Aspetti di Genere nella Sclerosi Multipla

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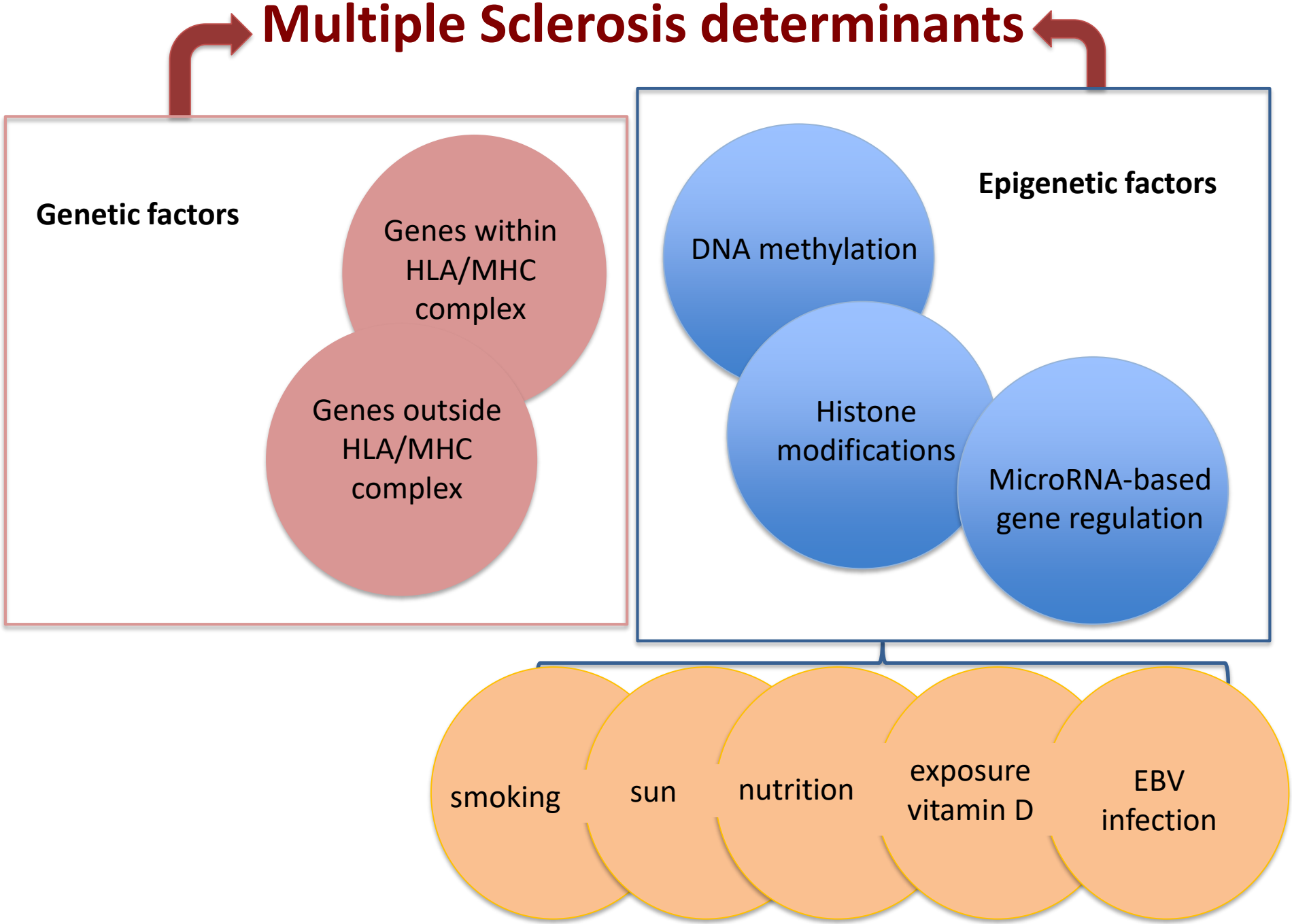
Ospedale San Camillo Forlanini, Roma

Multiple Sclerosis determinants



Outline

- Sex differences in susceptibility to MS
- Sex differences as major MS modifier on disease activity and progression.
- Sex differences in treatment response
- Impact of exogenous hormones on MS immune response and disease activity

Sex and MS susceptibility

Women *are more susceptible to MS than men*

	women to men ratio
Global	**2:1**
Early onset (<17 years)	**3:1**

Sex and MS susceptibility

Potential factors involved in the sex bias in MS

1. **Sex-related differences in immune responsiveness** and in responses to infection

   - **Women** have a **stronger TH1 immune response** mediated by interferon gamma (Zhang MA et al 2012).

   - Animal studies have shown that **female mice** have **greater antibody production capacity**, **increased cell-mediated responses**, and **increased production of interferon gamma**, **IL-1**, and **IL-6** compared to males (Edinger D et al 1972)

   - **Immune responses to myelin Ag are greater among women** with MS than among men with MS (Moldovan IR et al. 2008).

Sex and MS susceptibility

Potential factors involved in the sex bias in MS

2. Different responses to environmental factors (sun exposure and vitamin D supplements)

- only **female EAE mice** had a milder disease course upon feeding with a vitamin D enriched diet (Spach KM, 2005)
- supplementary **vitamin D** has been shown to have a greater immunomodulatory effect in women with MS than in men (Correale J, 2010)

Sex and MS susceptibility

*A **female** specific increase in the incidence of MS over time has been documented*

Sex ratio of multiple sclerosis in Canada: a longitudinal study

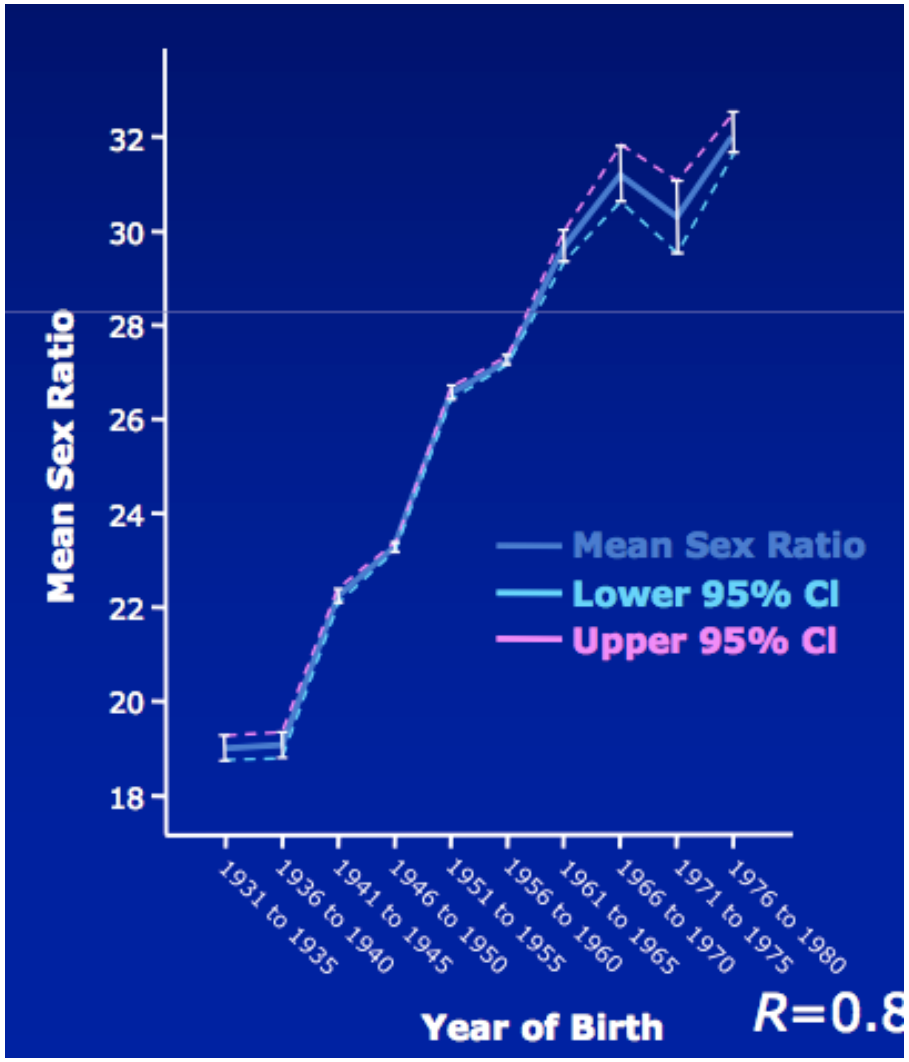
Sarah-Michelle Orton, Blanca M Herrera, Irene M Yee, William Valdar, Sreeram V Ramagopalan, A Dessa Sadovnick, George C Ebers, for the Canadian Collaborative Study Group*

Lancet Neurol 2006; 5: 932–36

Methods Since environmental risk factors seem to act early in life, we calculated sex ratios by birth year in 27 074 Canadian patients with multiple sclerosis identified as part of a longitudinal population-based dataset.

Findings The female to male sex ratio by year of birth has been increasing for at least 50 years and now exceeds 3·2:1 in Canada. Year of birth was a significant predictor for sex ratio ($p<0·0001$, $\chi^2=124·4$; rank correlation $r=0·84$).

