Sex differences in Alzheimer's disease: what we know and what we do

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Sex differences — **Biological**

Interaction

Gender differences — **Societal**

Behavior

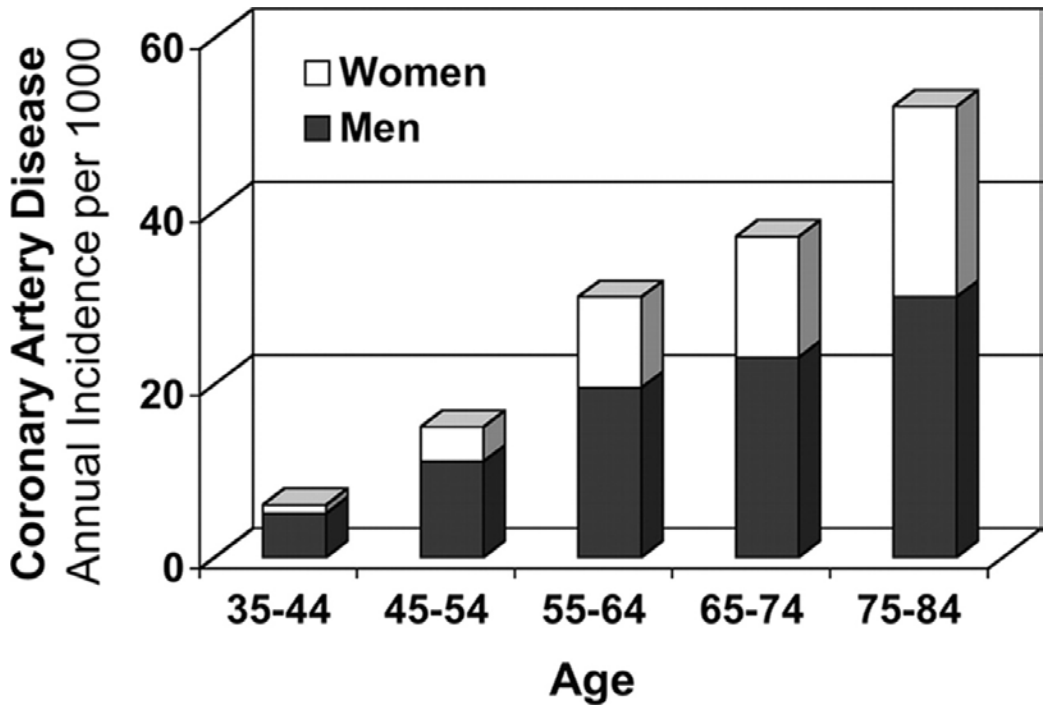
The biological purpose of these differences is: **Reproduction**

So there is male and female typical **behavior**: Are there male typical and female typical **diseases**???

Many diseases affect men and women differently

Sex is a biological factor in health and disease

Most striking example: **Cardiovascular disease**



Clear sex differences in disease risk between men and women

- Affects more males than females at reproductive age
- Different symptoms
- Different response to treatments
- Very little is still known about these sex differences

Meyer MR et al,. Hypertension, 2006

Despite obvious sex differences, why do we know so little about these?

Why are these not studied more?

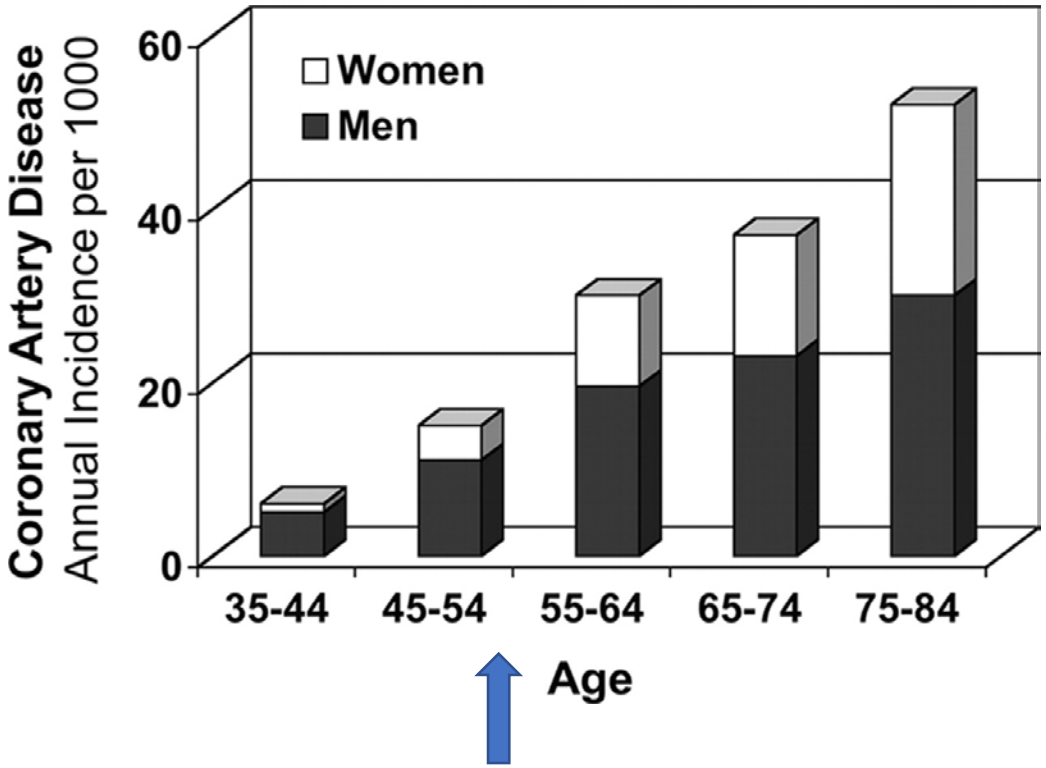
Sex and gender differences are understudied

Adds cost and time to the research project!!

- If mostly males or females are affected why study sex differences?
- All trials and treatments need to double - expensive
- The scope of the *in vitro* model need to include XX and XY cells
- Epidemiological and clinical studies would need to be stratified by sex
  – Reduces sample sizes with potential to give inconclusive results
- What about XY chromosomal aberration genotypes, or transgenders??
- How to incorporate gender aspects in the research project?

Sex as a biological factor in health and disease

Most striking example: **Cardiovascular disease**



Clear sex differences in disease risk between men and women

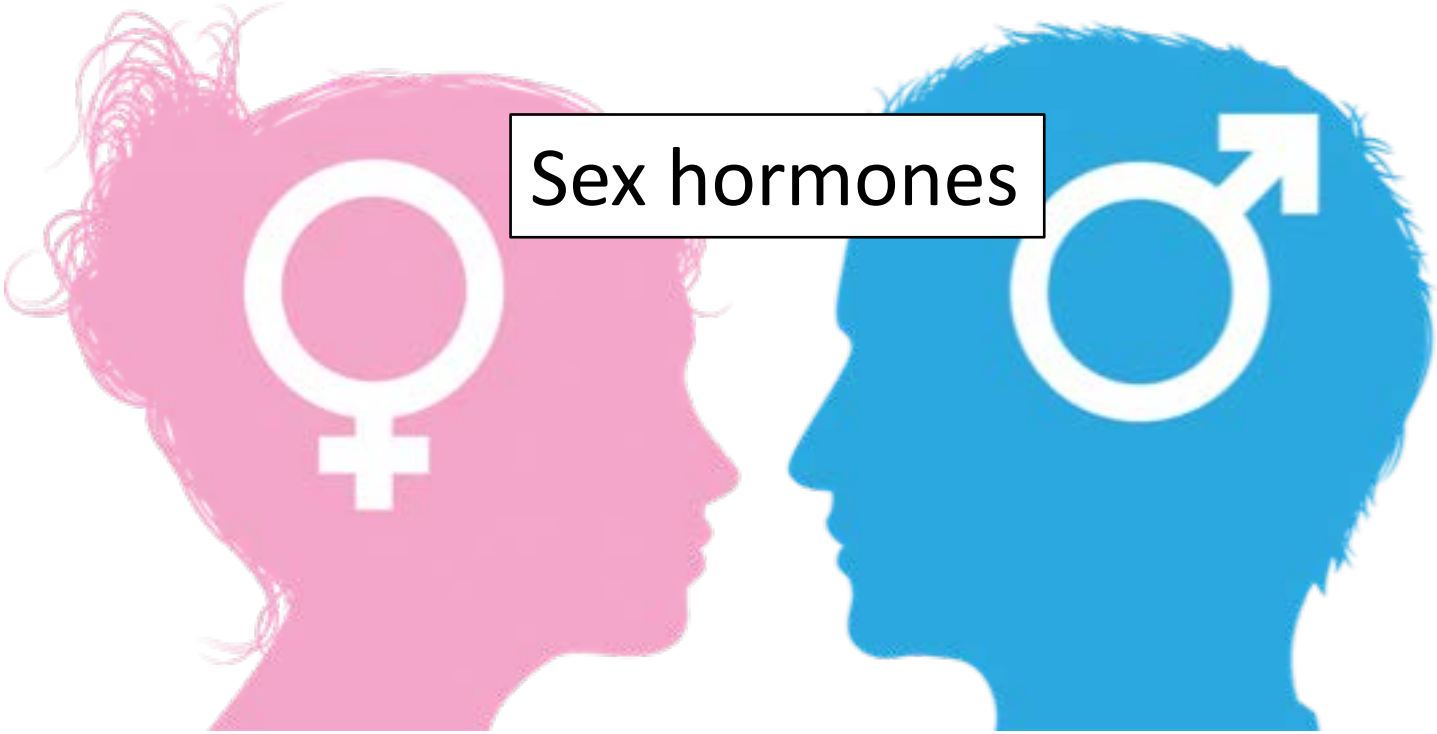
- Affects more males than females at reproductive age
- Different symptoms
- Different response to treatments

- **Differences are lost to a large extent after menopause**
- **Sex hormones are likely involved**
- **In fact women are at higher risk of dying of a cardiovascular event than men**

Meyer MR et al,. Hypertension, 2006

What about sex differences in neurological diseases?

Well, we do know that there are clear sex and gender differences in behavior



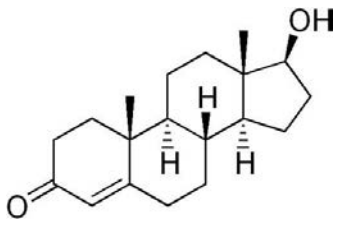
Is this because male and females brains are different?

