

Steroidal Receptor Signaling and Cardiovascular Function

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Estrogen Replacement Therapy: before and after the Women's Health Initiative (WHI)

Before WHI

Estradiol

Relaxing Factors ↑
Constricting Factors ↓
Anti-Oxidant ↑
Cholesterol ↓
RASS ↓
Antimitogenic Effects ↑

↓ **Cardiovascular Disease**

After WHI

Estradiol

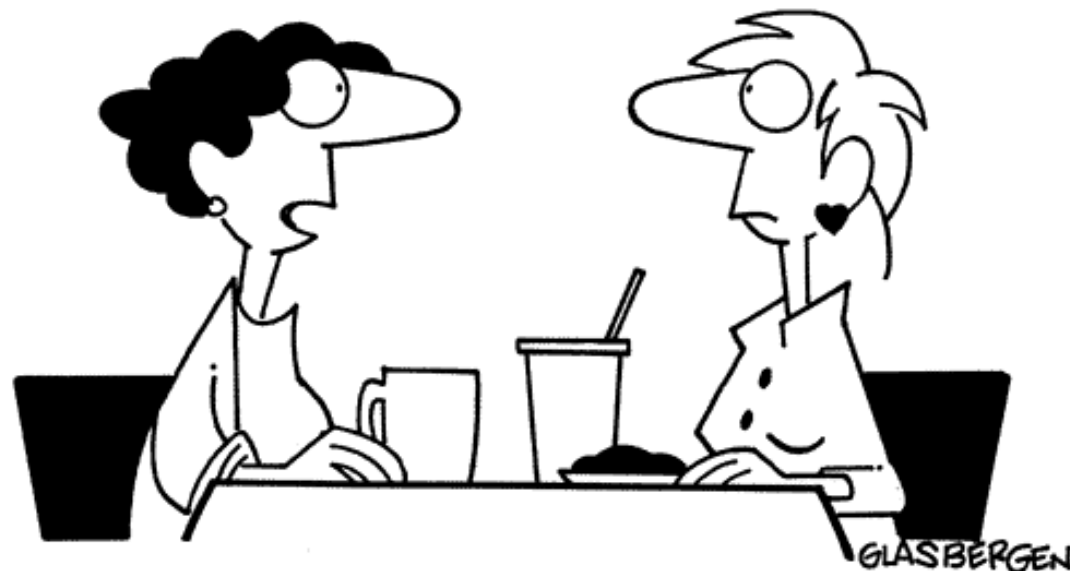
Venous thrombo-
embolism ↑
Stroke ↑
Coronary heart
disease ↑
Cholesterol Ø

↑ **Cardiovascular Disease**

WHI?