Sex and MS susceptibility

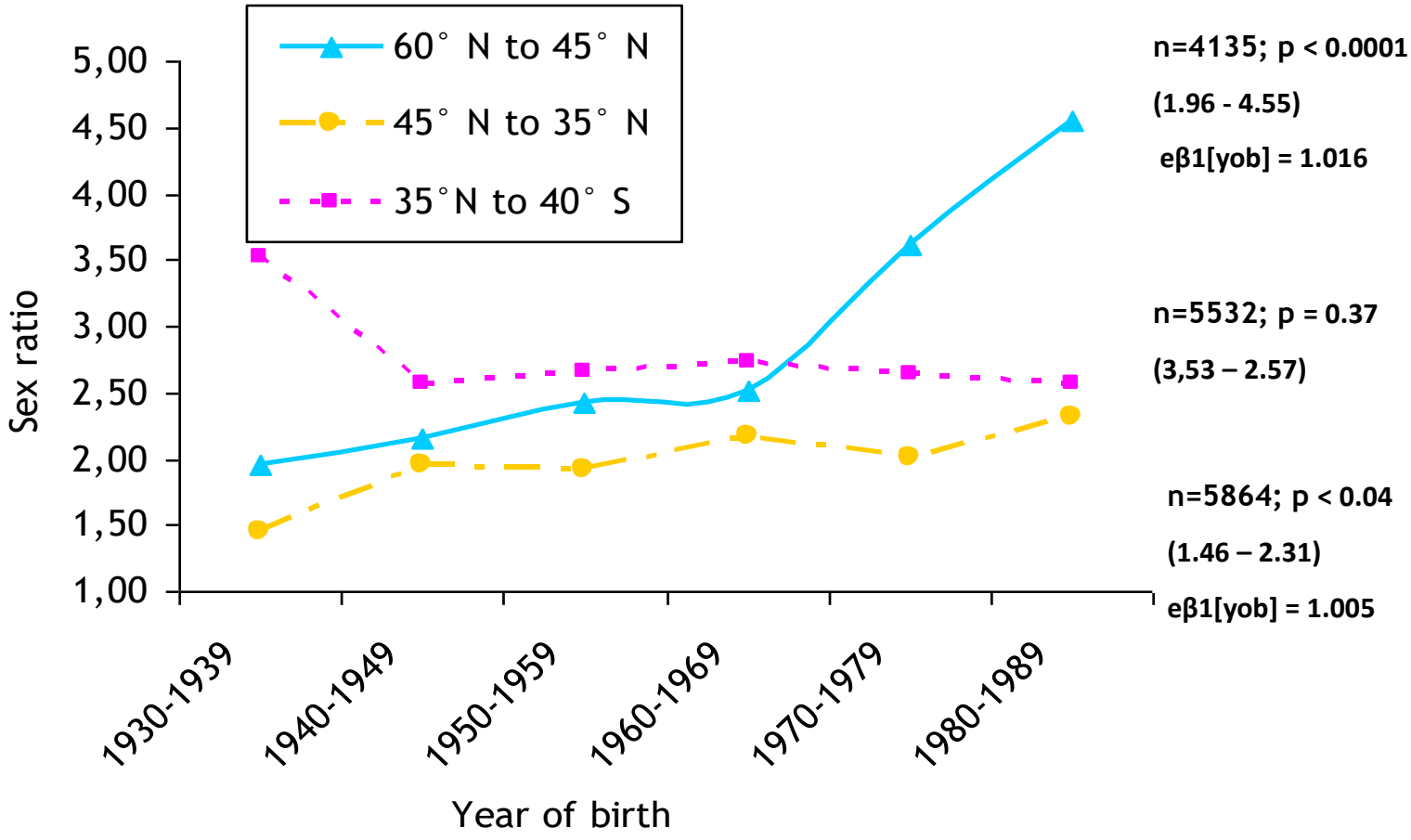


- **Female to male ratio** increases with time (from 1.9 to 3.2)
- No possible counfonders identified
- Effect indipendent of diagnostic changes (*its progressive nature and the use of year of birth as the predicting variable make it unlikely that the findings could be explained by any artifact related to ascertainment)*.

[Orton et al. 2006]

Sex and MS susceptibility

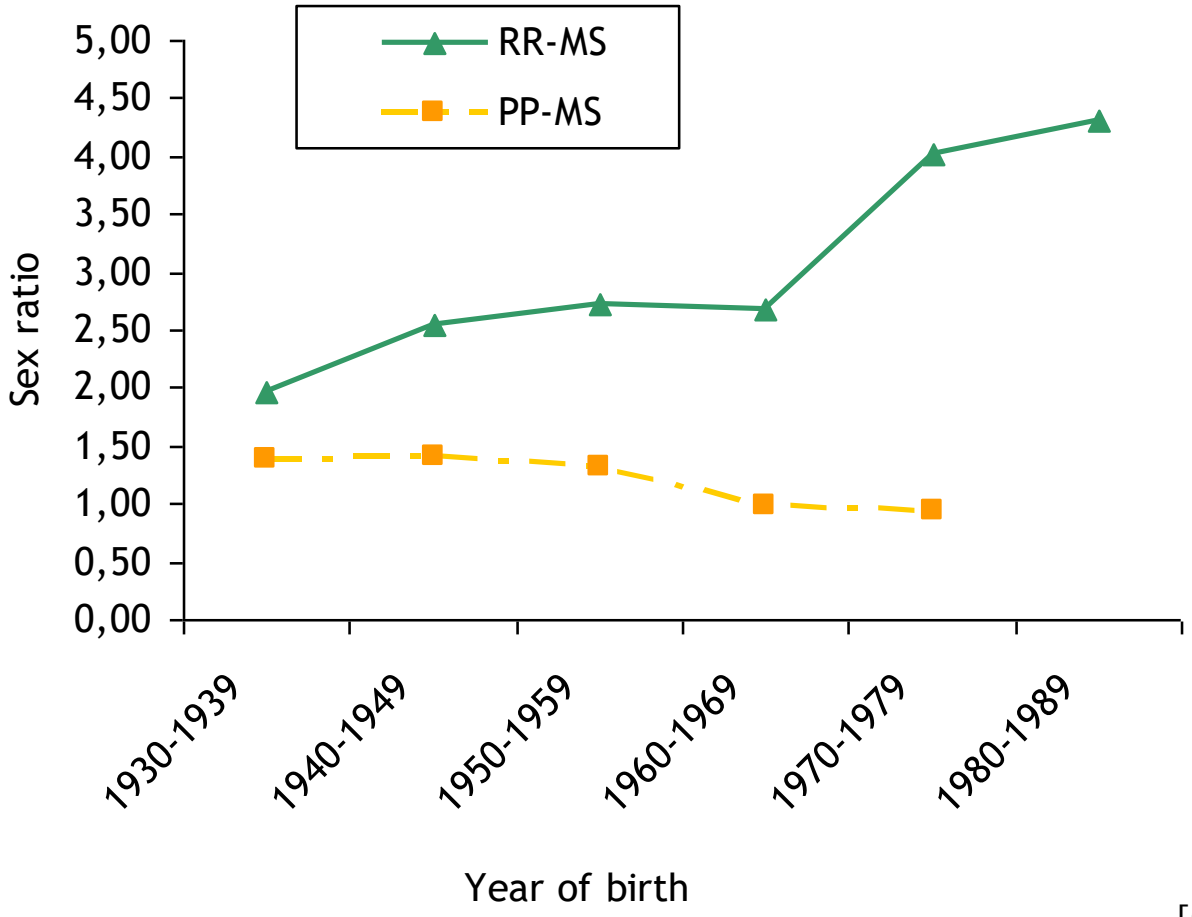
Plot of ***sex ratio*** by year of birth in MS patients stratified by latitude



[Graziano et al PLoS One 2012]

Sex and MS susceptibility

The increase of the **F/M ratio** arises mainly from an increase in the number of female patients with **RRMS**



n=3445; p < 0.0001

(1.93 - 4.39)

eβ1[yob] = 1.014

n=435; p = 0.28

(1.38 – 0.93)

[Graziano et al. PLoS One 2012]

Sex and MS susceptibility

ARTICLE

Incidence of MS has increased markedly over six decades in Denmark particularly with late onset and in women

Nils Koch-Henriksen, DMSc, Lau Caspar Thygesen, PhD, Egon Stenager, MD, Bjarne Laursen, PhD, and Melinda Magyari, PhD

Neurology® 2018;0:e1-e10. doi:10.1212/WNL.0000000000005612

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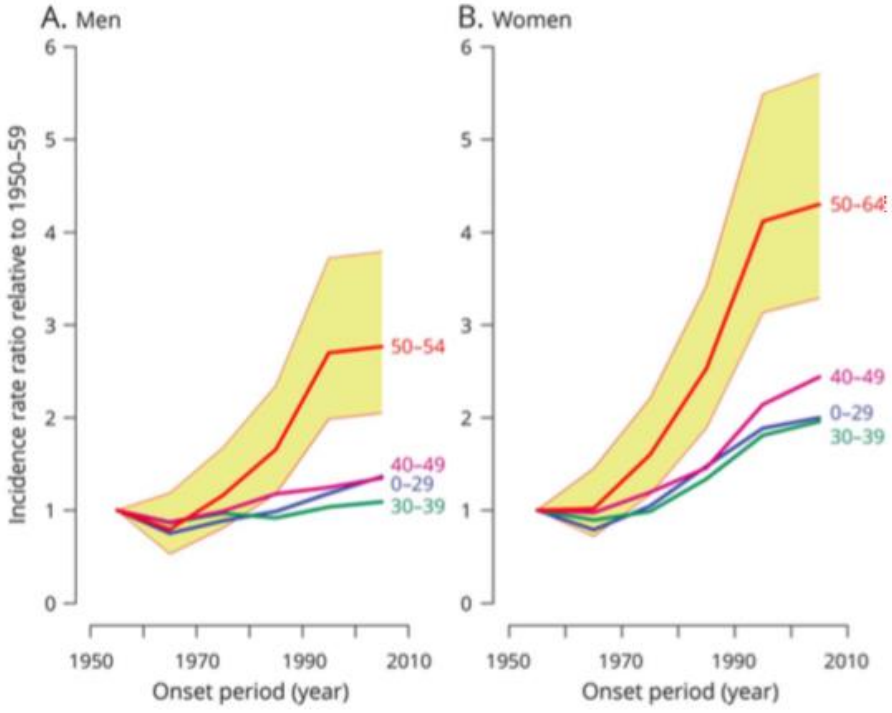


Danish Multiple Sclerosis Registry

19,536 cases with clinical onset from 1950 to 2009

- The incidence of MS in women has more than **doubled** over 60 years, and in the same period, it has only moderately increased in men (24%).
- **The increase in women was most prominent with onset of MS at age 50 years or older**

Sex and MS susceptibility

*Potential factors involved in the increased susceptibility in **women***

- **genetic (??)**
- **gene-environment interaction** (including sex differences in expression and environmental effects on candidate genes on the X or Y chromosome)
- **lifestyle changes** (contraception, diet, obesity, smoking, sunlight exposure, vitamin D deficiency, higher age at first childbirth, and fewer life-time pregnancies).