Sex hormones

SRY gene on the Y chromosome (XY)

Development of male testes

Production of testosterone



Testosterone

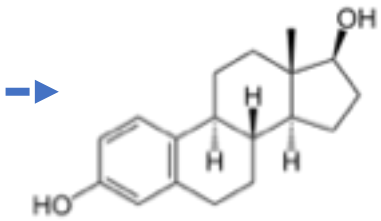
Aromatase enzyme

Androgen receptor

Lack of *SRY* gene (XX)

Default development of ovaries

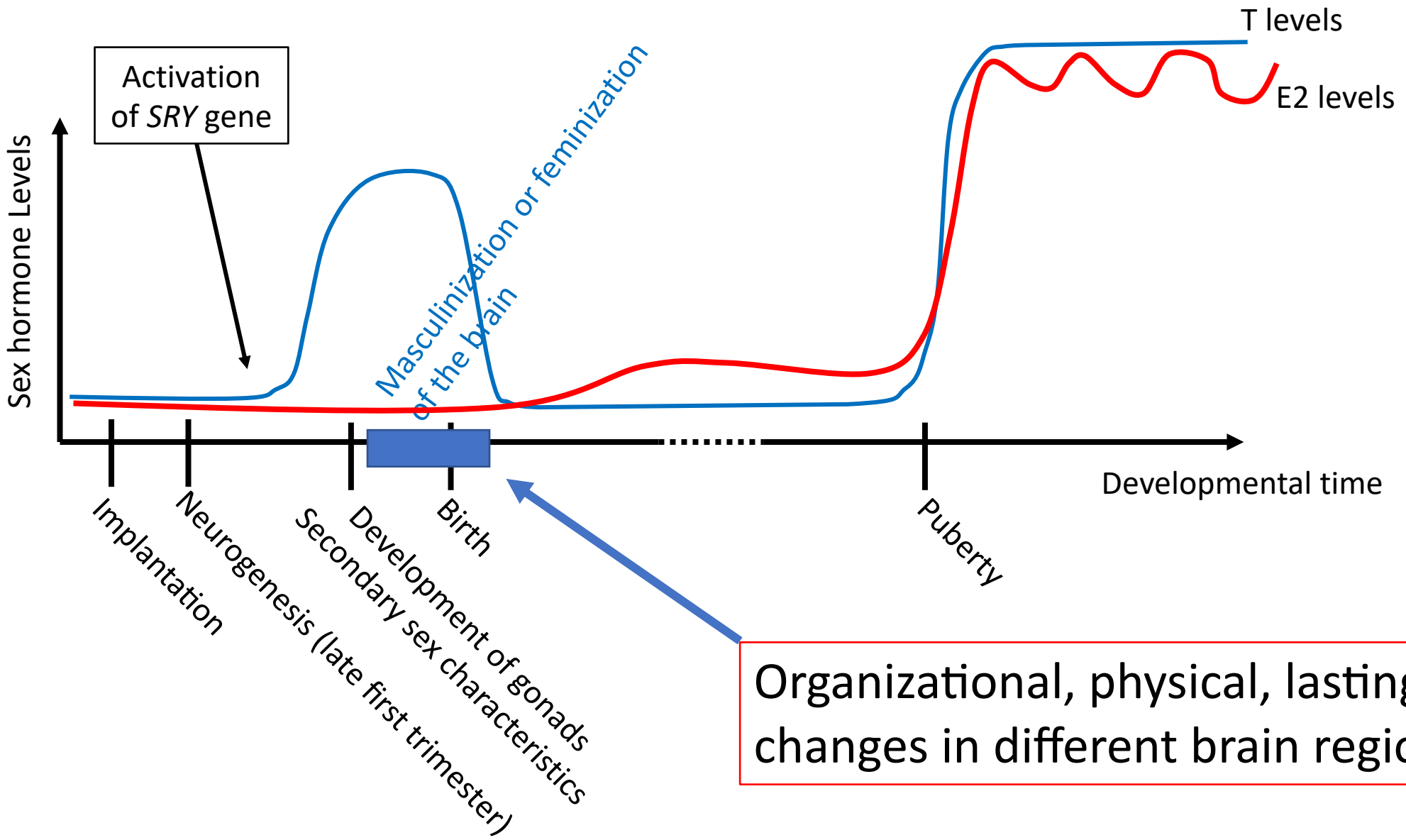
Production of estrogen



Estrogen
(Estradiol, E2)

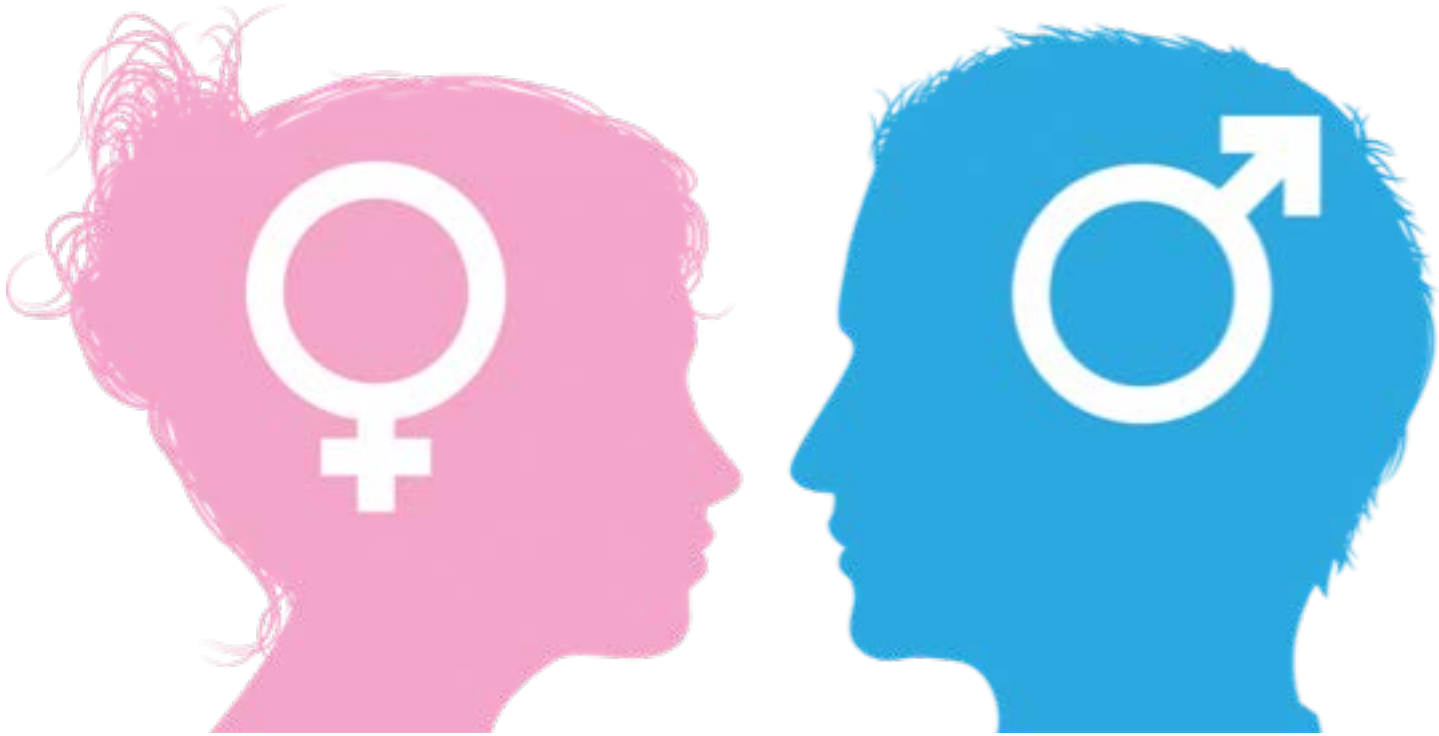
Estrogen receptor alpha (ERα)
Estrogen receptor beta (ERβ)
GPER1

Estrogen and testosterone in neurodevelopment

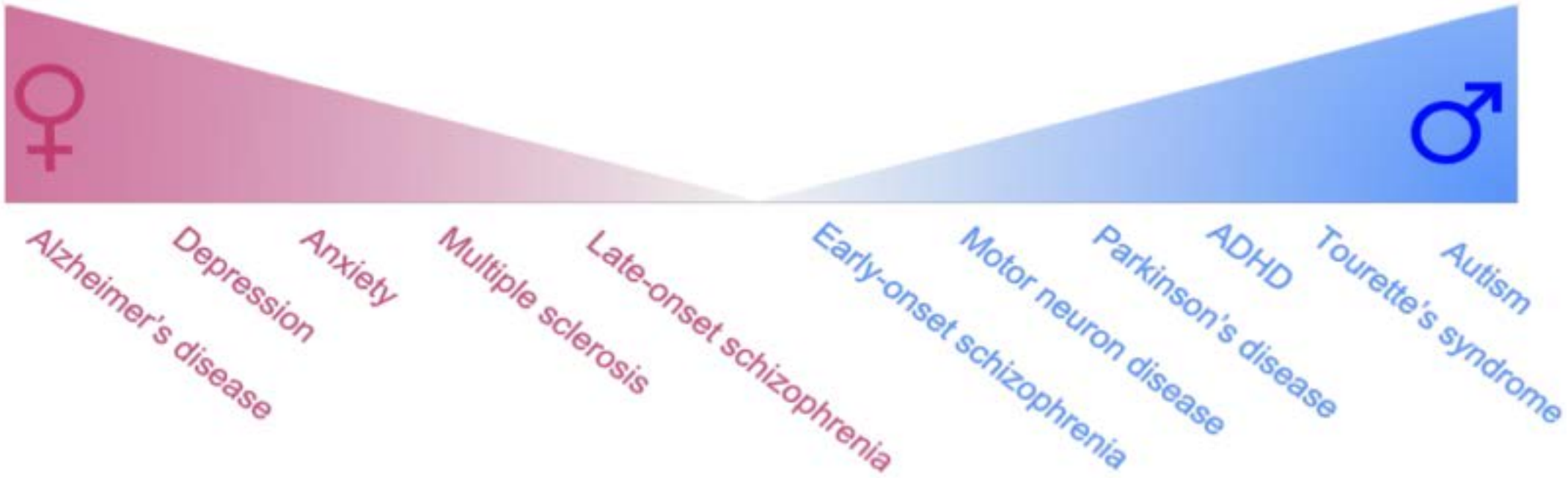


Organizational, physical, lasting changes in different brain regions

So, on a neurosignalling level there are significant sex differences between the male and female brain

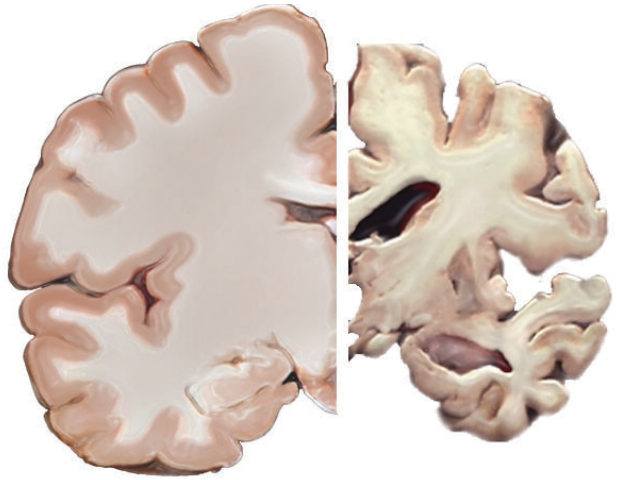


What about sex differences in neurological diseases?



