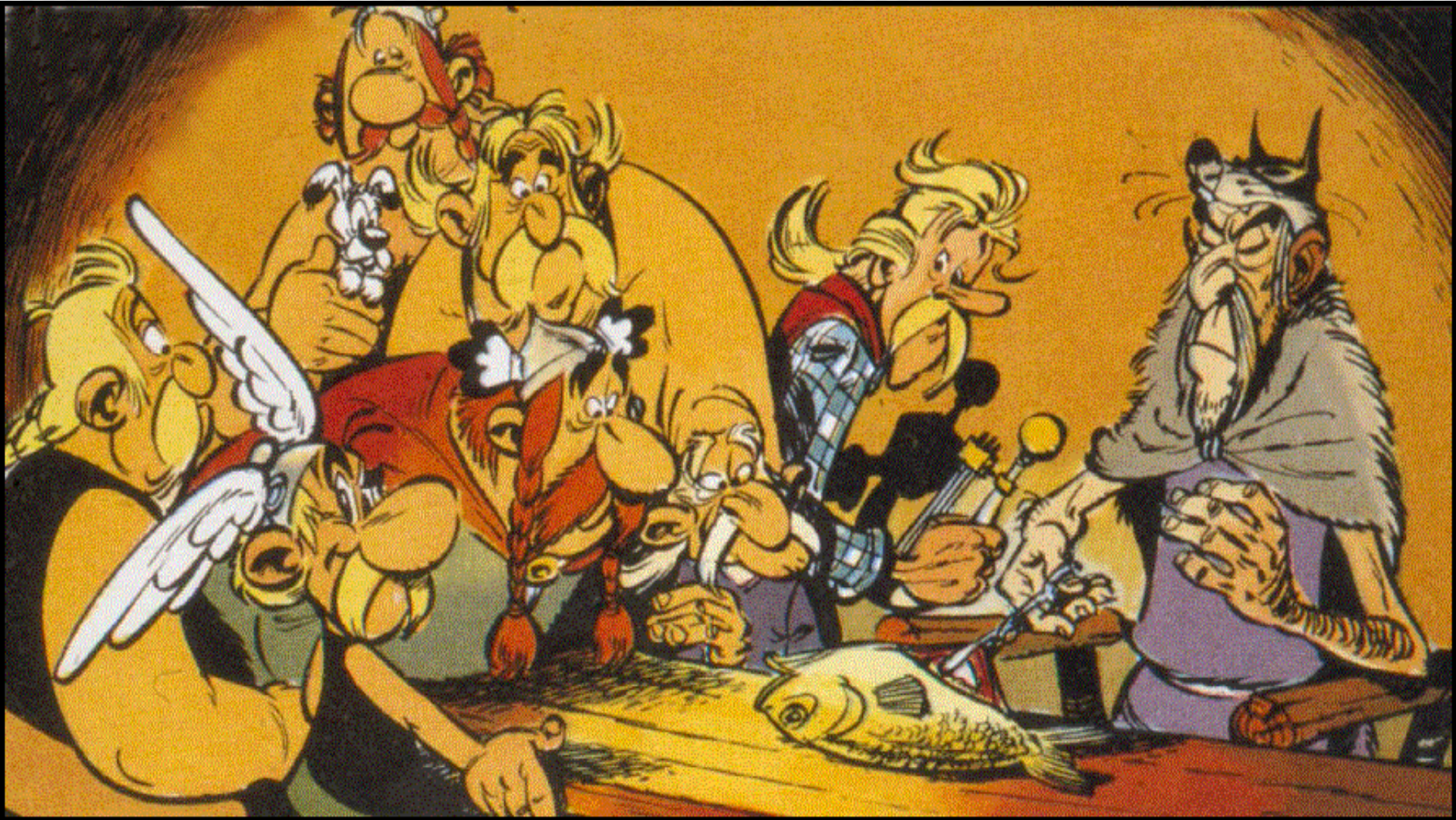
- Equal for all samples - Variable results
- Fit for Purpose