Sex and MS susceptibility

*Potential life style changing involved in the increased susceptibility in **women***

Table 1. MS risk factors may be differentially regulated in males and females.

Behavioral risk factors Westernizing gender norms	1. In some areas where the F:M ratio in MS is increasing, girls have experienced rapid increases in the time spent indoors, as a result of rapid urbanization, education and participation in the workforce. 2. There has been a dramatic shift in women's reproductive choices and trajectories, in the past century.
Smoking	1. Smoking may increase the risk of MS in women only. 2. Potential mechanisms may include an interaction between sex/gender and smoking, yielding increased levels of mature peripheral functioning T cells (OKT3+) in F smokers. 3. The increasing F:M ratio in MS parallels that in smoking; but in smoking, a higher F:M ratio may be driven both by a decrease in M rates, as well as by an increase in F's smoking rates.[6]
Sunlight Dietary risk factors Vitamin D	Sunlight deprivation has worse consequences in F than in M.[7] A functional synergy between 1,25(OH) D(3) and 17-β estradiol is observed, mediated through estrogen receptor α, mainly in the F and secondarily in M: As a consequence, Vitamin D may play a more important immunomodulatory role in F with MS than in M.[6]

[Bove R et al Mult Scler 2014]

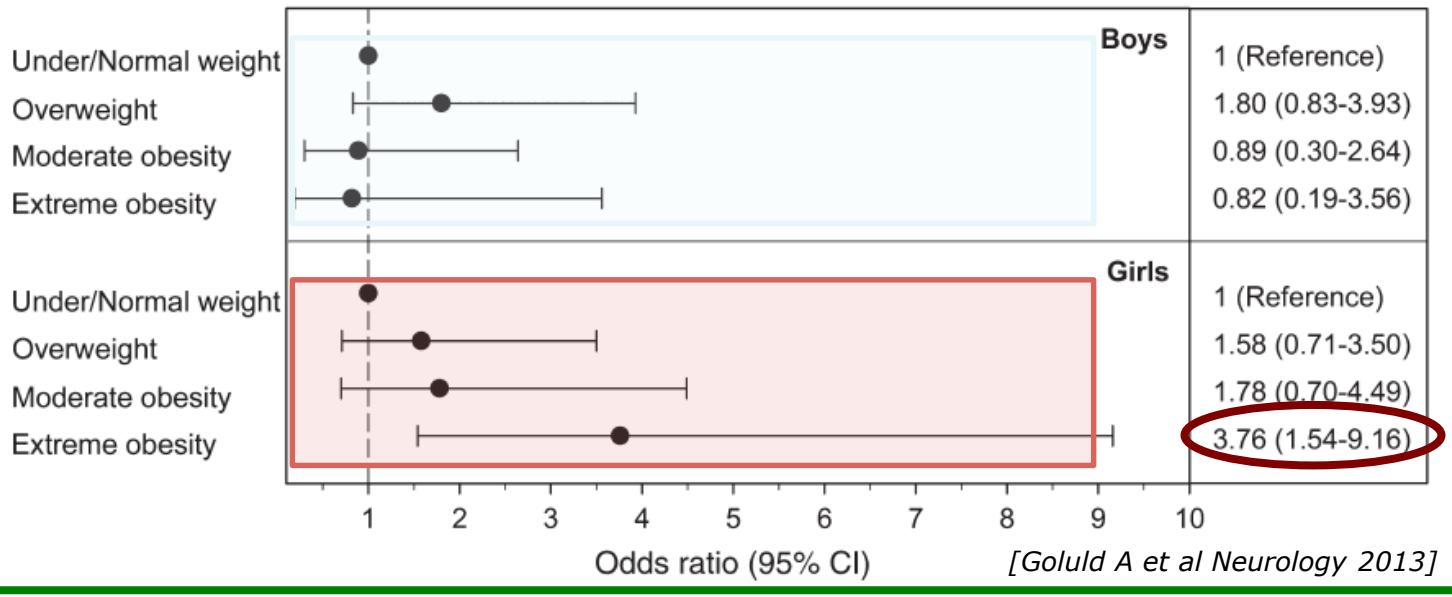
Sex and MS susceptibility

*Potential life style changing involved in the increased susceptibility in **women***

Diet and metabolism

1. Overweight/obesity at 18–20 years of age may double MS risk.[8]
2. Only in adult F obesity at MS onset may be associated with a 2-fold increased OR of a relapsing course at onset.[9]

Figure Association between weight class and pediatric multiple sclerosis/clinically isolated syndrome by sex

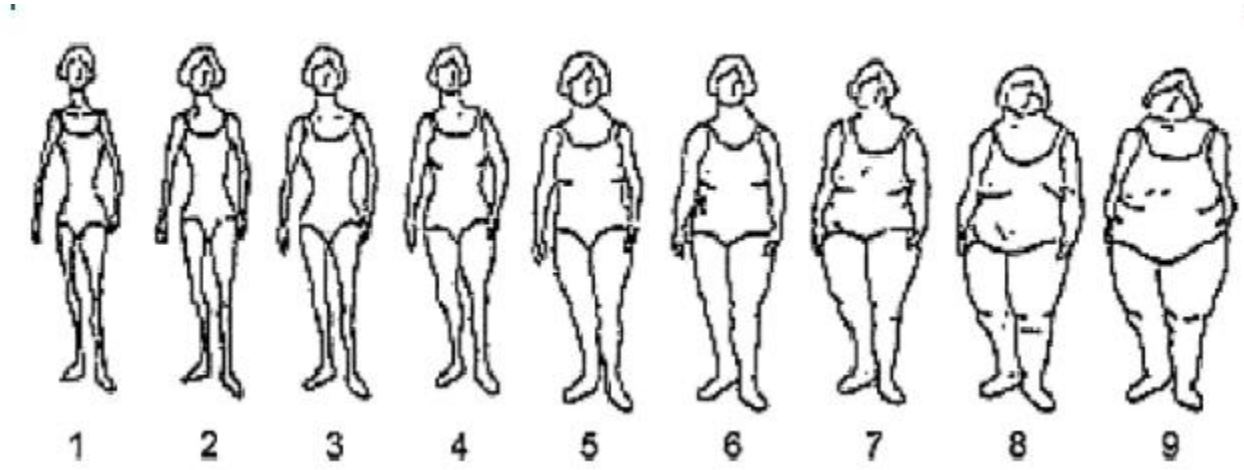


[Goluld A et al Neurology 2013]

[Bove R et al Mult Scler 2014]

Sex and MS susceptibility

*Potential factors involved in the increased susceptibility in **women***



- Adipose tissue produces and releases a variety of **proinflammatory cytokines**, **including leptin**, which promotes Th1 responses and reduces regulatory T-cell activity
- It is well-known that **obesity is potentially associated with increased estrogen levels in both sexes and lower androgen levels in males**

[Munger KL et al Neurol 2009]

Outline

- Sex differences in susceptibility to MS
- **Sex differences as major MS modifier on disease activity and progression.**
- Sex differences in treatment response
- Impact of exogenous hormones on MS immune response and disease activity

Gender as MS modifier

The course of the disease is different between sexes

- **Women** have an increased risk of developing clinically-definite MS after a first demyelinating event
  - [Optic Neuritis Study Group et al , 2008]

- **Women** have more often a **benign form** (F/M ratio 4:1) [Hawkins S.A., 1999, Bove R and Chitnis, 2013]

- **Women** have more often a **RR** course and higher relapse rate. The **progression** is little slower in women
  - [Duquette P.,1998, Confavreux C., 2003, Kalincik T. 2013]

- **Males** are more likely to display a **progressive disease onset**, **poor recovery after initial attacks**, more rapid rapid clinical progression and an overall more malignant course.