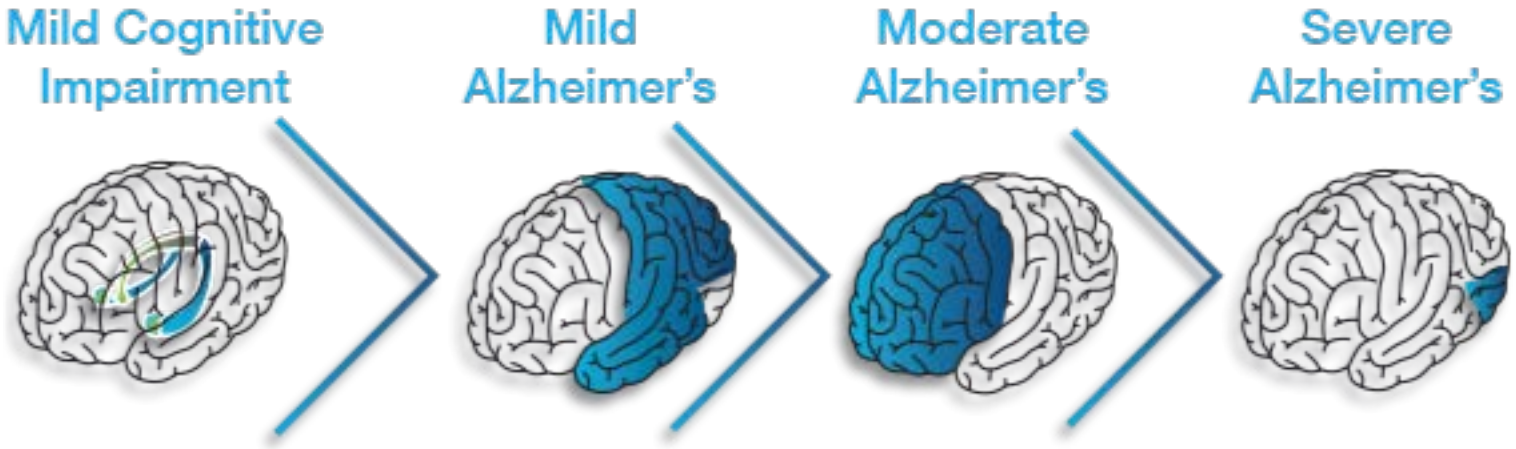
Pinares-Garcia et al, *Brain Sci*., 2018

Alzheimer's disease (AD)

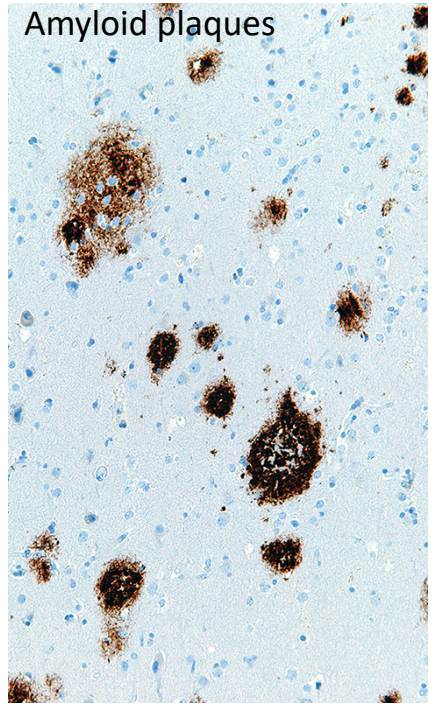


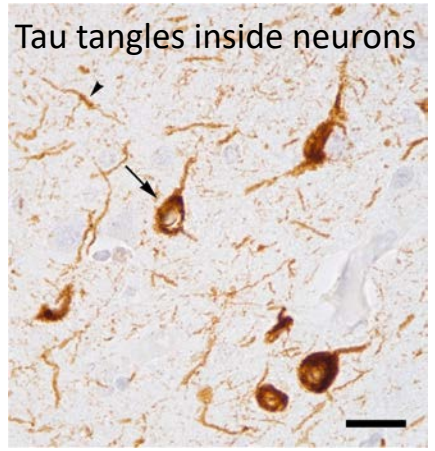
- Most common type of dementia
  - 50 million diagnosed cases, ca 60-70% of all dementia
  - 11% of people aged 65+ have AD
  - 32% of people aged 85+ have AD
- Cause of AD is unknown, except for <5% of cases with known genetic mutations (familial AD)
- > 95% of AD cases are sporadic, where ca 70% of all cases appear to be inherited
- Complex interaction between genes, disease history and environment
- Characterized by misfolded amyloid beta depositions in and around synapses (amyloid plaques) that leads to intraneuronal misfolded Tau-tangles inside neurons, extensive neuroinflammation and neuronal death
- No cure!

Alzheimer's disease progression

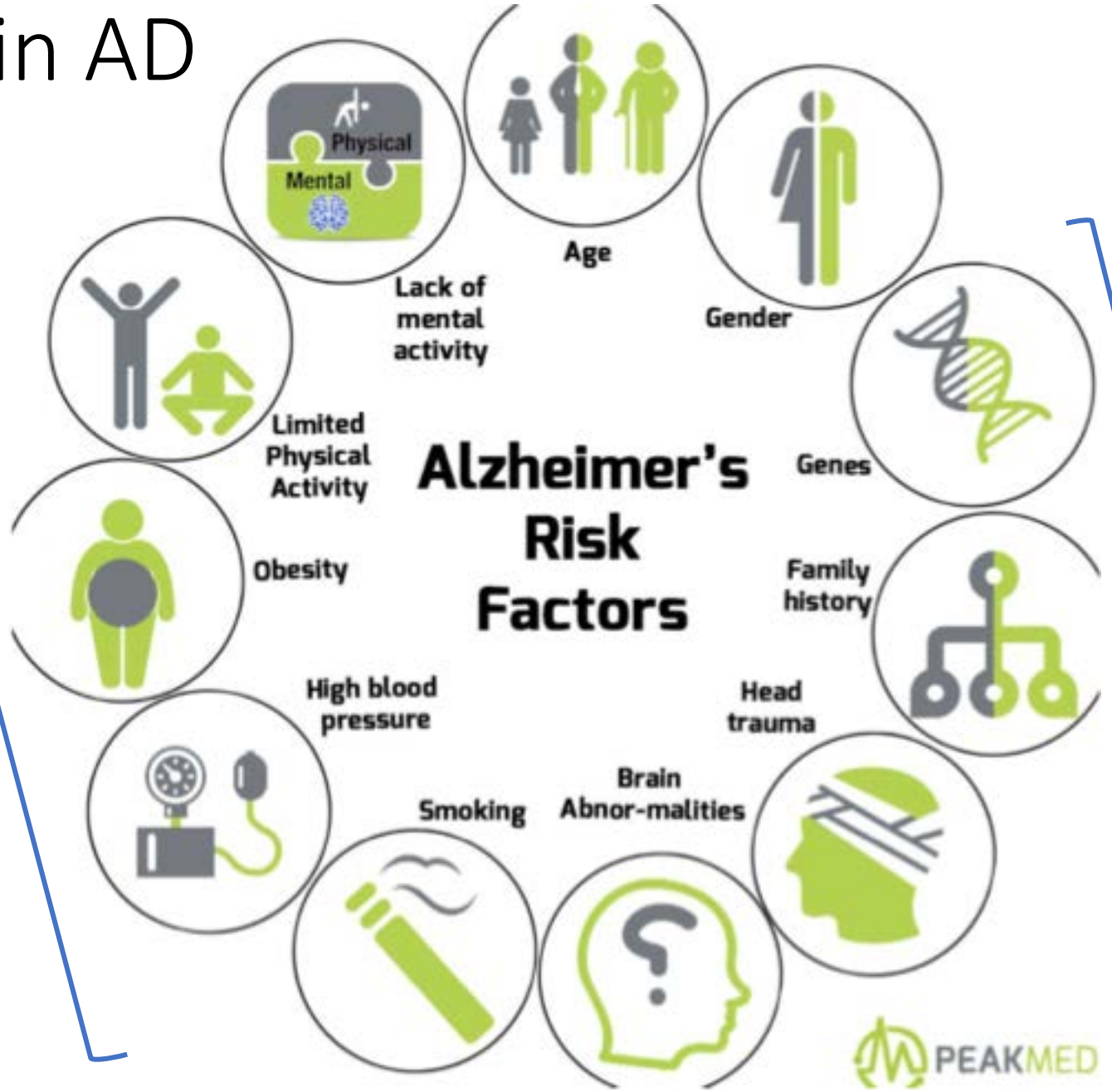


Amyloid pathology	++	++	+++	+++
Tau pathology	(+)	+	++	+++
Neuroinflammation	+	++	+++	+++