Pre-analytical variations

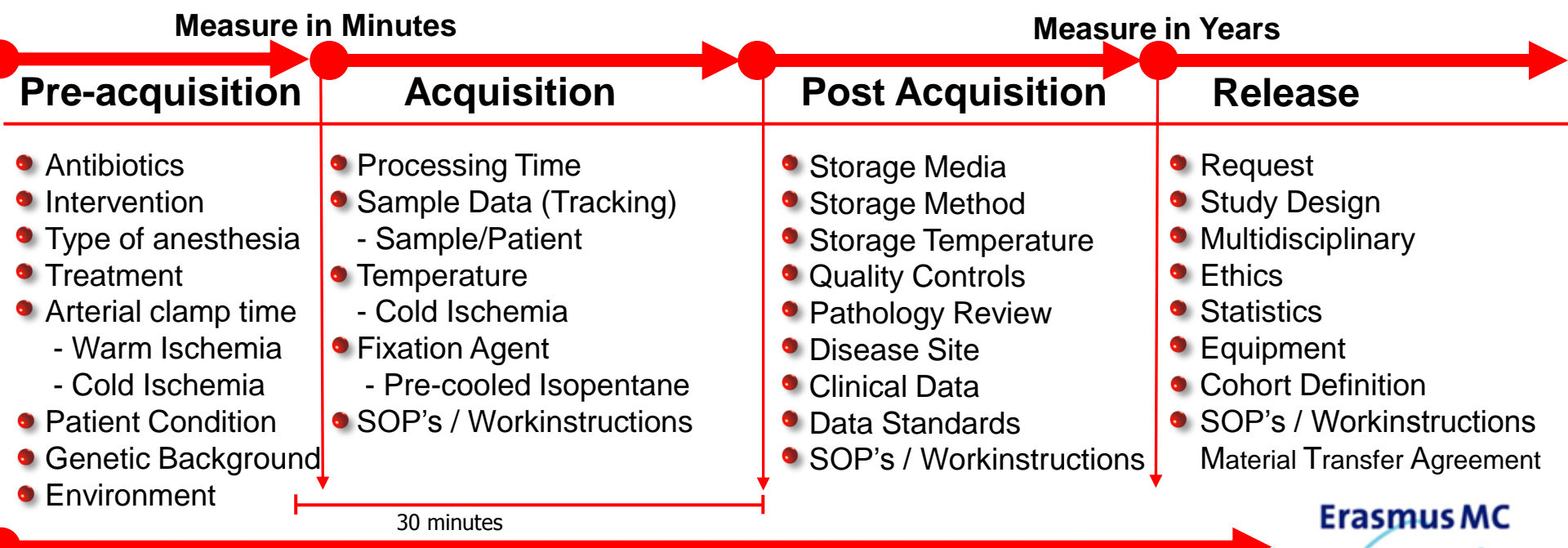
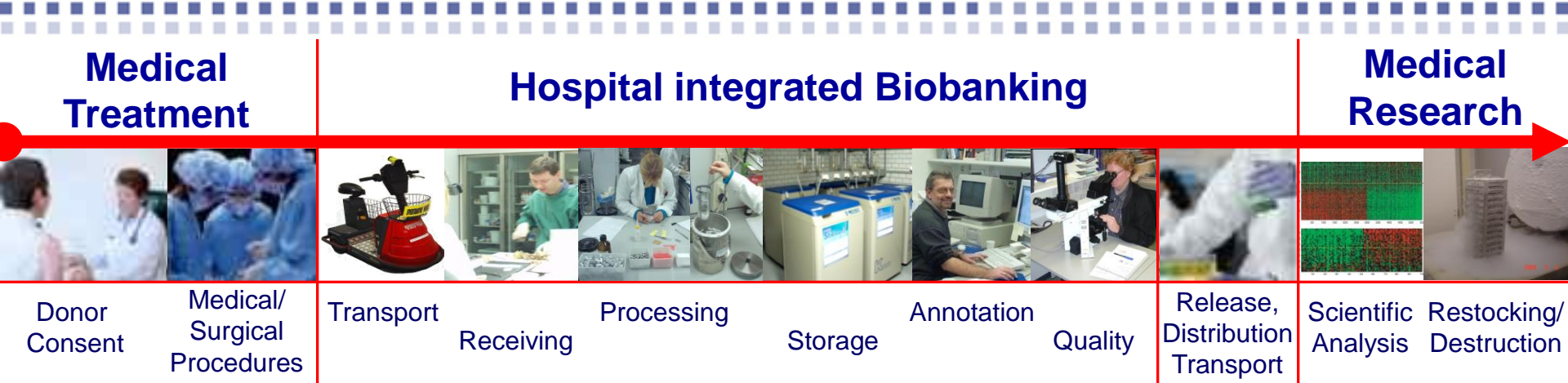
- Standardize
- Avoid
- Record variation



Reproducible conclusions ?