Gender as MS modifier

The course of the disease is different between sexes

- Males with MS may also be at higher risk of developing **cognitive impairment** [Benedict et al 2011]

Table 3 General characteristics of the study population by sex and by degree of cognitive impairment

Variables	Men				Women			
	Preserved (n = 70)	Mild (n = 97)	Severe (n = 16)	p Value	Preserved (n = 151)	Mild (n = 143)	Severe (n = 26)	p Value
Age, years, mean (SD)	37.1 (9.6)	39.4 (9.3)	44.8 (9.4)	0.02[a]	36.6 (9.6)	36.1 (8.9)	36.2 (7.9)	0.98[a]
Age at onset, years, mean (SD)	27.2 (9.5)	27.5 (9.5)	29.7 (10.3)	0.70[a]	27.2 (8.3)	26.9 (8.6)	25.2 (8.5)	0.62[a]
Disease duration, years, mean (SD)	9.9 (7.5)	12.0 (8.5)	15.1 (7.2)	0.02[a]	9.3 (7.8)	9.1 (7.2)	11.0 (7.5)	0.32[a]
Disease course, no. (%)								
RR	54 (77.1)	70 (72.2)	13 (81.2)	0.23[b]	127 (84.1)	117 (81.8)	19 (73.1)	0.42[b]
SP	11 (15.7)	25 (25.8)	3 (18.8)		18 (11.9)	23 (16.1)	5 (19.2)	
PP	5 (7.2)	2 (2.0)	0 (0.0)		6 (4.0)	3 (2.1)	2 (7.7)	
Education, years, mean (SD)	11.9 (3.8)	11.2 (3.6)	7.8 (3.4)	0.002[a]	11.7 (3.5)	11.1 (3.8)	9.8 (4.2)	0.08[a]
EDSS score, median (range)	2.5 (0–7)	3.5 (0–8.5)	5.8 (2–8.5)	< 0.002[a]	2.5 (0–8.5)	2.5 (0–8.5)	3.5 (1–7.5)	0.13[a]
Beck score, median (range)	8.5 (0–39)	10.0 (0–63)	7.0 (0–18)	0.39[a]	9.0 (0–36)	11.0 (0–51)	11.0 (0–37)	0.36[a]
APOE, no. (%)								
ε4+	12 (17.1)	12 (12.4)	7 (43.8)	0.016[b]	19 (12.6)	21 (14.7)	3 (11.5)	0.83[b]
ε4–	58 (82.9)	85 (87.6)	9 (56.2)		132 (87.4)	122 (85.3)	23 (88.5)	

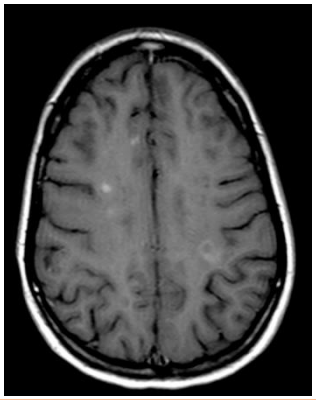
[a] Kruskal-Wallis test; [b] χ^2 test
All statistically significant variables were adjusted for Bonferroni correction

Males with MS and cognitive decline may be more likely to carry the apolipoprotein **ε4-ε4 risk allele** for dementia [Savettieri et al 2004].

Gender as MS modifier

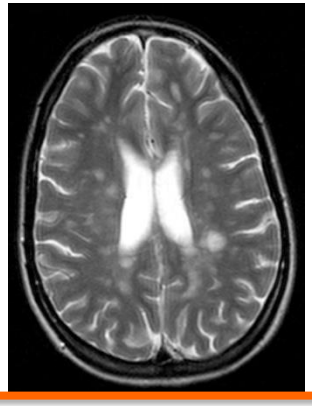
Gender difference in brain damage in MS

Mean No. Gd-enhancing lesions



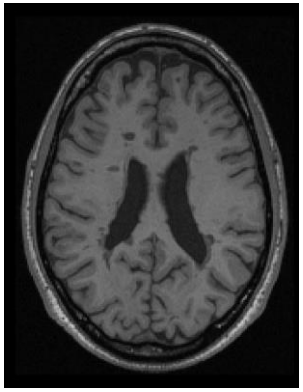
M vs F:
1.24 vs 1.86
p=0.01

Mean No. T2-hyperintense lesions



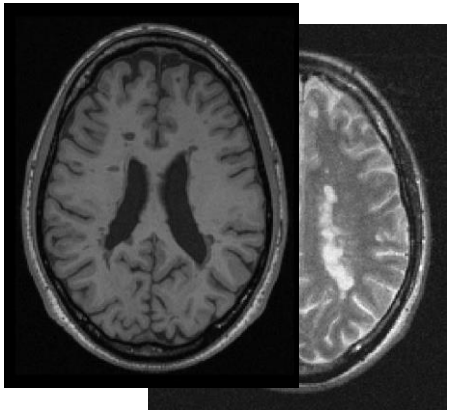
M vs F:
48.3 vs 50.1,
p=ns

Mean No. T1-hypointense lesions



M vs F:
19.1 vs 15.8
p=0.09

T1/T2 ratio: ***proportion of demyelinating lesions becoming "black holes"***



M vs F:
0.38 vs 0.29,
p=0.001

Men are prone to develop less inflammatory but more destructive lesions than women supporting a *modulation of MS pathological changes by gender*.

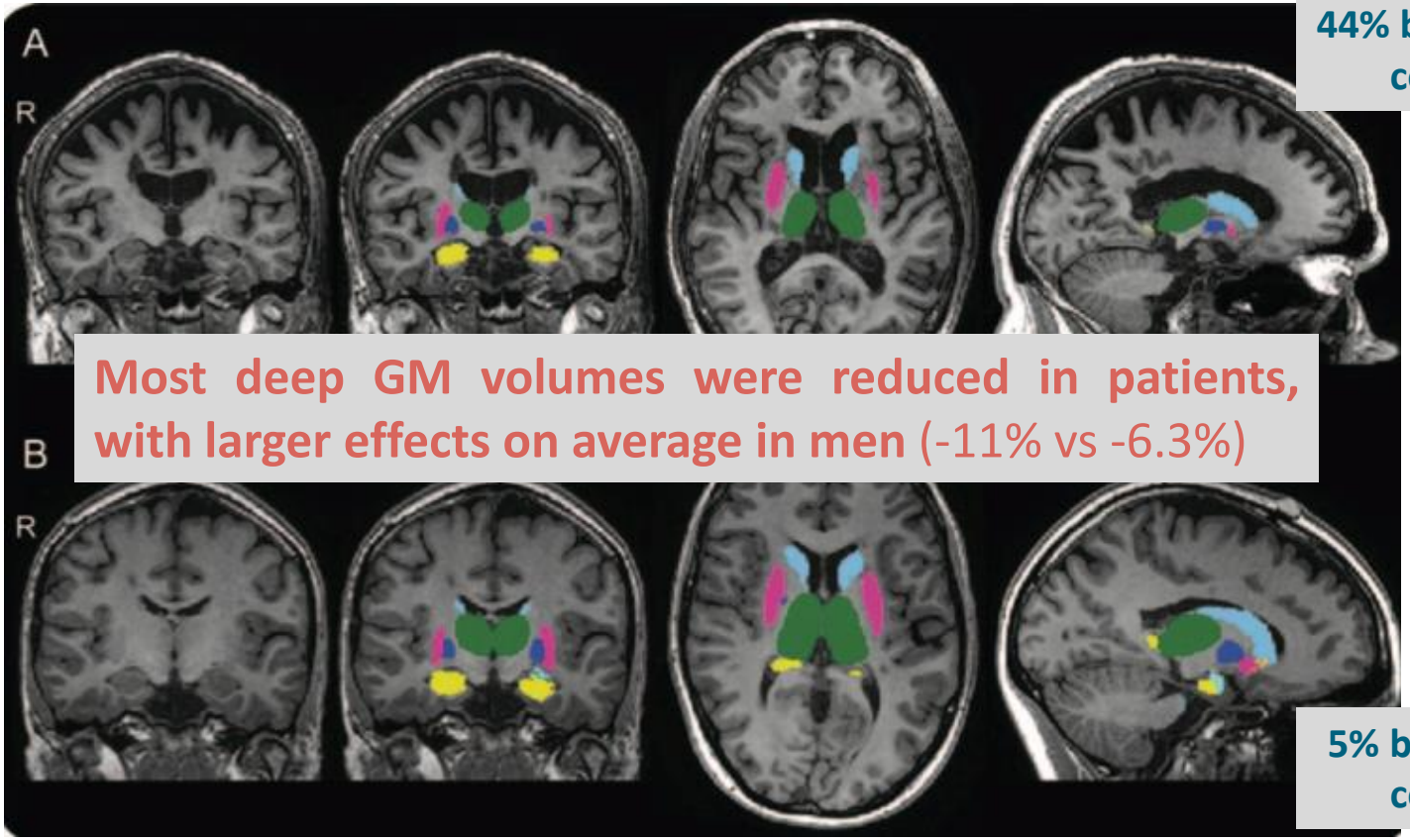
[Pozzilli C et al. 2003]

Gender as MS modifier

Gender difference in brain damage in MS

Male

Female



44% below male controls

Most deep GM volumes were reduced in patients, with larger effects on average in men (-11% vs -6.3%)

5% below male controls

120 pts
early inception cohort, 6 yrs postdiagnosis

Gray matter atrophy (particularly thalamic) is noted to be particularly harmful for cognition, functional connectivity and network efficiency.

Gender as MS modifier

Gender difference in brain damage in MS

J Neurol (2012) 259:2105–2110
DOI 10.1007/s00415-012-6464-z

ORIGINAL COMMUNICATION

Higher Hcy levels are associated with male sex, suggesting a role of Hcy in neurodegenerative processes of MS, which are prominent in male patients

Elevated plasma homocysteine levels in patients with multiple sclerosis are associated with male gender

Stefano Zoccolella · Carla Tortorella · Pietro Iaffaldano · Vita Direnzo · Mariangela D'Onghia · Damiano Paolicelli · Paolo Livrea · Maria Trojano

OPEN ACCESS Freely available online

PLoS

Low Serum Urate Levels Are Associated to Female Gender in Multiple Sclerosis Patients