Risk factors in AD

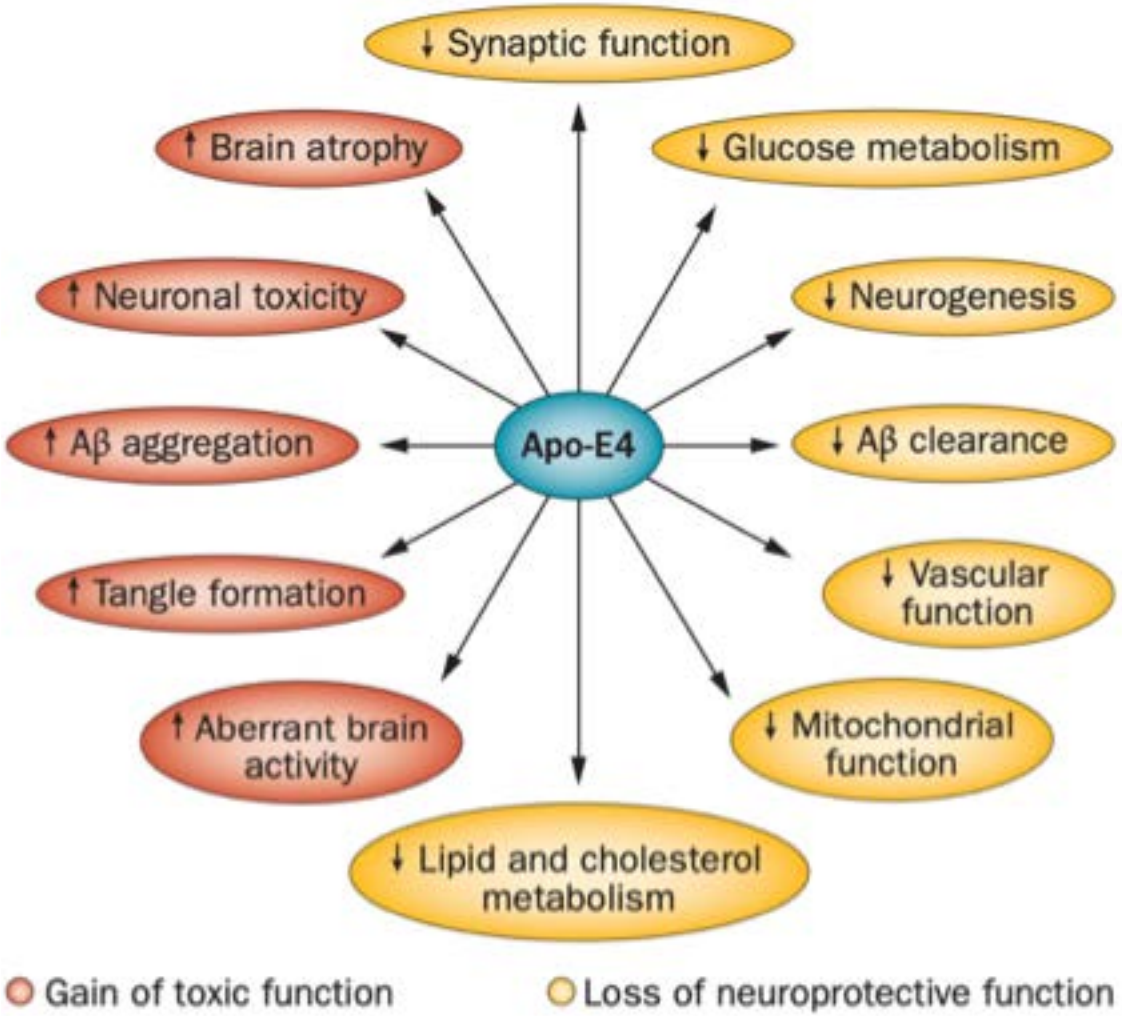


(Cardio-) Metabolic disorders

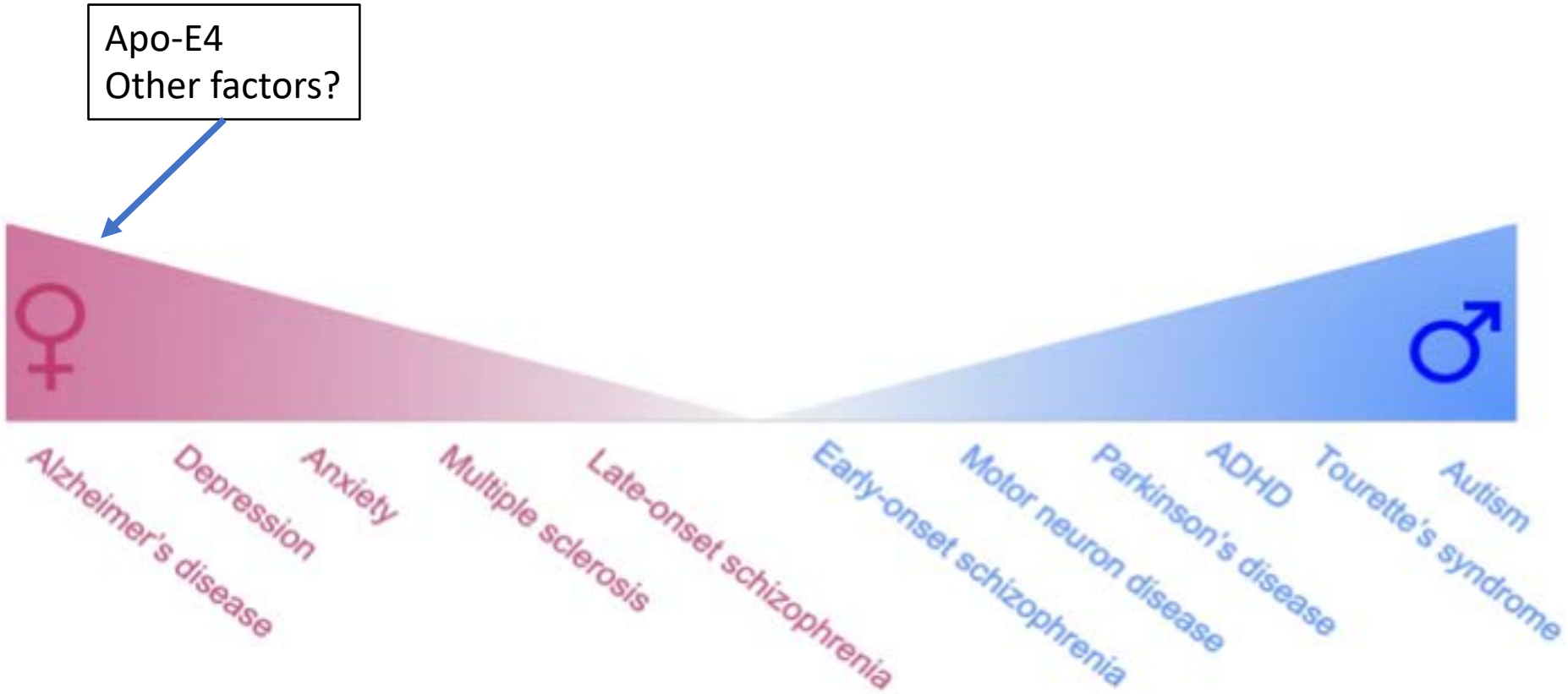
APOE

Risk factors in AD: Apo-E4

- *APOE4* allele status is the most common genetic risk factor for *sporadic* AD (14%)
- Apo-E is mainly expressed in astrocytes in the brain and participates in cholesterol transport and in proteolysis of Amyloid beta in the brian
- Polymorphic *APOE*, *APOE4*, is not as effective in these processes
- Females heterozygous for *APOE4* have a 4-fold higher risk of developing AD than heterozygous men. Not known why.



Liu CC et al, *Nat Neuro* 2013



Pinares-Garcia et al, *Brain Sci*., 2018

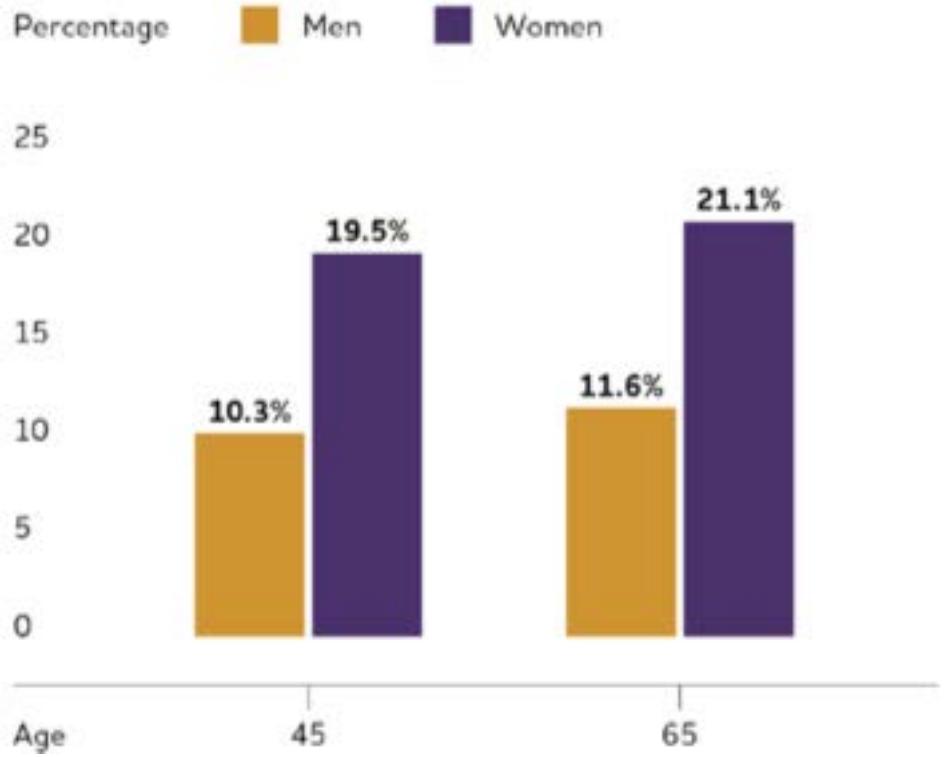
Epidemiological data

AD Facts & Figures:

1) In USA: 3.2 M women and 1.7 M men have AD (2014)
   - Being female is a risk factor for AD
   - Females have a higher incidence of developing AD. However, no excess risk in USA – regional differences!
2) Women live on average longer than men
3) Women born before ~1950 have lower education than men
4) Men who survive middle age cardiovascular events are healthier and less prone to develop AD ("survival bias")
   - This is controversial!

5) Women with *APOE4* genotype have a higher risk of developing AD compared with men with the same genotype
   - **Estrogen** has been suggested to be involved!

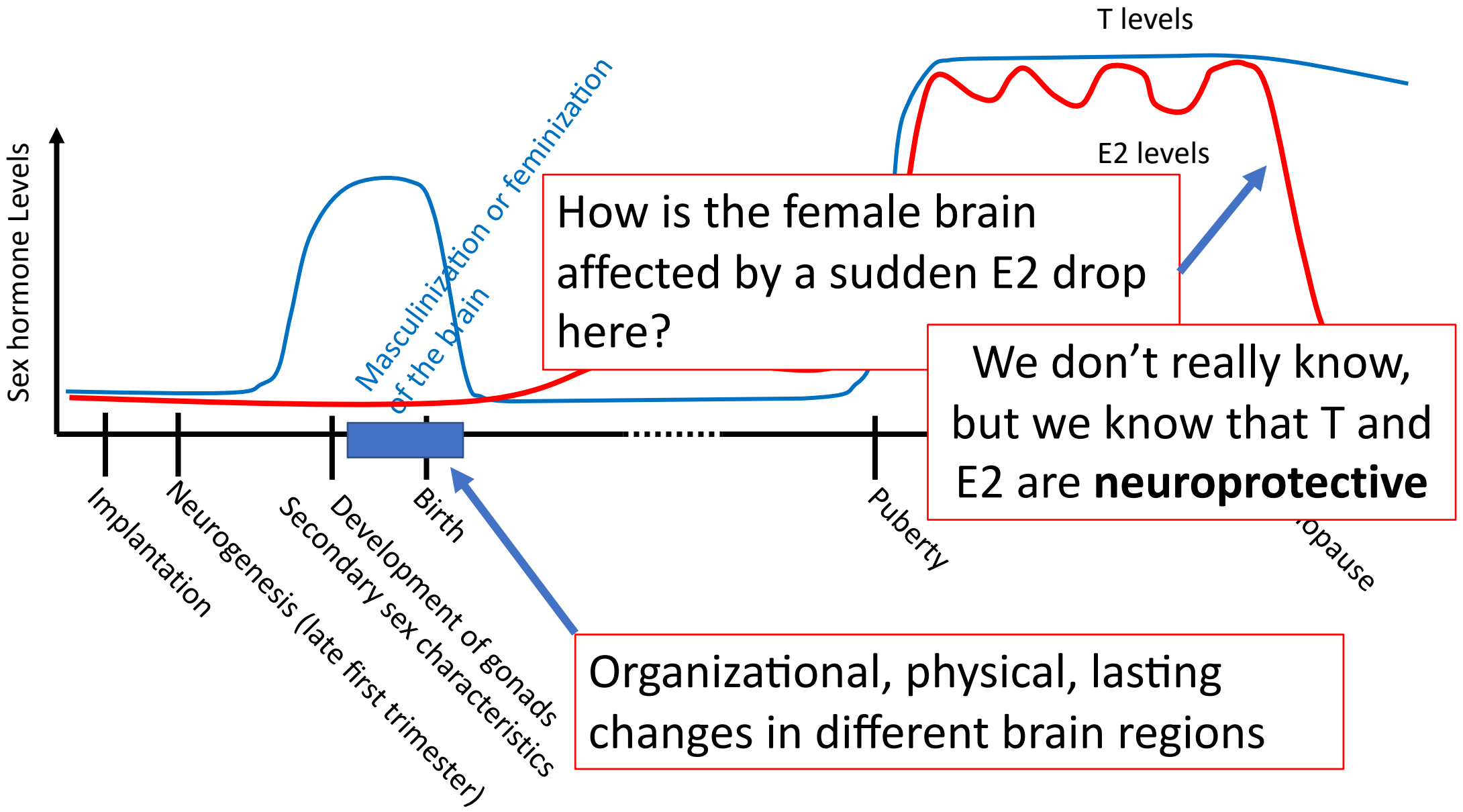
Farmingham Heart Study (USA), 1975-2009, 7901 participants:



