WS 1.3. Gender in basic research, Febr 2015 , Berlin

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<th>Participants – look at website</th>
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<tr>
<td>Arnal</td>
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<td>Brunelleschi</td>
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WS 1.3 Rationale for basic research on S&G differences

- Women and men differ in many diseases

- Understanding S&G differences may contribute to improvements in therapy

- Are there common S&G specific mechanisms that play a role in many diseases?
How to study sex and gender in clinical and basic research?

- NIH expects that sex as a biological variable will be factored into research designs, analyses, and reporting in vertebrate animal and human studies.

- Common S&G mechanism in all diseases

- Asthma
- Lung cancer
- Rheumatic diseases
- Autoimmune diseases
- Myocardial infarction
- Stroke
- Heart failure
- Renal diseases
### WS 1.3. Molecular and cellular mechanisms contributing to sex differences

**Genetic and Epigenetic processes**

- Sex specific genetic background (Erdmann, Kararigas)
- Sex differences in histone modifications in rat models (Mieth/Hübner)
- Sex differences in the anti-inflammatory response of cells stressed by e.g. cigarette smoke. (Bunelleschi)
- Sex chromosomes and behaviour (Majdic)
- Epigenetic processes are the key to all sex differences in health and disease? (Junien)

**Sex, sex hormones and cell function**

- Cell sex and determination of cell survival and death. (Malorni)
- Neuron interactions-sex differences of factors reducing mental health? (Reynolds)
- Sex differences in endothelial function (Randall)
- Mitochondria and sex differences in cytoprotection.(Ventura-Clapier)
- Sex hormones and reactive fibrosis (Dworazek)

**Translational**

- Lipoprotein and glucose metabolism - effects of cycle and hormone status? (Hofmann)
- Effect of testosterone on cardiomyocyte function (Giannetta/Lenzi)
- Sex hormone receptor modifications. (Arnal)

**Drug development**

- Sex differences in drug targets (Fliegner)

**Output 1**: Review paper of the consensus statements on sex differences in basic biomedical research and preclinical drug development
Percent of GWAS Papers with X Chromosome Analysis

How to Include Chromosome X in Your Genome-Wide Association Study

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- X Chromosome still underrepresented in GWAS
- Advanced analysis tools are available for X Chromosomal data
- Including X Chromosome will provide new insights into pathogenesis of cardiovascular disease
Dissemination is important ...


- **X chromosomal variation is associated with slow progression to AIDS in HIV-1-infected women.**


- AIDS has changed from a mostly male-specific health problem to one that predominantly affects females. Although sex differences in HIV-1 susceptibility are beyond doubt, the extent to which sex affects the onset and progression of AIDS has remained elusive. Here, we provide evidence for an influence of X chromosomal variation on the course of retroviral infection, both in HIV-1-infected patients and in the rhesus macaque model of AIDS. A two-stage, microsatellite-based GWAS of SIV-infected monkeys revealed MHC class I markers and a hitherto-unknown X chromosomal locus as being associated with a nominal score measuring progression to AIDS (Fisher's exact p < 10(-6)). The X chromosomal association was subsequently confirmed in HIV-1-infected patients with published SNP genotype data. SNP rs5968255, located at human Xq21.1 in a conserved sequence element near the RPS6KA6 and CYLC1 genes, was identified as a significant genetic determinant of disease progression in females (ANOVA p = 8.8 x 10(-5)), but not in males (p = 0.19). Heterozygous female carriers of the C allele showed significantly slower CD4 cell decline and a lower viral load at set point than TT homozygous females and than males. Inspection of HapMap revealed that the CT genotype is significantly more frequent among Asians than among Europeans or Africans. Our results suggest that, in addition to the individual innate and adaptive immunity status, sex-linked genetic variation impacts upon the rate of progression to AIDS. Elucidating the mechanisms underlying this sex-specific effect will promote the development of antiretroviral therapies with high efficacy in both sexes.
Cell sex – male and female cells differ in mechanisms of cell death

Endogenous defences:
Antioxidants,
Metabolic responses, e.g. fatty acids mobilization
ER modulation

Exogenous agents:
Antioxidants,
Nutrients,
Estrogens
Epigenetically active therapies

Stressors (e.g. inflammatory cytokines, oxidative stress)
Nutritional/Metabolic stress

Mitochondrion
Nucleus
Lysosome

ROS increase
Cytoskeletal changes
Morphological changes

Mitochondrial alterations

Pro-apoptotic agents (drugs, radiation)

Senescence and autophagy
Cell survival
(increased autophagic cytoprotection)

Increased apoptotic markers
Cell death (Apoptosis)

XX cells
XY cells

V Malorni et al
Sex differences in cardiac fibrosis in humans

- Patients with aortic stenosis
  (Villari et al., 1995; Petrov & Dworatzek et al., 2014)

- Patients with coronary artery disease
  (Campbell et al., 2011)

- Patients with atherosclerosis
  (Ambale Venkatesh et al., 2014)

- Healthy persons

Men develop more easily fibrosis than women

Petrov & Dworatzek et al., JACC Imaging 2014

This project the European Gender Medicine Network (EUGenMed) has received funding from the European Union’s Seventh Framework Programme for research, technological development and demonstration under grant agreement No 602050.
E2 decreases collagen III in females and increases it in male rat cardiac fibroblasts.