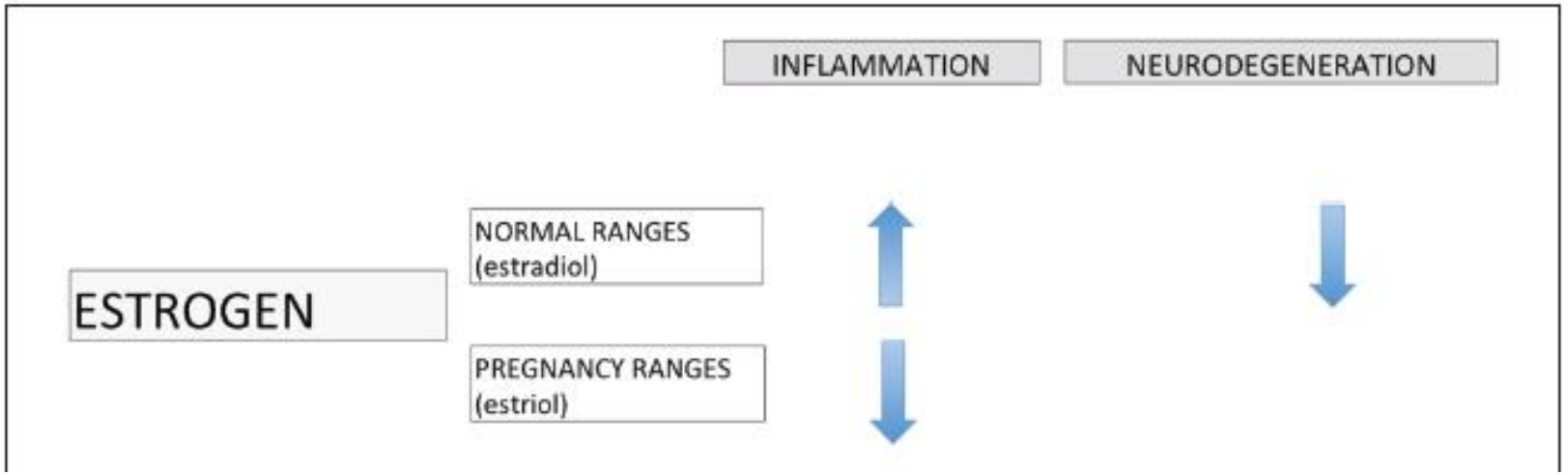
Stefano Zoccolella, Carla Tortorella, Pietro Iaffaldano, Vita Direnzo, Mariangela D'Onghia, Elena Luciannatelli, Damiano Paolicelli, Paolo Livrea, Maria Trojano*

- Uric acid is a natural anti-oxidant and a peroxynitrite scavenger.
- Low urate levels could be of significance in predominantly inflammatory phases of MS even at the early stage and mainly in females.

Gender as MS modifier

Estrogen and their effects on brain pathology

Hormones reflect a double-edged sword



No convincing data that low level of ovarian hormones are disease promoting

Estrogens in high levels, especially estriol in the levels found in pregnancy, can reduce EAE (Kim S. *Neurology* 1999)

Oral ethinylestradiol suppresses EAE (Subramanian S, *J. Immunol.* 2003)

Puberty and MS

Several lines of evidence suggest that the pubertal period may be a key regulator of MS risk.

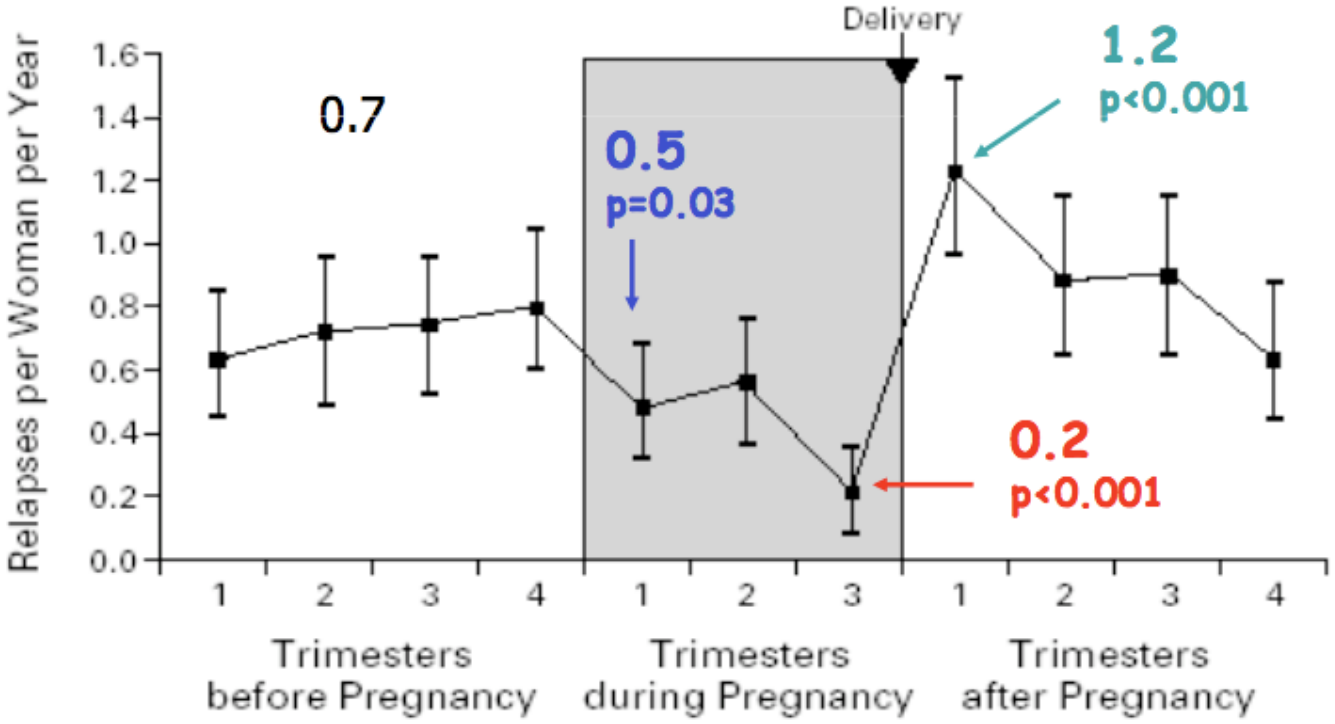
1. Dramatic female-specific increase in MS risk after puberty [Chitnis T. et al. 2013]

2. Earlier puberty is a risk factor for MS [Chitnis, 2013]

3. Being >15 years of age at menarche was associated with a lower risk of MS/CIS [Langer-Gould A. et al 2018]

Pregnancy and relapse

Relapse Rate per woman per year for each 3-month period before, during, and after pregnancy (227 pregnancy resulting in a live birth)



[Confavreux 1998]

Pregnancy:
downregulation of cellular immune responses

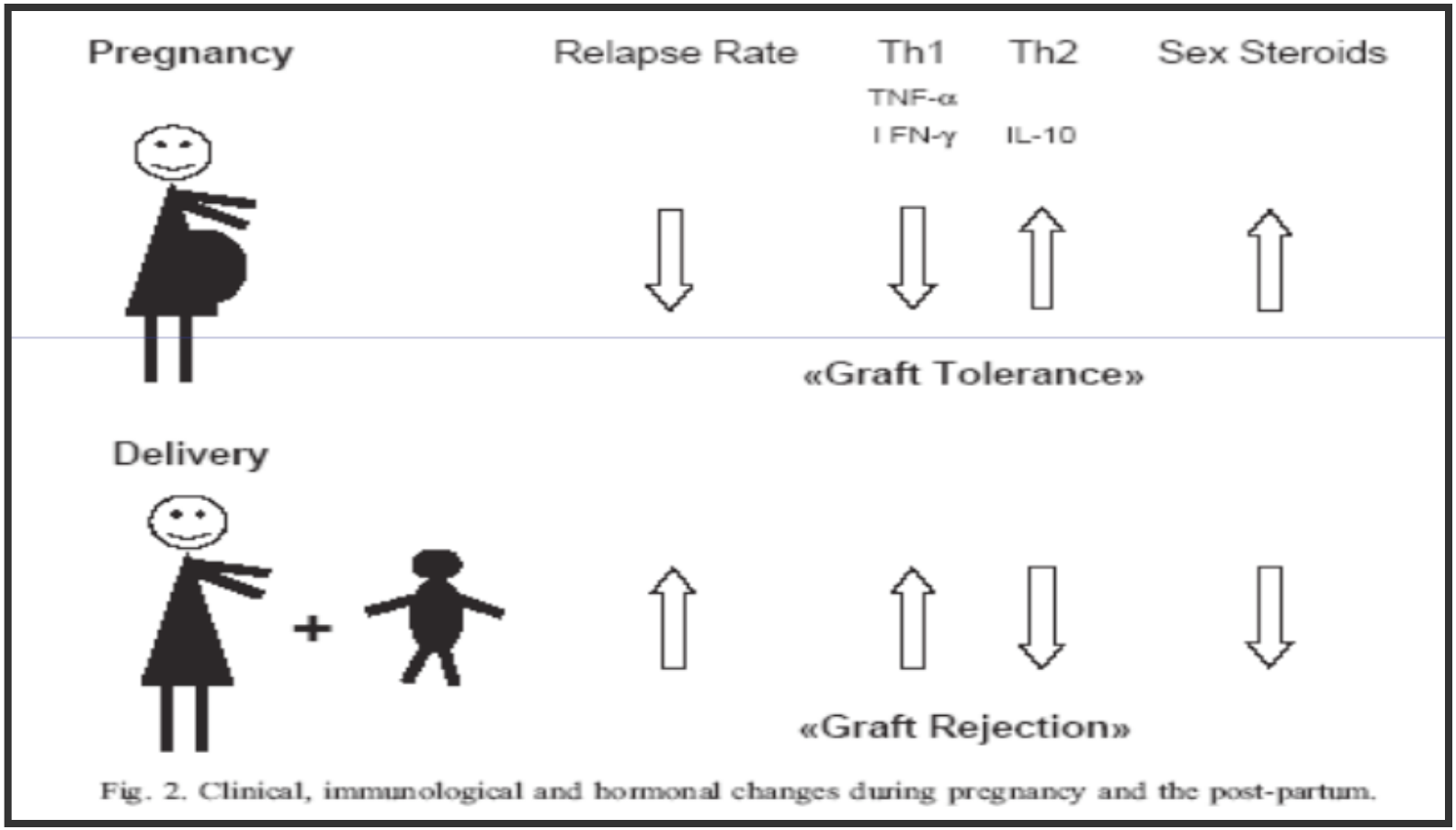
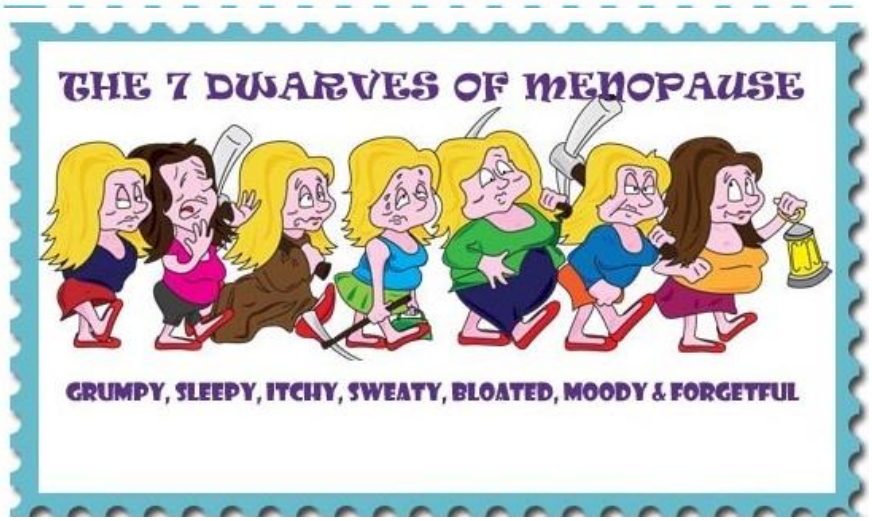