Lifetime risk of developing AD at age 45 and 65

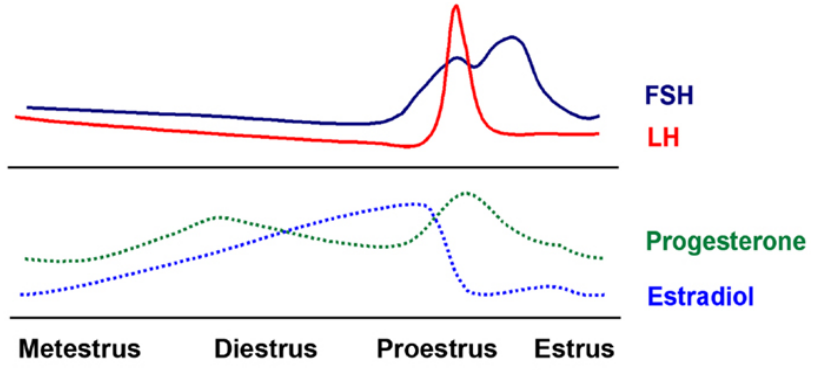
Alzheimer's disease Facts & Figures. *Alzheimer & Dementia,* 2018

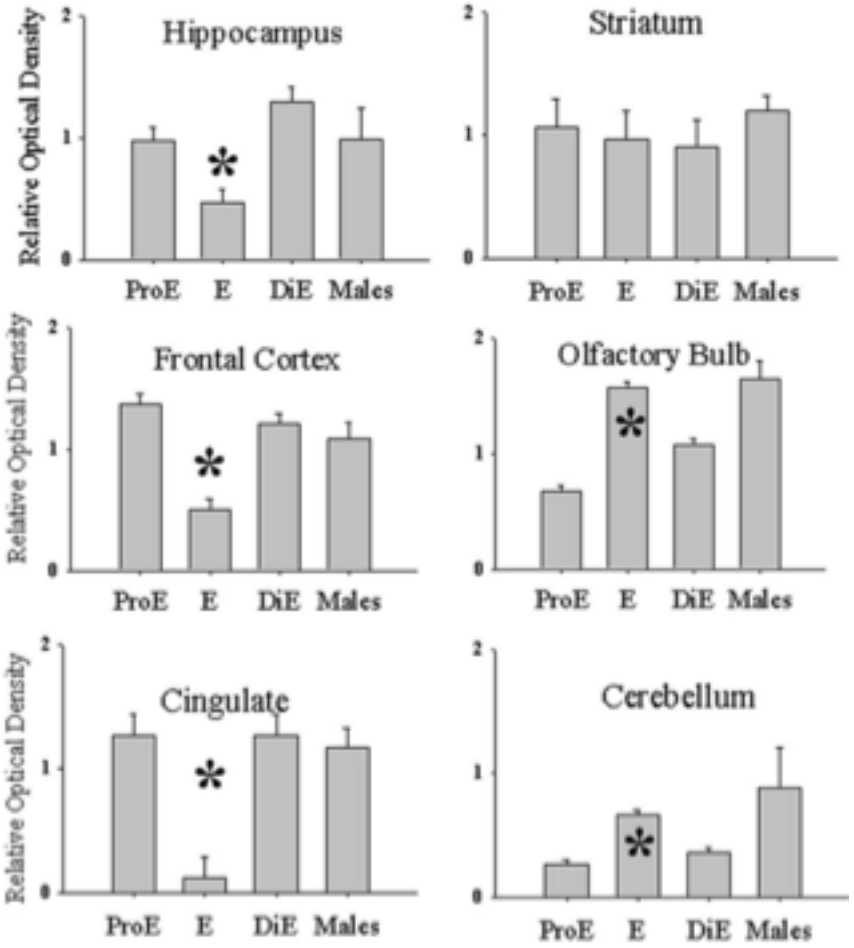
Wait. How could estrogen be involved in AD?



1. E2 regulates ApoE

- ApoE participates in amyloid beta degradation and brain cholesterol transport
- ApoE levels in mice change according to estrus cycle
- Different effects in different brain regions
- Not known how – likely indirect effects of E2 on ApoE

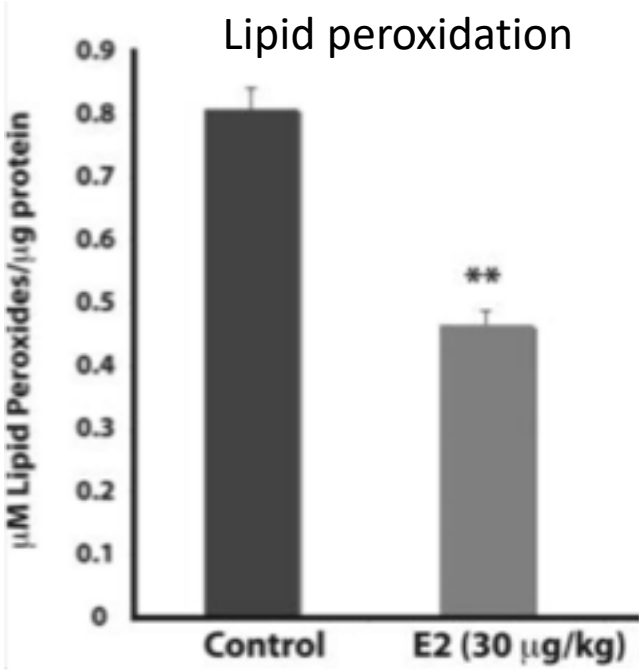




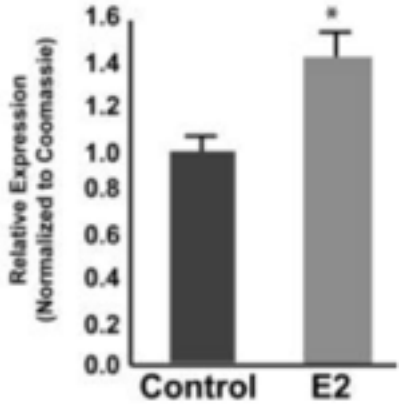
Struble RG, et al, *Exp Neurology*, 2003

2. E2 is antioxidative

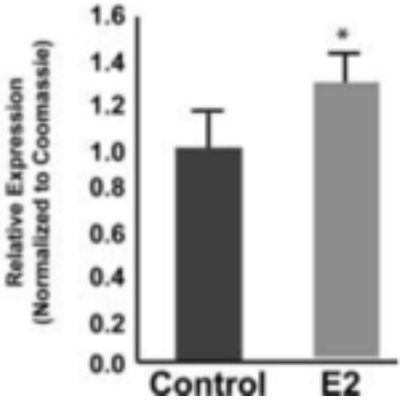
Lipid peroxidation



MnSOD



Peroxiredoxin V



- E2 and progesterone (P4) decreases mitochondrial electron leakage and thereby **lowers brain mitochondrial lipid peroxidation**
- E2 upregulates expression of key antioxidant enzymes in the brain

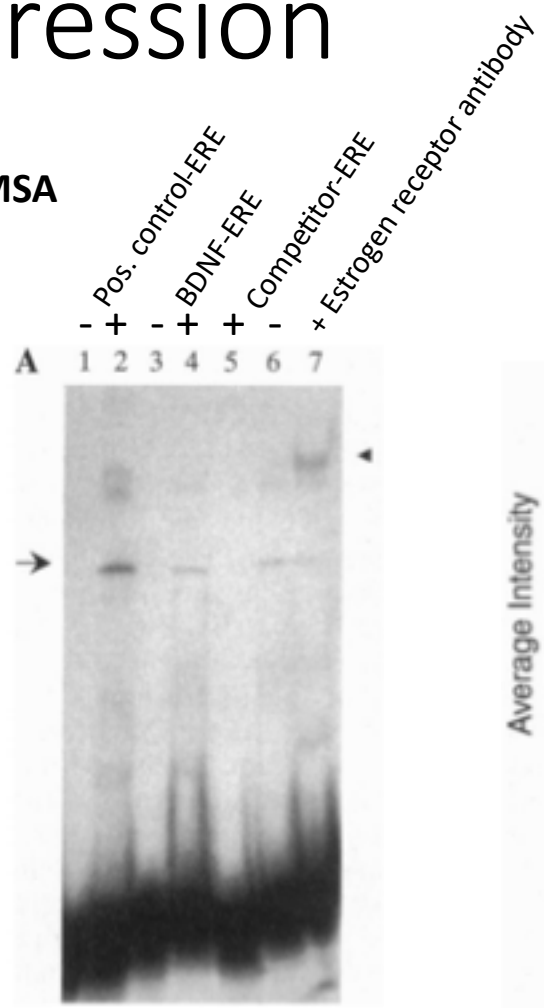
Irwin et al, *Endocrinology*, 2008

3. E2 modulates BDNF expression

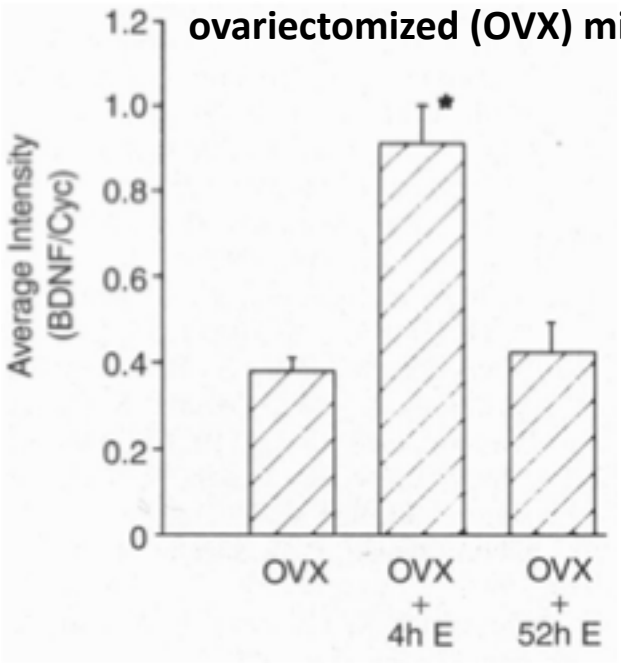
BDNF (Brain-derived neurotrophic factor) promotes:

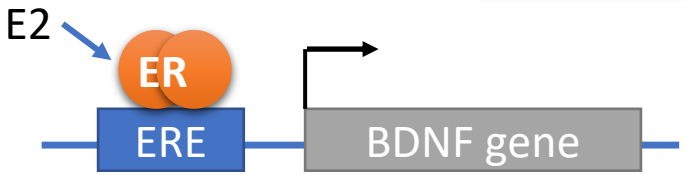
- Neuronal survival
- Neuronal growth and differentiation
- Neurogenesis (from neural stem cells)
- Synapse formation
- Long-term memory