Mean ± SEM, n=4 carried out in triplicates; **p≤0.01, *p≤0.05 comparison was performed using unpaired t-test.
Sex specific modulation of fibrosis

Cardiac fibroblasts

E₂

ERβ

ERα

TGF-β1

Fibrosis-associated genes:
Col I/III, Periostin, MMPs, SMADs, TGFβ-1

transcription

miRNA

collagen

Diseases

Rheumatoid arthritis

Systemic sclerosis

Heart failure

Renal diseases

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Models for S&G research are complicated and costly

- Sex and gender research need very specific and complicated animal models
- Research funding organisations must support the development of these models by specific calls for gender research projects
- Examples:
  - transgenic mouse models, knock-out or overexpression of estrogen or androgen receptors
  - 4 core genotypes – generating **XX males** or **XY females**

![Diagram showing methods and genetic models](image)

Coop J F Jaisser, Inserm
Diabetes and lipid levels

High density lipoprotein (HDL)-based drug therapy for women with diabetes to promote regression of increased cardiovascular disease (CVD)

- High rates of diabetes are of particular concern because of the enhanced elevation in CVD risk documented in women with diabetes compared to men with diabetes
- Relation between diabetes and lipid levels
- Oxidation of HDL, different HDL function in women and men


S. Hofmann, Munchen
S&G differences at cellular level are relevant for many diseases

S&G differences affect basal cellular functions, outside sexual organs

They are relevant for many different diseases

Studies need specific and complicated tools
  e.g. cross sex hormone receptor modified mice with disease prone mice

Translational approaches are promising
  Diabetes
  Heart failure
Conclusion

- Sex and SH hormones affect basal cellular physiology
- This is important for many organs and diseases
- Therefore S&G differences must be studied in as many disease models as possible – S&G as cross sectional element
- ...and in a disease independent manner - in an own discipline, with own methods, tools, research questions

This leads to own research questions, hypothesis, methods, training and career opportunities

It needs own specific calls
Outcomes

- Publication: Gender in Basic research, V Regitz-Zagrosek et al, in preparation
- Contributions to policy briefs on CVD, Stroke, Diabetes, Asthma and lung cancer
- Slide set at EUGenMed homepage

Steps towards implementation

- Contributions to ESC meeting 2015 London; a sessions with basic research gender topics
- Planning sessions at congress of the International Society for Gender Medicine in Berlin, Sept 2015 (www.igmcongress.com/): epigenetic mechanisms in sex differences, sex differences in cells,
Examples: Sex & G specific mechanisms affecting many disease entities

- Female sex/E2 affect fibrosis differently in women and men:
  - HF, MI, Rheumatic diseases

- Calcium handling
  - E controls macrophage phentoytypes – proinflammatory M1 in men, antiinflammatory M2 in women – and autoimmunity.
    - Relevant for infections and septic shock, rheumatic arthitis, thyroid disease, lupus, systemic sclerosis, myocarditis, heart failure, healing of MI, transplant rejection

- Female sex/E2 favor autophagy; male sex necrosis and apoptosis, relevant for heart failure, MI, ........
  - SH modulate mitochondrial function differently in F and m, relevant for HF, mitochon diseases (reye syndrome, kearns sayre,....)
1. Gender as an independent research area

1. Establish Gender as an independent research area:
   Like: metabolism, nuclear receptors, hormones

Defining gender medicine as a research area means:
   own nomenclature, definitions, research questions, methodology, textbooks, topics,
   thematic calls, review panels, sessions, journals, institutes, career opportunities

In contrast/ in addition to:
   Gender as a cross-sectional topic that is important in all areas
1. Sex Chromosome Effects
Effects of XX vs. XY sex chromosomes not mediated by the gonads

2. Organizational Gonadal Hormone Effects
PERMANENT
Gonadal hormone-induced irreversible commitment of a tissue to a masculine or feminine phenotype.

3. Activational Gonadal Hormone Effects
REVERSIBLE
Sex differences in traits that are abolished by gonadectomy in adulthood