Fig. 2. Clinical, immunological and hormonal changes during pregnancy and the post-partum.

Menopause and MS



In late onset MS (LOMS), MS (onset after age 50)

- The F:M sex ratio decreases to a range of 1.4–1.9 to 1
- There are fewer differences between males and females in the time to reach an EDSS of 6, even when stratifying for disease type.

[Bove R. et al. 2012]

Gender as MS modifier

Testosterone and their effects on brain pathology

- Data showing that **high level of testosterone** in **young male mice is protective**: removal of physiological levels of testosterone from male mice via castration increased disease susceptibility
- In **male** the onset of MS tends to be relatively later in life (30s-40s), coinciding with the beginning of the decline in bioavailable testosterone
- It has been reported the 24% of **male MS** patients had significantly lower levels of testosterone as compared to age matched healthy men

Thus has been postulated that relatively **high levels of testosterone** in young men may be playing a temporarily protective role in men who are otherwise genetically predisposed to developing MS

[Palaszynsky Neuroimmunol 2004, Liva J. Immunol 2001, Voskull Lancet Neurol 2011]

Gender as MS modifier

Low testosterone is associated with disability in men with multiple sclerosis

R Bove, A Musallam, BC Healy, K Raghavan, BI Glanz, R Bakshi, H Weiner, PL De Jager, KK Miller and T Chitnis

Multiple Sclerosis Journal

1–9

DOI: 10.1177/1352458514527864

In **men** with MS :

an association of **low baseline testosterone** levels with **disease severity** and with longitudinal **changes in cognition** was found, suggesting that testosterone treatment may potentially have a higher benefit/risk ratio in MS patients, relative to a healthy population

Outline

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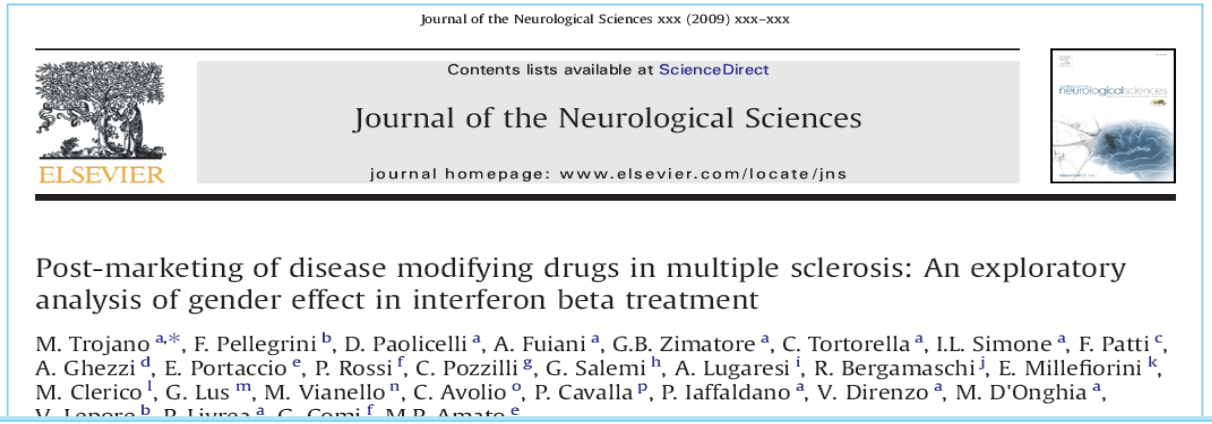
- **Sex differences in treatment response**
- Impact of exogenous hormones on MS immune response and disease activity

Gender and treatment response



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Post-marketing of disease modifying drugs in multiple sclerosis: An exploratory analysis of gender effect in interferon beta treatment

M. Trojano [a,*], F. Pellegrini [b], D. Paolicelli [a], A. Fuiani [a], G.B. Zimatore [a], C. Tortorella [a], I.L. Simone [a], F. Patti [c], A. Ghezzi [d], E. Portaccio [e], P. Rossi [f], C. Pozzilli [g], G. Salemi [h], A. Lugaresi [i], R. Bergamaschi [j], E. Millefiorini [k], M. Clerico [l], G. Lus [m], M. Vianello [n], C. Avolio [o], P. Cavalla [p], P. Iaffaldano [a], V. Direnzo [a], M. D'Onghia [a], V. Lepore [b], P. Livrea [a], G. Comi [f], M.P. Amato [e]

Male patients respond to IFNbeta treatment differently than females

2570 IFN beta treated MS pts (in 15 Italian MS Centers). FU: 7 years

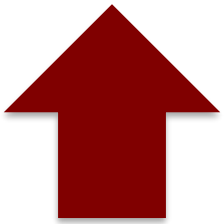
PS-matched Cox regression time

Males vs Females

Endpoint	IRR	95% CI	p-value
Time to 1st bout	0.87	0.77-0.97	0.0164
Time to 1 point EDSS progression	1.17	0.98-1.40	0.0882

Gender and treatment response

Male patients respond to IFNbeta treatment differently than females



a. **Differences in immune response and inflammatory reaction between sex (>in women), higher susceptibility to degenerative processes in men might explain the different efficacy of the treatment**

b. **Moreover modification and increase of estrogen and progesteron receptors induced by IFNβ treatment might play, also, a role in the different response between females and males**

Gender and treatment response

Females respond to Natalizumab treatment better than males

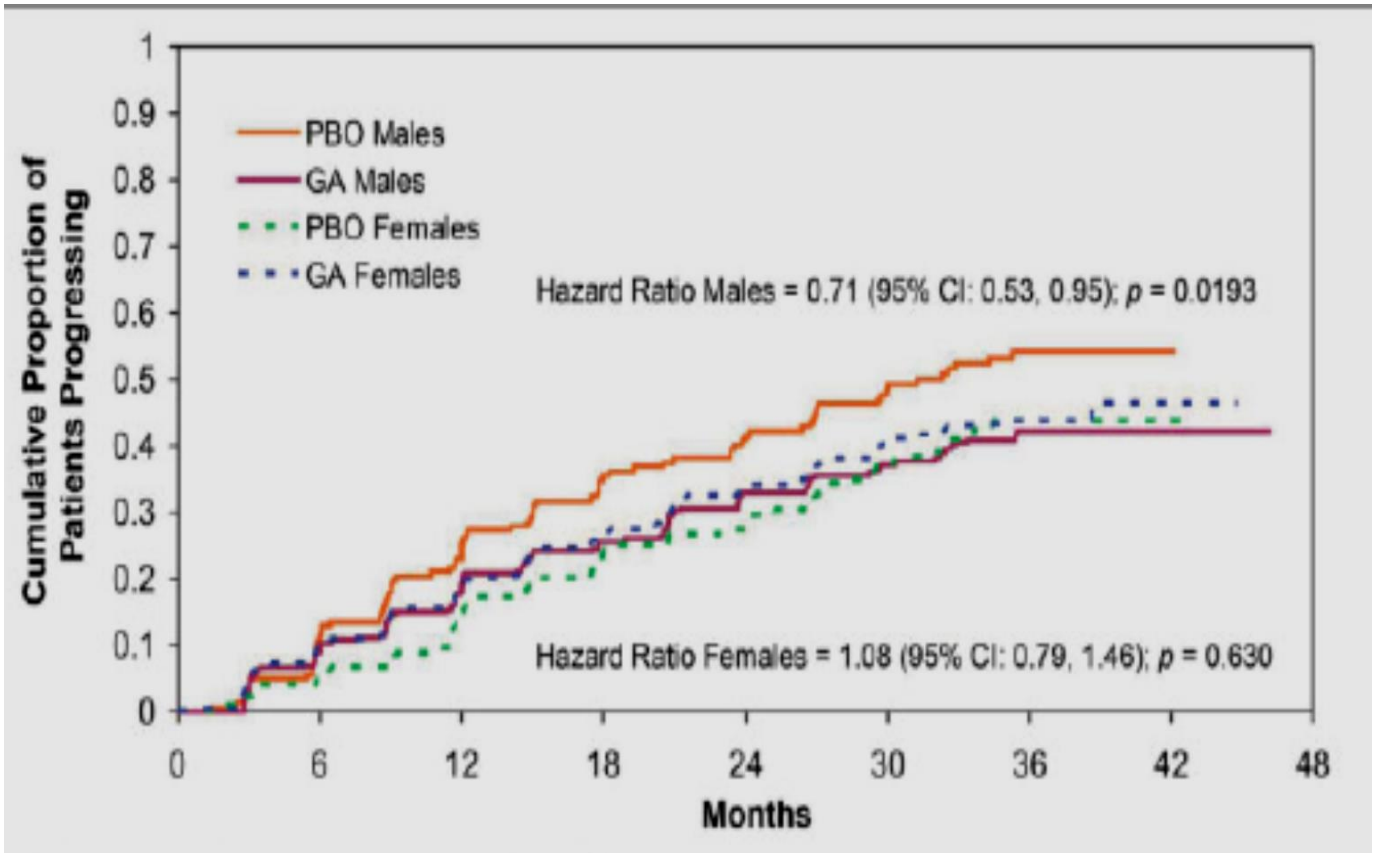
In SENTINEL study, natalizumab significantly reduced the risk of sustained disability progression in the following subgroups:

- or = 9 T2 lesions at baseline,
- > or = 1 Gd+ lesions at baseline,
- **female patients ***
- patients < 40 years of age.