EMSA



BDNF RNA expression in cerebral cortex of ovariectomized (OVX) mice

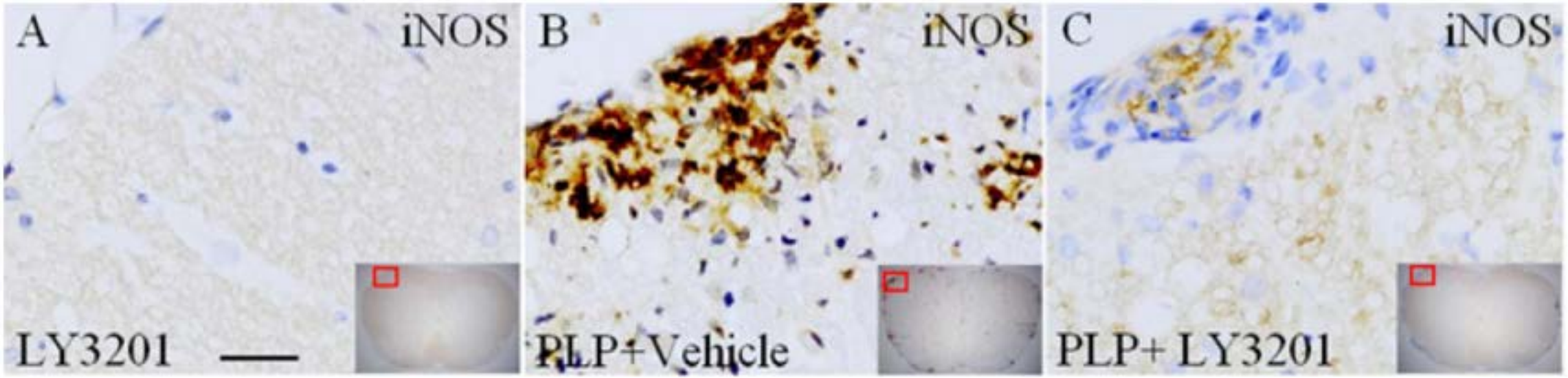




ERE = Estrogen response element in promoter region

Sohrabji F, et al, *PNAS*, 1995

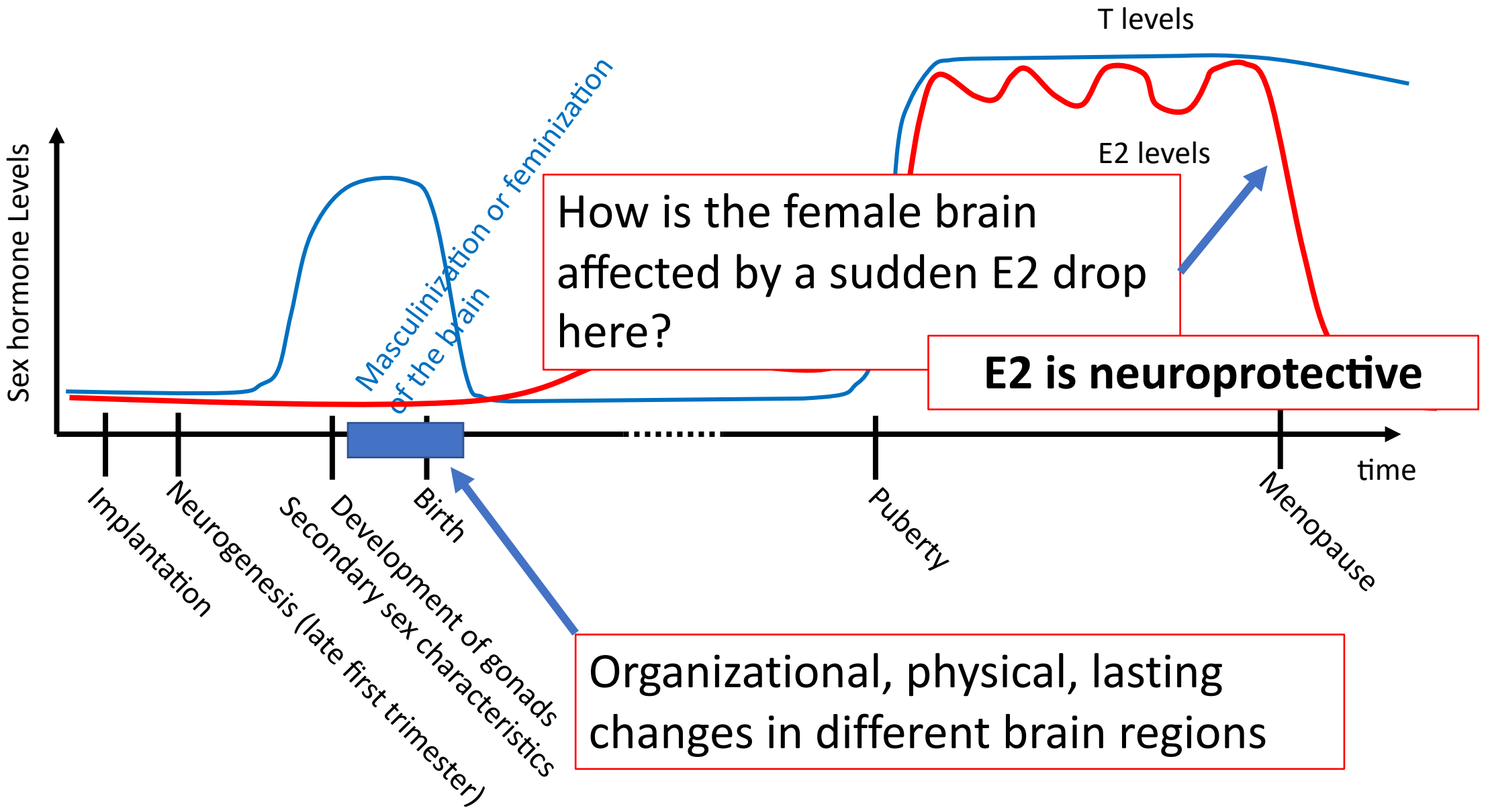
4. E2 is anti-inflammatory



Selective ERβ agonist (LY3201) decreased reactive pro-inflammatory microglia (iNOS expressing) in a mouse model of Multiple Sclerosis (PLP treatment)

Wu WF et al, *PNAS*, 2013

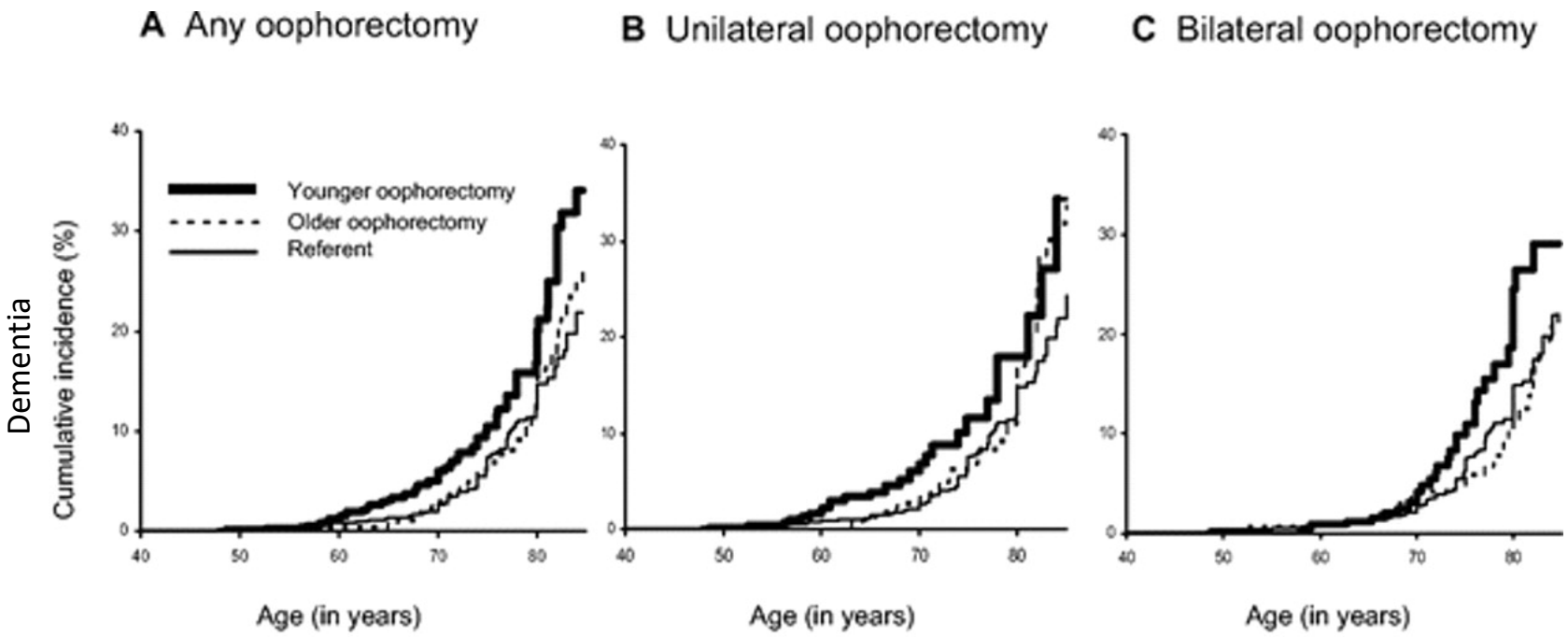
Is menopause a risk factor for AD??



Is menopause a risk factor for AD??

Back to epidemiology!

Looking at the extreme cases: surgical menopause (oophorectomy) & dementia risk



Oophorectomy is a risk factor for dementia (HR: 1.46), especially if performed at a younger age (HR: up to 4.61)

Rocca WA et al, *Neurology*, 2007

Could hormone replacement therapy (HRT, ERT) at menopause lower the risk of AD and dementia later in life??

In such case, when and how should HRT be given??

Several variables to take into consideration:

- HRT type (e.g. 17β-estradiol or conjugated equine estrogens, ±progestins)
- Route of administration (oral, vaginal, or dermal)
- Timing
- Duration
- Interaction with other sex-biased risk factors? – e.g. cardiometabolic disease
- Interaction with other gender-biased risk factors? – e.g. education, smoking

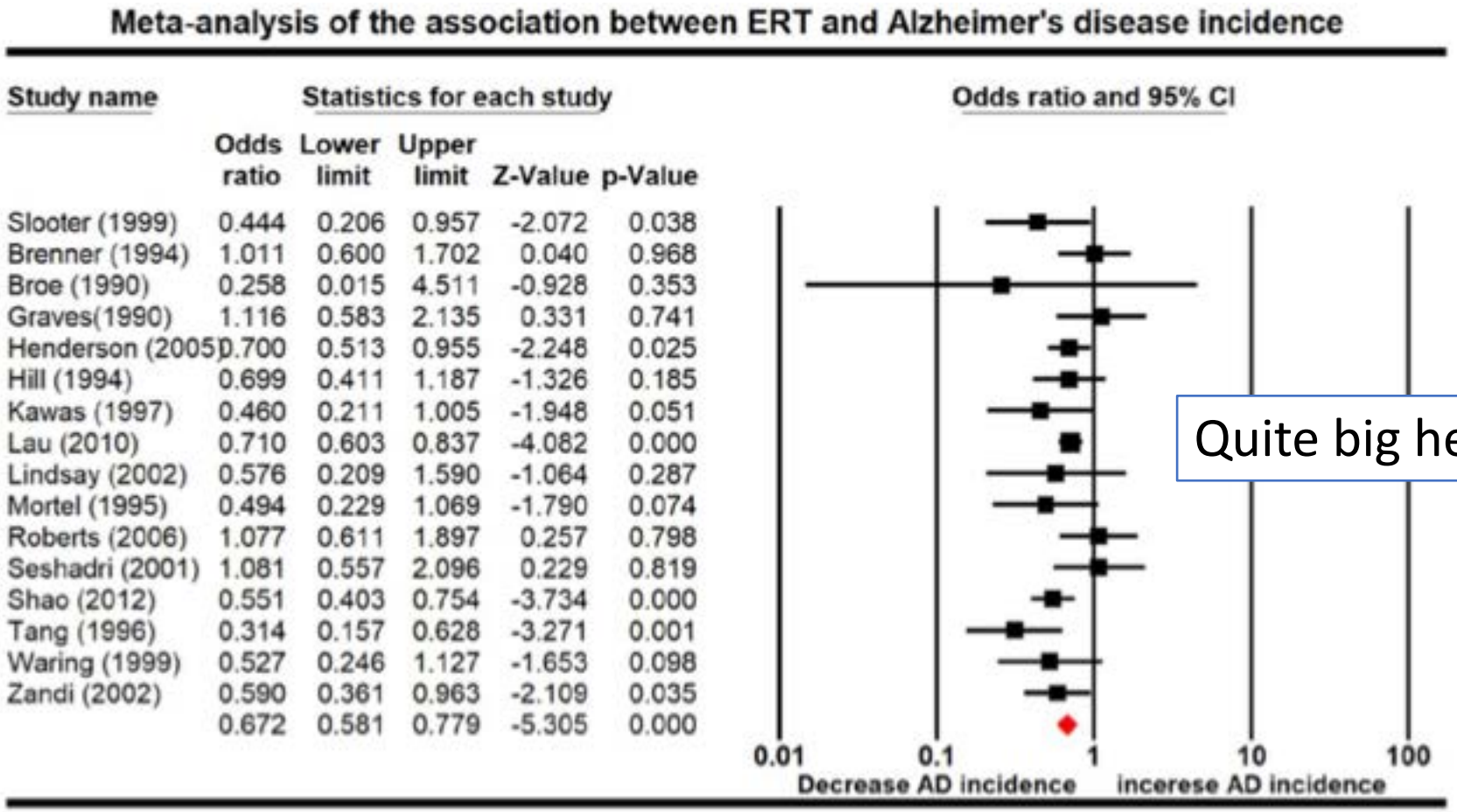
What about possible side effects of HRT?