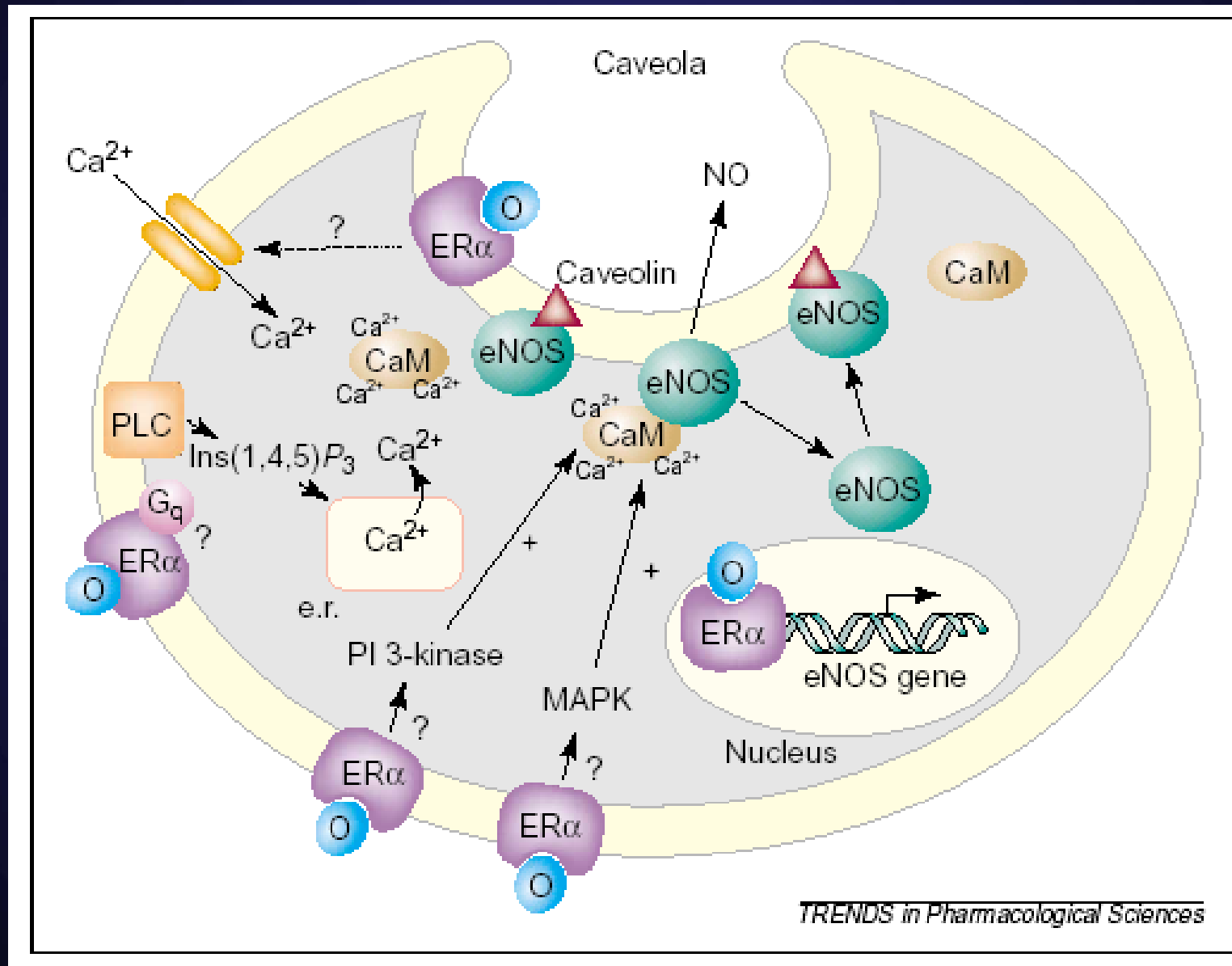
Equine Estrogen vs. Natural Estrogen

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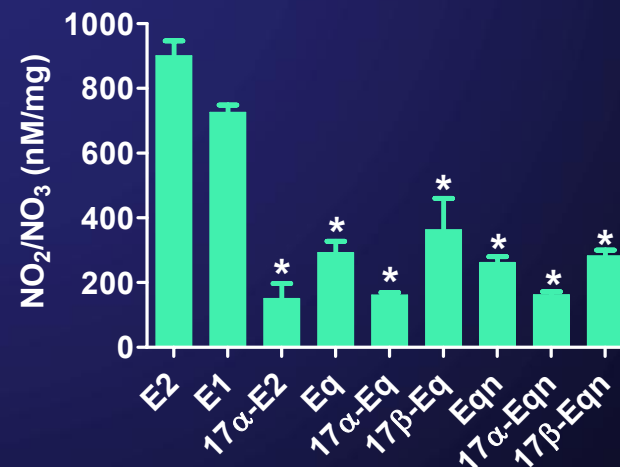
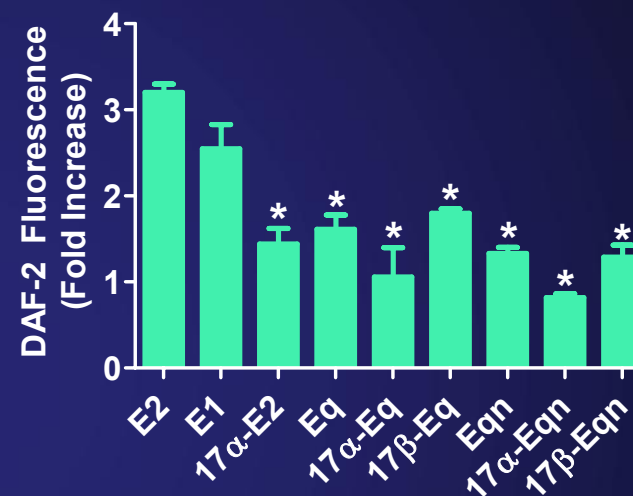
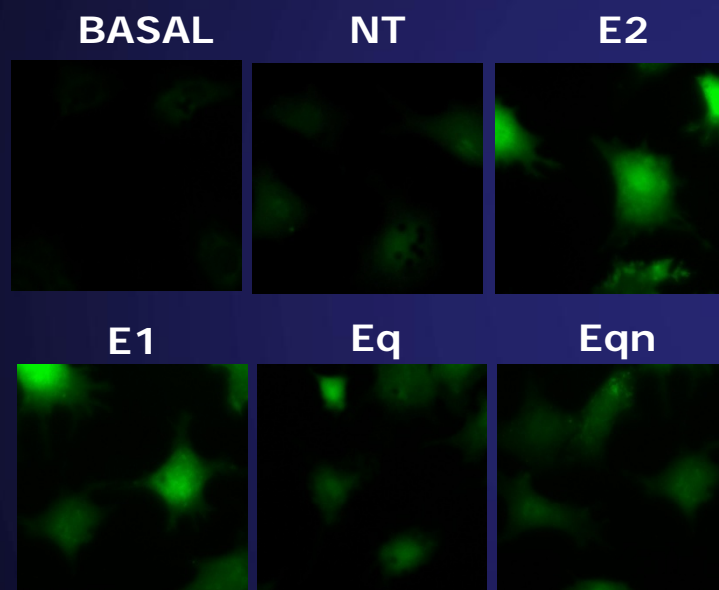