Quality of tissue samples



Pre-analytical phase

Variables for FFPE

- **Fixation time** – Over fixation
- **Fixative** – Concentration / Ratio
- **Treatment** – Drugs, warm ischemia
- **Patient** – Condition, Genetic background
- **Lag time** – Transport



**Tissue Micro Array, Immunohistochemistry
DNA, RNA mi-RNA and protein isolation**

Poor Molecular Quality

Variables for Frozen Tissue

- **Treatment** – Drugs, warm ischemia
- **Patient** – Condition, Genetic background
- **Lag time** – Transport

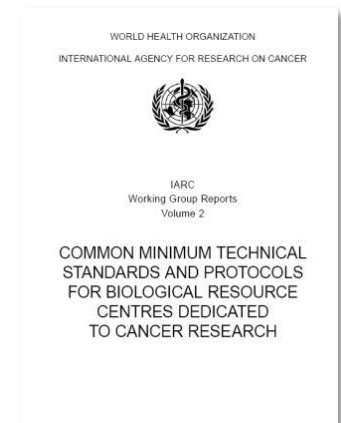
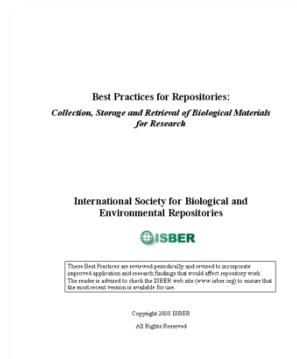
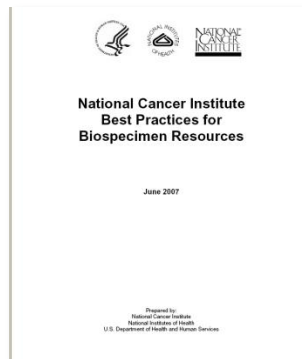


**Genomics, Transcriptomics, Proteomics,
Metabelomics**

High Molecular Quality

Guidelines / Best Practices Biobanking

- Guidelines / Best practices, ISBER / NCI / OECD
- IARC Common Minimal Standards
- OECD-TuBaFrost Standardization for Frozen Tissue
- Role of the pathologist in tissue banking:
European Consensus Expert Group Report (BBMRI)



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**Tissue removal from patient until freezing
30 minutes lag time**



**Conservation of morphology
Snap Freezing**

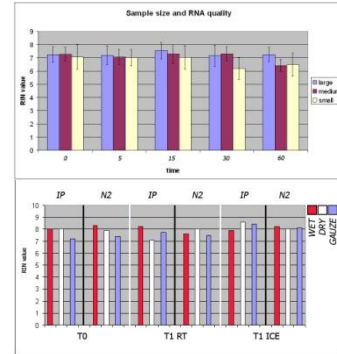
- Pre-cooled Isopentane -150°C



Evidence Based Biobanking RNA integrity / Expression

- Influence sample size:**

- Significant in medium (1cm³) more pronounced in small samples (0,5 cm³) RNA on RIN and expression



- Transport, freezing method and delay:**

- Morphology & expression altered in 0.9% salt solution
- RIN, expression and morphology optimal dry or moist gauze and ice