- Increased risk for endometrial and breast cancer
- Increased risk for thromboembolism

Overall benefits must be weighed against overall risks!

Some studies exist on hormone replacement therapy (HRT, ERT) and AD risk later in life

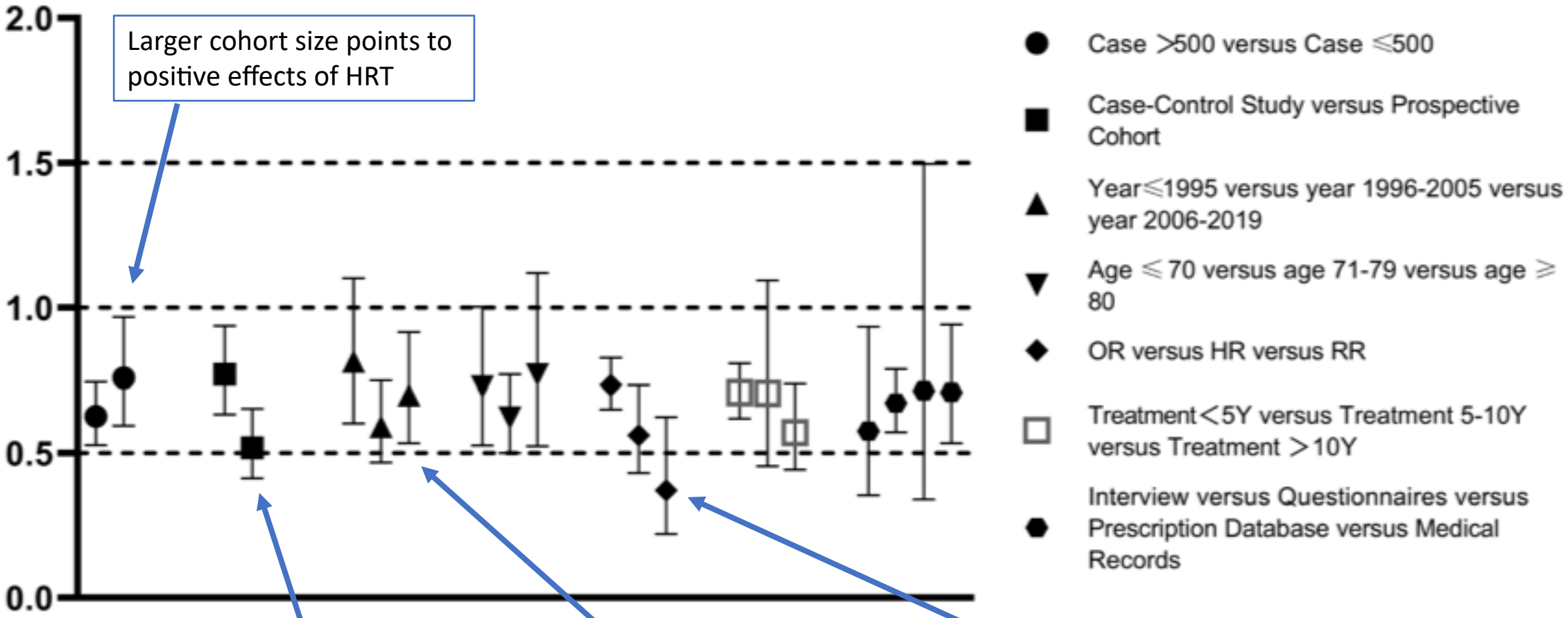
About 20 epidemiological studies have been performed



Quite big heterogeneity…

Song Y-J, et al, *Fornt Neuorsci*, 2020

Why the heterogenous results??



Larger cohort size points to positive effects of HRT

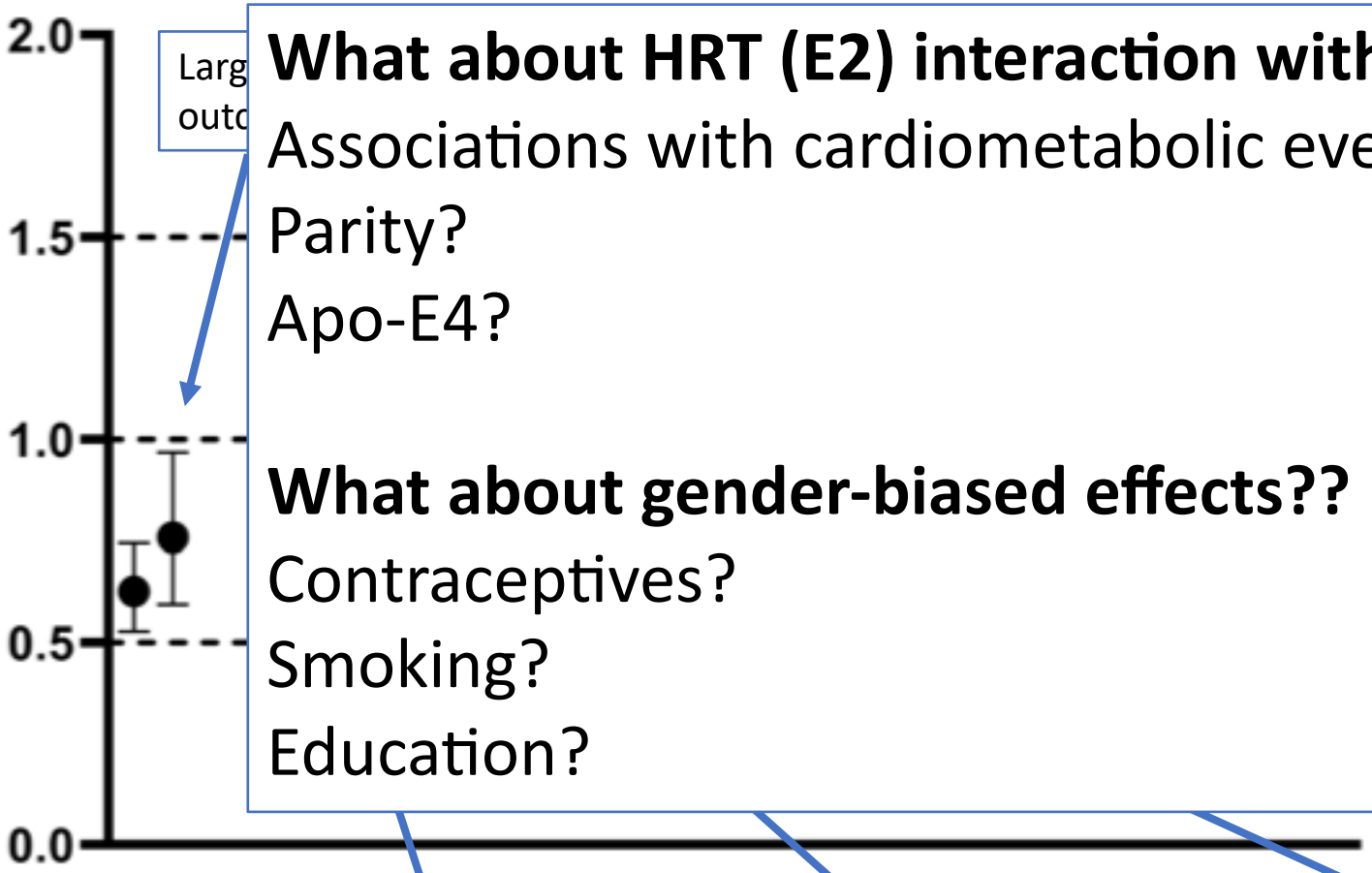
Prospective longitudinal studies show stronger beneficial effects of HRT than retrospective case-control studies

Somewhat stronger beneficial effects observed in newer studies

Studies displaying results as relative risks (eg prospective studies) show stronger beneficial effects

Song Y-J, et al, *Fornt Neuorsci*, 2020

Why the heterogenous results??



Larg
outc

What about HRT (E2) interaction with other sex-biased effects??

Associations with cardiometabolic events?

Parity?

Apo-E4?

What about gender-biased effects??

Contraceptives?

Smoking?

Education?

We don't know!

How does E2 interact with AD risk on a mechanistic level??

We don't really know that either!

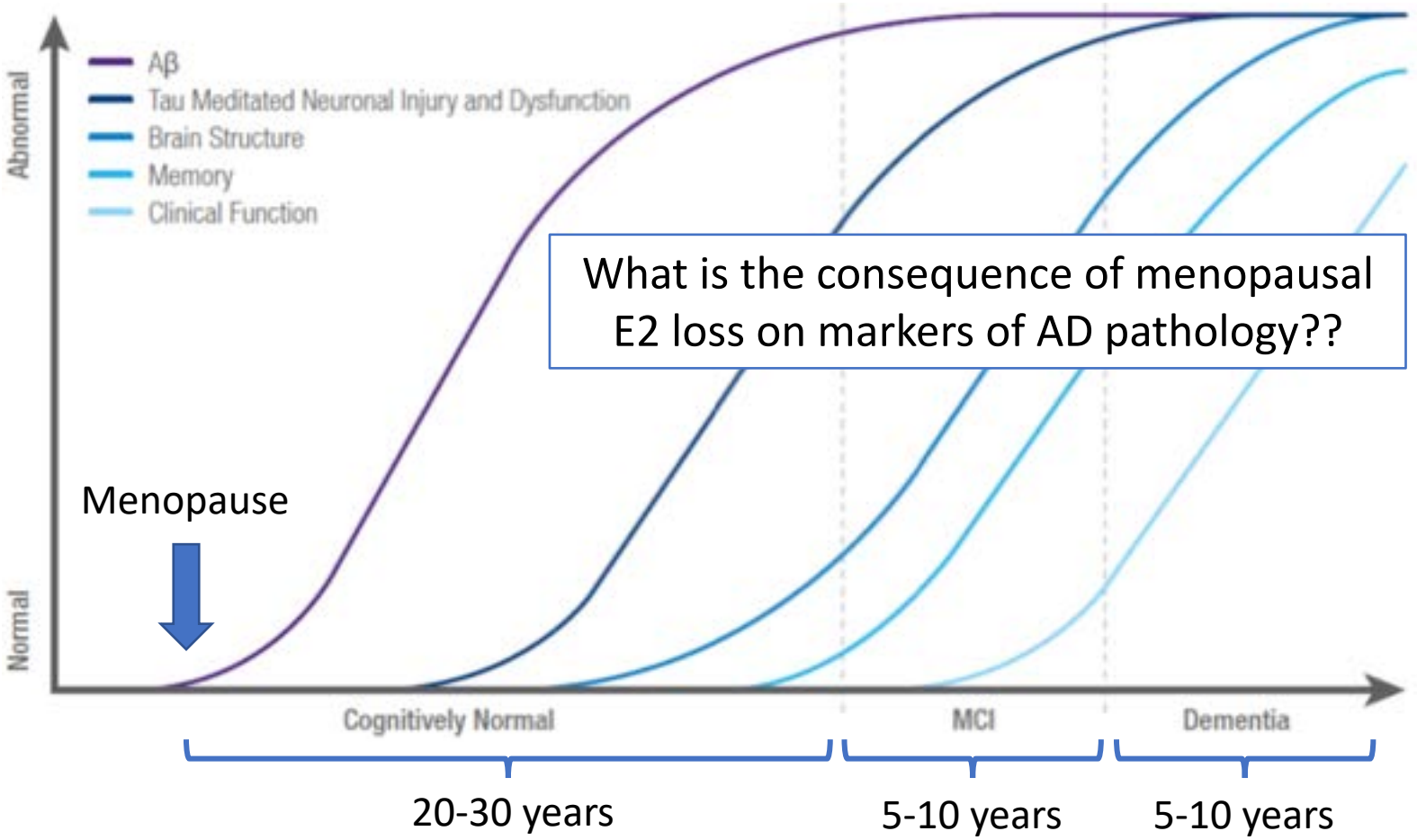
Prospective longitudinal studies show stronger beneficial effects of HRT than retrospective case-control studies