*The authors did not control for disease duration nor baseline EDSS in these analyses.

[Hutchinson et al. Ann. Neurol. 2007]

Gender and treatment response



Results in **male** patients demonstrated that **GA** significantly delayed time to sustained progression of accumulated disability compared with PBO treatment (p=0.01)

[Wolinsky JS et al. Ann. Neurol. 2007]

Outline

- Sex differences in susceptibility to MS
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- Sex differences in treatment response
- **Impact of exogenous hormones on MS immune response and disease activity**

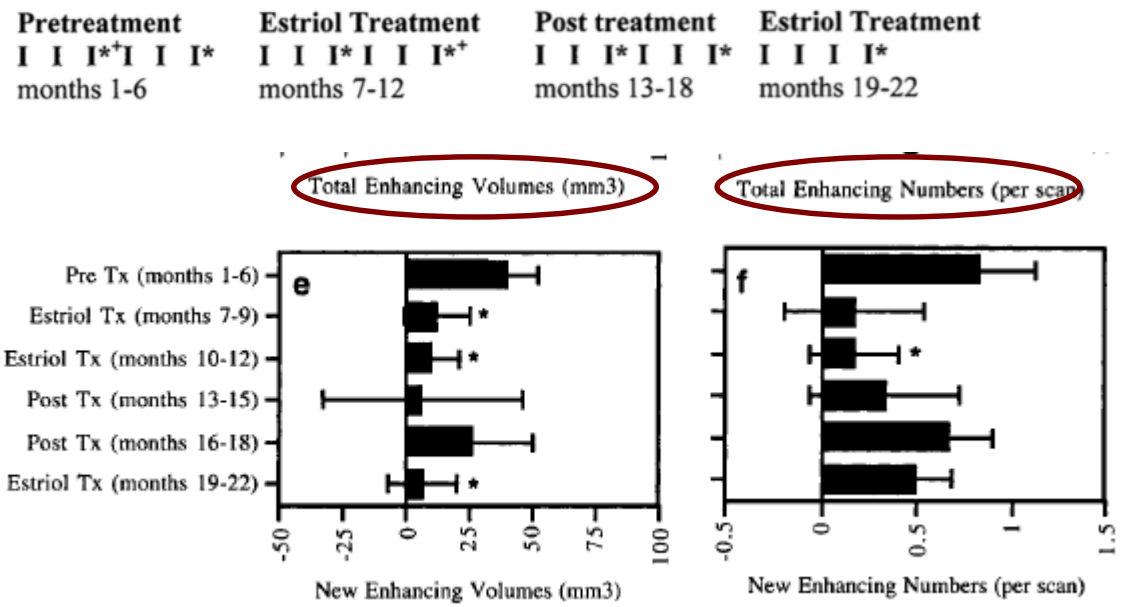
Exogenous hormones to treat MS

Treatment of Multiple Sclerosis with the Pregnancy Hormone Estriol

Nancy L. Sicotte, MD,[1] Stephanie M. Liva, PhD,[1] Rochelle Klutch, RN,[1] Paul Pfieffer, BS,[1] Seth Bouvier, BS,[1] Sylvia Odesa, BS,[1] T. C. Jackson Wu, MD, PhD,[2] and Rhonda R. Voskuhl, MD[1]

Ann Neurol 2002;52:421–428

A first pilot trial of 8 mg oral oestriol per day



Relapsing Remitting pts treated with oral estriol (8 mg/day) demonstrated **decreased gd-enhancing lesion numbers and volumes on MRI**.

When estriol treatment was stopped, enhancing lesions increased to pretreatment levels

Exogenous hormones to treat MS

ORIGINAL CONTRIBUTION

Testosterone Treatment in Multiple Sclerosis

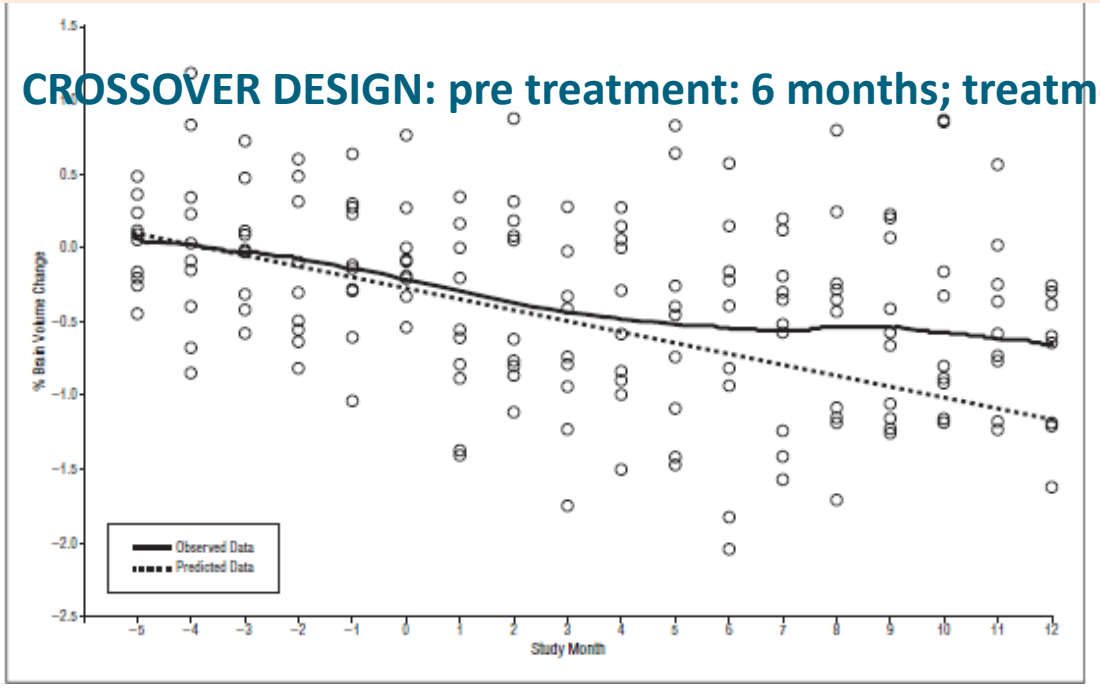
A Pilot Study

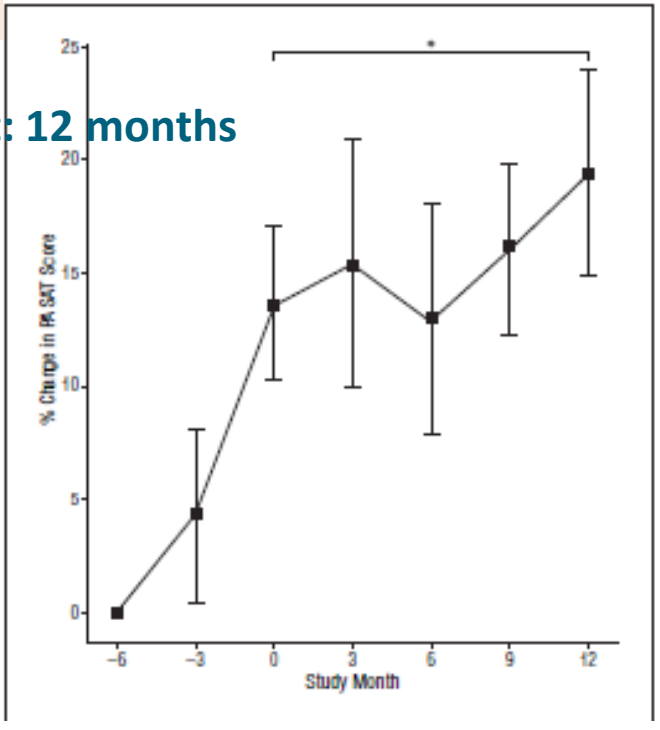
Nancy L. Sicotte, MD; Barbara S. Giesser, MD; Vinita Tandon, MD; Ricki Klutch, RN; Barbara Steiner, RN; Ann E. Drain, BS; David W. Shattuck, PhD; Laura Hull, BS; He-Jing Wang, PhD; Robert M. Elashoff, PhD; Ronald S. Swerdloff, MD; Rhonda R. Voskuhl, MD

Arch Neurol. 2007;64:683-688

One year of treatment with testosterone gel was associated with improvement in cognitive performance (*P =.008) and a slowing of brain atrophy (p.001).*