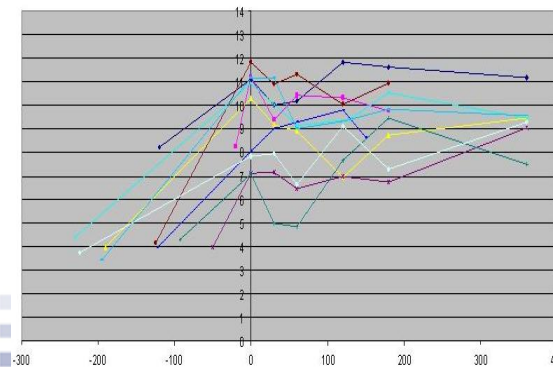
- Tissue type and storage type:**

- QC → 5% RIN < 6.5
- Fat, Necrotic, Fibrous and Pancreatic tissue
- Long time storage -80°C no differences
- No apparent differences LN2 and -80°C freezers



- Warm Ischemic phase:**

- Small % significant change RNA expression
Liver wedge vs. needle biopsies



Frozen tissue QA and QC

- **Protocols for collection and storage (ISO 15189)**

- **Review of 1% of new cases annually**

Cases selected at random (Rare cases are re-chosen)

- Sample identification (bar code, labelling)
- Confirm diagnosis with stained HE sections
- RNA extraction for RNA quality assessment



- **Protocols for collection and storage (ISO 15189)**

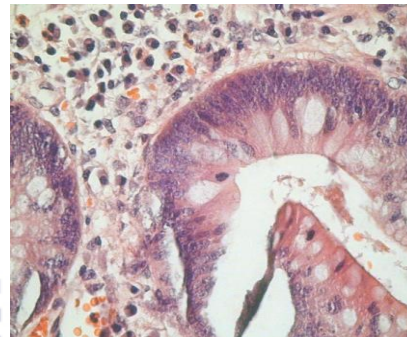
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Cases selected at random (Rare cases are re-chosen)

- Sample identification (bar code, labelling)
- Confirm diagnosis with stained HE sections
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- **QC Fast diagnostics by comparison the known clinical diagnosis with the H&E stained slide**

Also produced for recording virtual Microscopic digital images for the Biobank database.



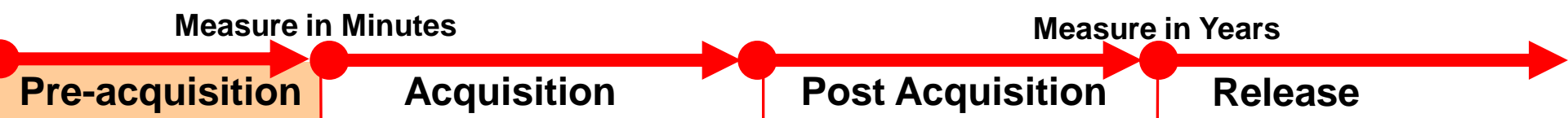
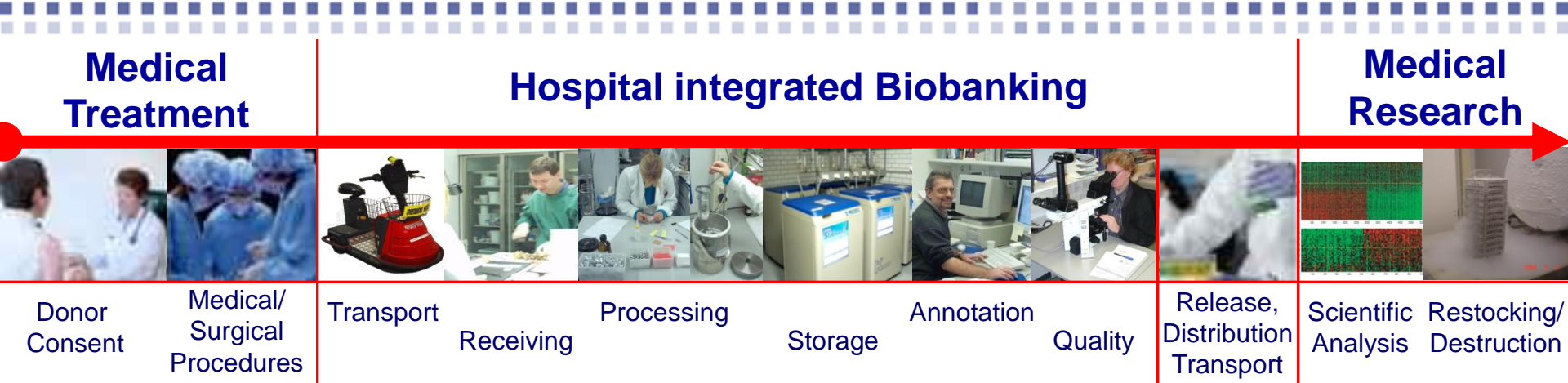
ISO