Studies di
(eg prosp
beneficial

Somewhat stronger beneficial effec
observed in newer studies

Song Y-J, et al, *Fornt Neuorsci*, 2020

But wait! Menopause occurs ca 20-30 years before any signs of cognitive impairment!



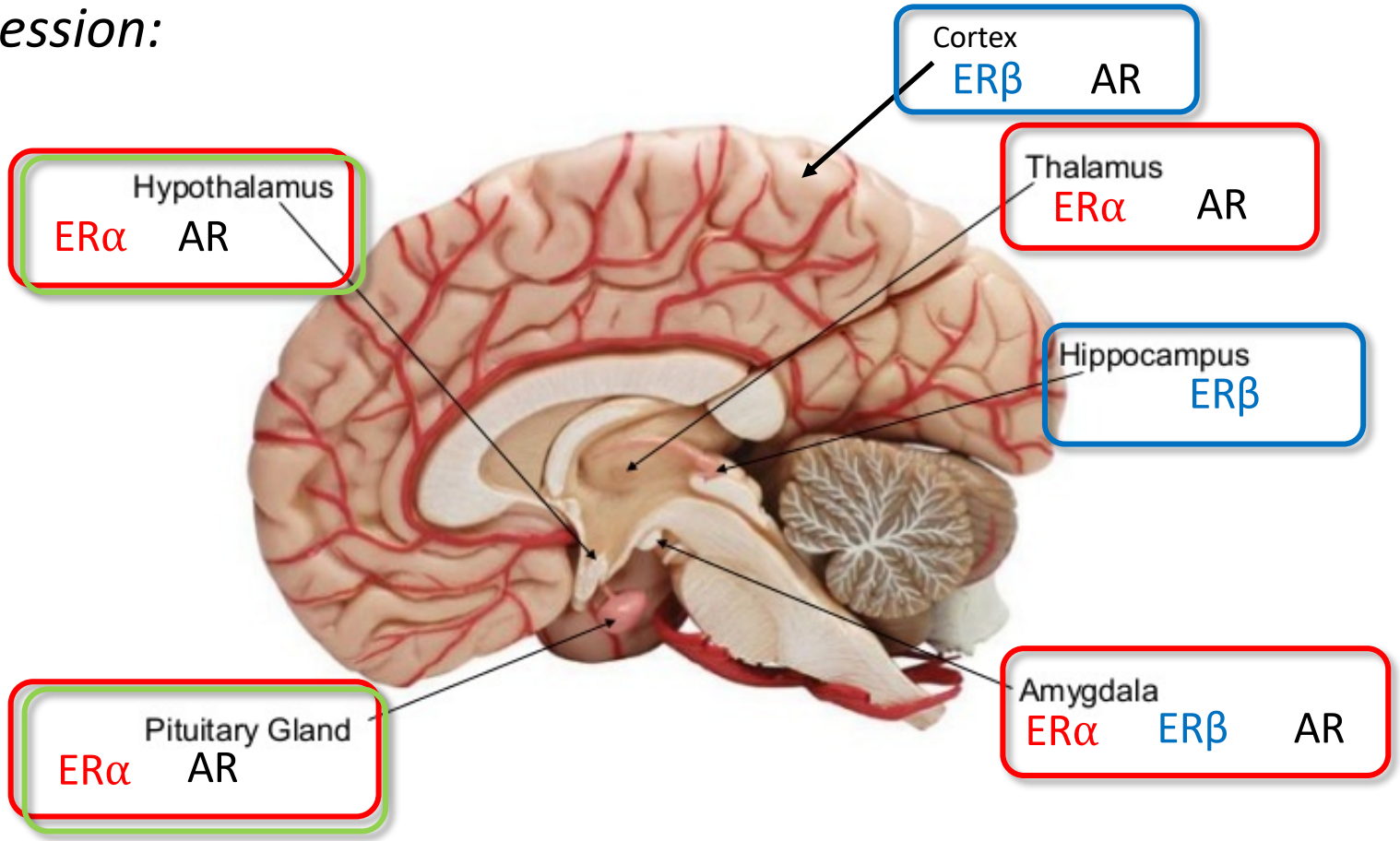
Jack et al., 2010

Estrogen and Androgen receptors in the **adult brain**

Estrogen Receptor alpha (ERα)

Estrogen Receptor beta (ERβ)

Expression:

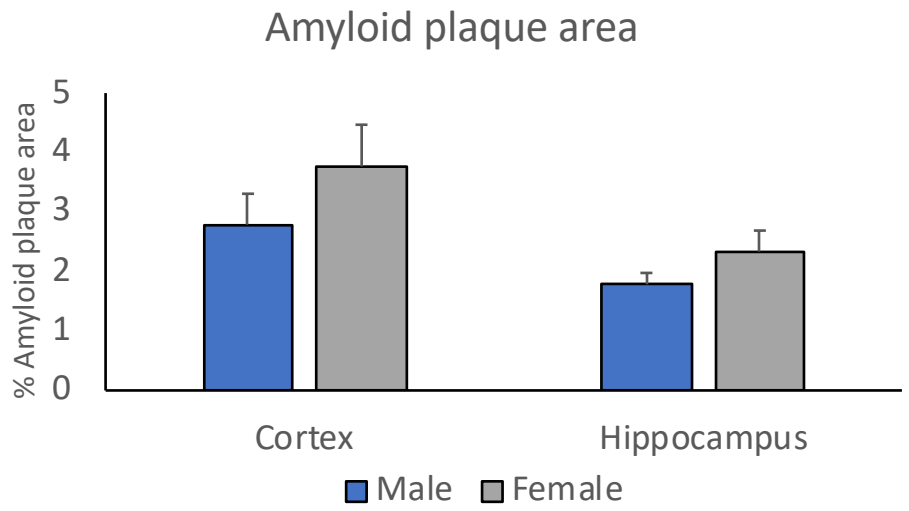


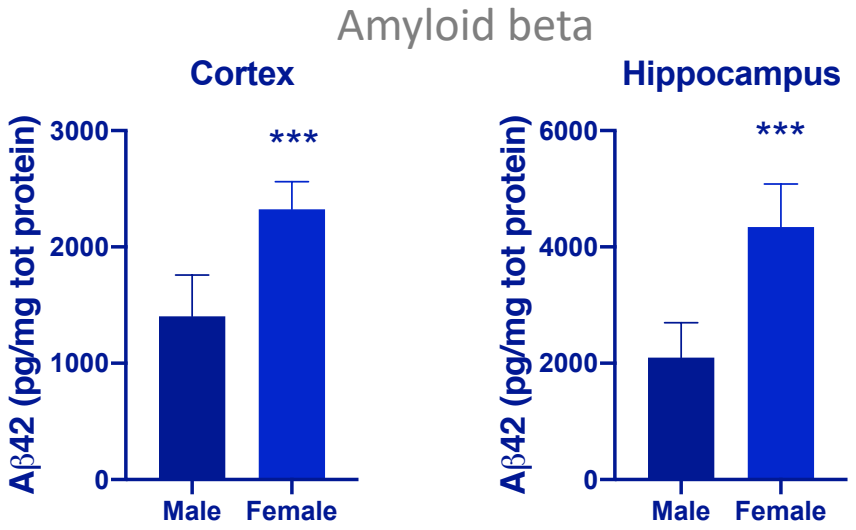
Limbic system (mood, emotions)

Cognition (memory, learning)

Reproduction (HPG-axis)

Males vs Females





- Female AD mice have a more aggressive AD pathology than male mice
- Ovariectomy may worsen pathology in frontal cortex
- Differences in neuroinflammatory response may be one contributing factor for these sex differences
  (lessons from ischemic stroke)

Still much is not known

- Other factors? Cholesterol metabolism?
- Species differences! Sex hormonal signaling in mice is different from humans

The complexity of human AD cannot be fully addresses only using experimental models

What about E2 interaction with other sex-biased effects??
Associations with cardiometabolic events?
Parity?
Apo-E4?

What about gender-biased effects??
Contraceptives?
Smoking?
Education?

COMPREHEND

Karin Leander, IMM, KI
Coordinator

”Combined cohorts of menopausal women – Studies of register-based health outcomes in relation to hormonal drugs”

- Pools over **80 000** Swedish postmenopausal women from different cohorts (**unique power!**)
- Detailed and unique information of HRT use (baseline 1987-2002)
- Type, route of administration, timing, duration
- Information on possible confounding variables exist, such as:
  Age at baseline, level of education, smoking status, body mass index, level of physical activity, age at menopause onset, type of menopause, parity (number of given births), contraceptives (oral, vaginal or transdermal), alcohol consumption, hypertension, diabetes, family history of cardiovascular disease, and dyslipidemia
- Genotype of most participants exist (Apo-E, other genes)

Participants are followed over time (prospective) through linkage to Swedish national registers regarding incidence of chronic disease: AD and non-AD dementia, Cardiometabolic disease (CVD, CHD)

Are AD treatment strategies different for men and women today?

There is no difference in treatments given to men and women at similar stage of disease

There is no difference in preventive recommendations given to men and women

There is no difference in how dementia is diagnosed between men and women

(women can perform better on verbal memory test despite presence of pathology)

Why?

Most animal experiments performed on **male** mice

Species differences: Rodent sex hormone signaling is different from human

AD etiology has a **complex** interaction with biological and environmental factors which affects one sex more than the other

More integrative research is needed!

ER signaling group at BioNut, KI

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Per Antonson
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Peik Brundin
Chiara Gabbi

Soon hiring PhD student and postdoc!

National Institutes of Health
VETENSKAPSRÅDET THE SWEDISH RESEARCH COUNCIL
STINT Stiftelsen för internationalisering av högre utbildning och forskning
The Swedish Foundation for International Cooperation in Research and Higher Education
Karolinska Institutet