CROSSOVER DESIGN: pre treatment: 6 months; treatment: 12 months



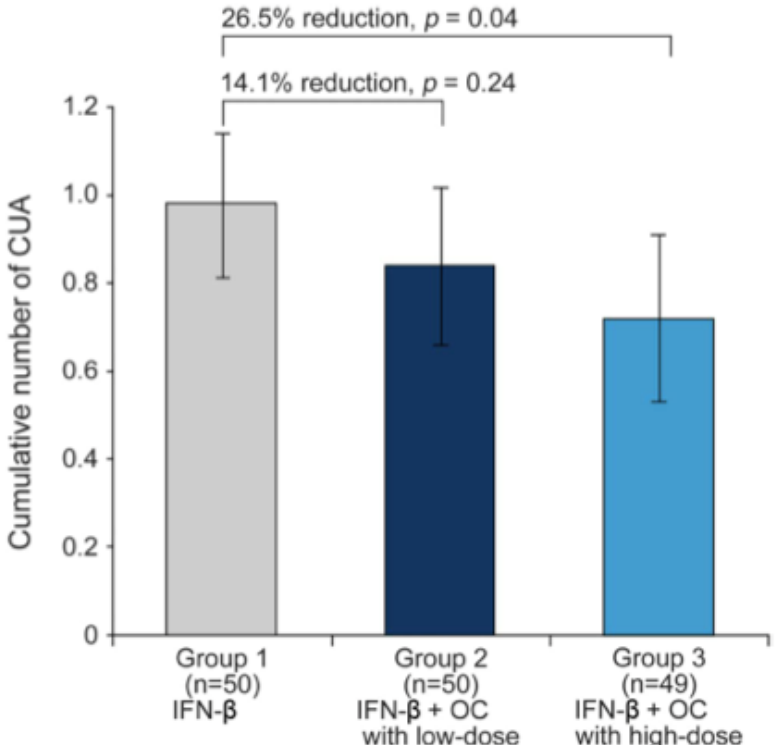


Exogenous hormones to treat MS

Oral contraceptives combined with interferon β in multiple sclerosis

OPEN

Figure 2 Primary endpoint: Cumulative number of combined unique active (CUA) lesions at week 96



IFN beta

+

ethinylestradiol 20 mg and desogestrel 150 mg

ethinylestradiol 40 mg and desogestrel 125 mg

24 months follow-up

[Pozzilli Cet al. Neurol Neuroiflamm. 2015]

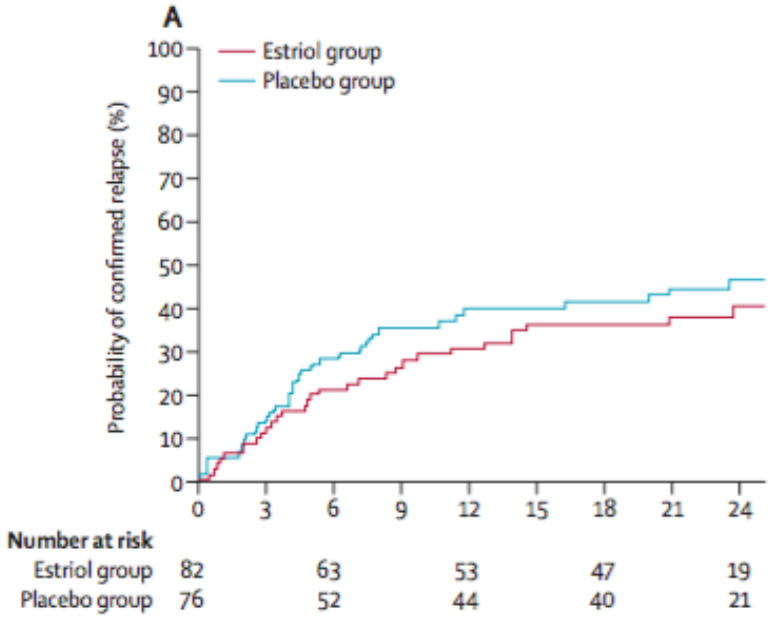
Exogenous hormones to treat MS

Estriol combined with glatiramer acetate for women with relapsing-remitting multiple sclerosis: a randomised, placebo-controlled, phase 2 trial

Lancet Neurol 2016; 15: 35–46

Rhonda R Voskuhl, HeJing Wang, T C Jackson Wu, Nancy L Sicotte, Kunio Nakamura, Florian Kurth, Noriko Itoh, Jenny Bardens, Jacqueline T Bernard, John R Corboy, Anne H Cross, Suhayl Dhib-Jalbut, Corey C Ford, Elliot M Frohman, Barbara Giesser, Dina Jacobs, Lloyd H Kasper, Sharon Lynch, Gareth Parry, Michael K Racke, Anthony T Reder, John Rose, Dean M Wingerchuk, Allan J MacKenzie-Graham, Douglas L Arnold, Chi Hong Tseng, Robert Elashoff

- 164 patients:
  - 83 "estriol group": daily oral estriol (8 mg) + GA
  - 81 "placebo group": placebo + GA
- The primary endpoint was ARR after 24 months



"Estriol plus glatiramer acetate met our criteria for reducing relapse rates, and treatment was well tolerated over 24 months. These results warrant further investigation in a phase 3 trial".


تمبوكتو ٥٢ يومًا
TOMBOUCTOU 52 Jours

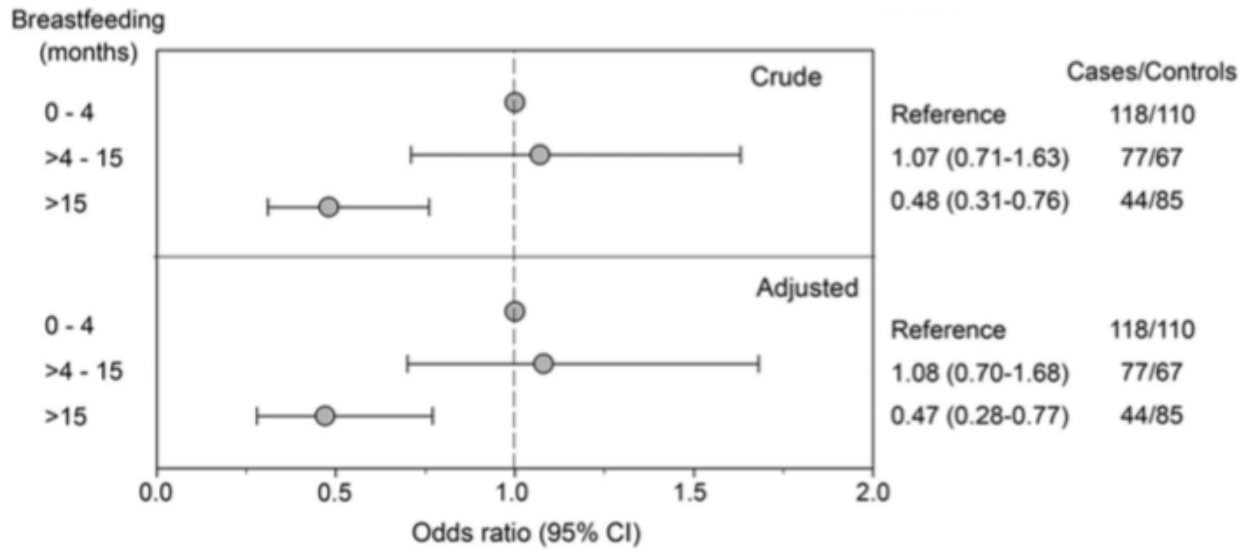
Breastfeeding

Breastfeeding, ovulatory years, and risk of multiple sclerosis

[Langer-Gould A. et al 2018]

397 patients
433 HC

Figure Duration of breastfeeding and MS



"a cumulative duration of breastfeeding for >15 months was associated with a reduced risk of MS/CIS" … ***Protective role of anovulation?***

Breastfeeding and post partum relapse

Table 1. Main studies assessing the influence of breastfeeding on disease activity in multiple sclerosis.[a]

	No of pregnancies	DMD	Assessment of exclusivity	Effect of breastfeeding	Other predictors of post-partum disease activity	MS-related covariates	Notes
Nelson et al.[28]	191	–	No	Neutral		Unadjusted	
Vukusic et al.[2]	210	–	No	Neutral		Unadjusted	Milder course in breastfeeding women
Langer-Gould et al.[10]	32	62% (interferons, GA)	Yes, with confirmation	Protective	–	Age; disease duration; relapse before pregnancy; DMD	Higher frequency of DMD in non-exclusive
Airas et al.[12]	61	Unknown	Yes, without confirmation	Neutral		Unadjusted	Milder course in exclusive breastfeeding women
Portaccio et al.[13]	302	46.4% (interferons, GA)	Yes, without confirmation	Neutral	Relapse before pregnancy; relapse during pregnancy	Age; age at MS onset; disease duration; EDSS at conception; DMD; relapse before pregnancy; relapse during pregnancy	Milder course in exclusive breastfeeding women
Hellwig et al.[11]	201	89% (interferons, GA, natalizumab)	Yes, with confirmation	Protective	Relapse during pregnancy	Age at onset; disease duration; DMD; relapse before pregnancy	Higher frequency of relapse during pregnancy and DMD in non-exclusive

DMD: disease modifying drugs; MS: multiple sclerosis; GA: glatiramer acetate; EDSS: Expanded Disability Status Scale.
[a]Full research articles published in international peer-reviewed journals, conducted on larger samples (≥30 subjects).

[Portaccio E, Amato MP 2019]

Gender as MS modifier

- **Sex differences in the regulation of the gut microbiome**

Sex Differences in the Gut Microbiome Drive Hormone-Dependent Regulation of Autoimmunity

Janet G. M. Markle,[1,2] Daniel N. Frank,[3] Steven
Robertson,[4] Leah M. Feazel,[3] Ulrike Rolle-Kamp
Kathy D. McCoy,[8] Andrew J. Macpherson,[8] Jay

"We have identified a **direct interaction** between **sex hormones** and **microbial exposures** and show that **microbiome manipulations can provoke testosterone-dependent protection from autoimmunity in a genetically high-risk rodent model".**