- Biobank personnel and users are both aware of sample quality
- Better support can be given in case of bad results

Post Audit Dip



Quality of tissue samples



QA & QC

SOP's & Workinstructions

ISO 15189:2007

- Antibiotics
- Intervention
- Type of anesthesia
- Treatment
- Arterial clamp time
 - Warm Ischemia
 - Cold Ischemia
- Patient Condition
- Genetic Background
- Environment

- Processing Time
- Sample Data (Tracking)
 - Sample/Patient
 - Temperature
 - Fixation Agent
 - Pre-cooling
- SOP's / Workinstructions

- Storage Media
- Storage Method
- Storage Temperature
- Quality Controls
- Disease Site
- Data Standards
- SOP's / Workinstructions

- Request
- Study Design
- Multidisciplinary
- Ethics
- Statistics
- Equipment
- Cohort Definition
- SOP's / Workinstructions
- Material Transfer Agreement

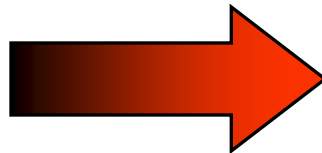
30 minutes

Pre-analytical phase

- **Bad Samples**
 - Bad Quality
 - Incomparable quality
- **Bad Research**
 - Bad study design
 - Bad experiments

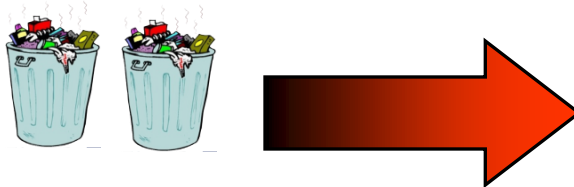
**Institutional
research projects**

**DEDICATED
PERSONNEL**



- ~~Bad Samples~~
 - ~~Bad Quality~~
 - Incomparable quality
- ~~Bad Research~~
 - Bad study design
 - Bad experiments

~~Institutional~~
research projects



~~Bad Samples~~

~~- Bad Quality~~

- Incomparable quality

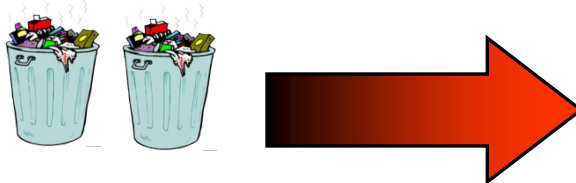
Bad Research

- Bad study design

- Bad experiments

Multi center research projects

- Record pre-acquisition phase
- External Quality Assurance
- Evidence Based Biobanking
- Proficiency testing



Study design

Study design:

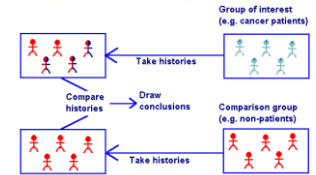
- Feasibility
- Strategy

Disciplines:

- Molecular Biologists
- Epidemiologists
- Pathologists
- Statisticians
- Clinicians
- Pharmacologists
- Bio-Informatics
- Clinical genetics

Cohort selection:

- Cohort Type:
 - Longitudinal population-based cohorts
 - Clinical case/control studies
 - Population isolate studies
 - Twin registries
- Amount and type of samples



Input -> Output:

- Unexpected Bias
- Analysis and expected results
- Test Sensitivity

Cooperation

**Biobank
Network
Performance**

Access rules

**Ethical Legal
& Social Issues**

**Data
Interconnectivity**

Sample Quality

**Sample
Exchangeability**

EQA

Metadata Pre-acquisition phase

Certification / Accreditation
Proficiency Testing

Quality Assurance
Quality Control

SOP's in document control system

Standard Operating Procedures (Guidelines)



European Committee for Standardization
Comité Européen de Normalisation
Europäisches Komitee für Normung

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WP10 Biobanking

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