“I’m in an experimental program that treats menopause with ostrich hormones. Now I only get hot flashes when I’m laying an egg.”

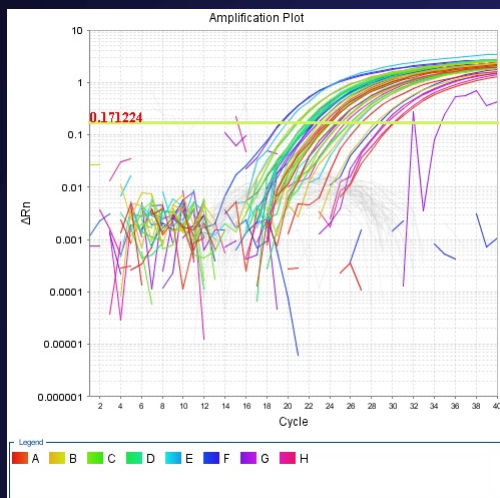
ESTROGEN REGULATION OF NITRIC OXIDE PRODUCTION



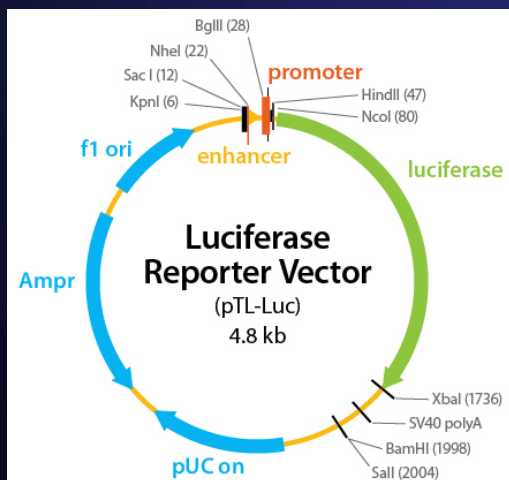
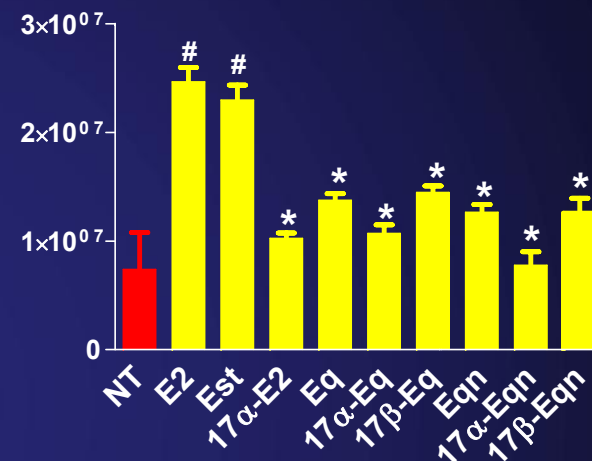
Estrogen-mediated NO production



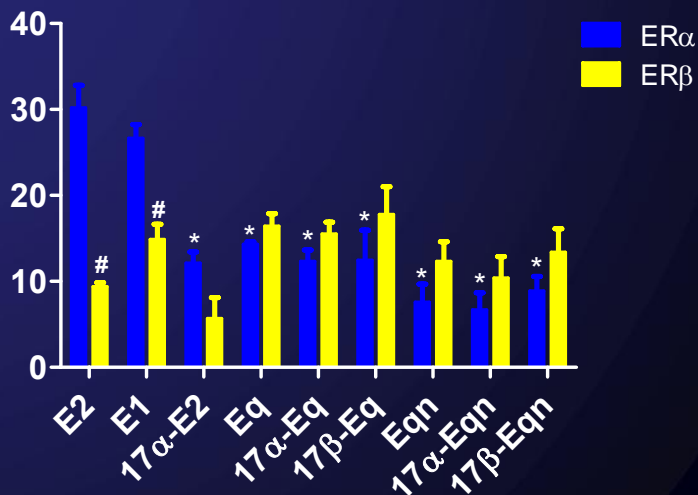
Estrogen effects on eNOS transcription



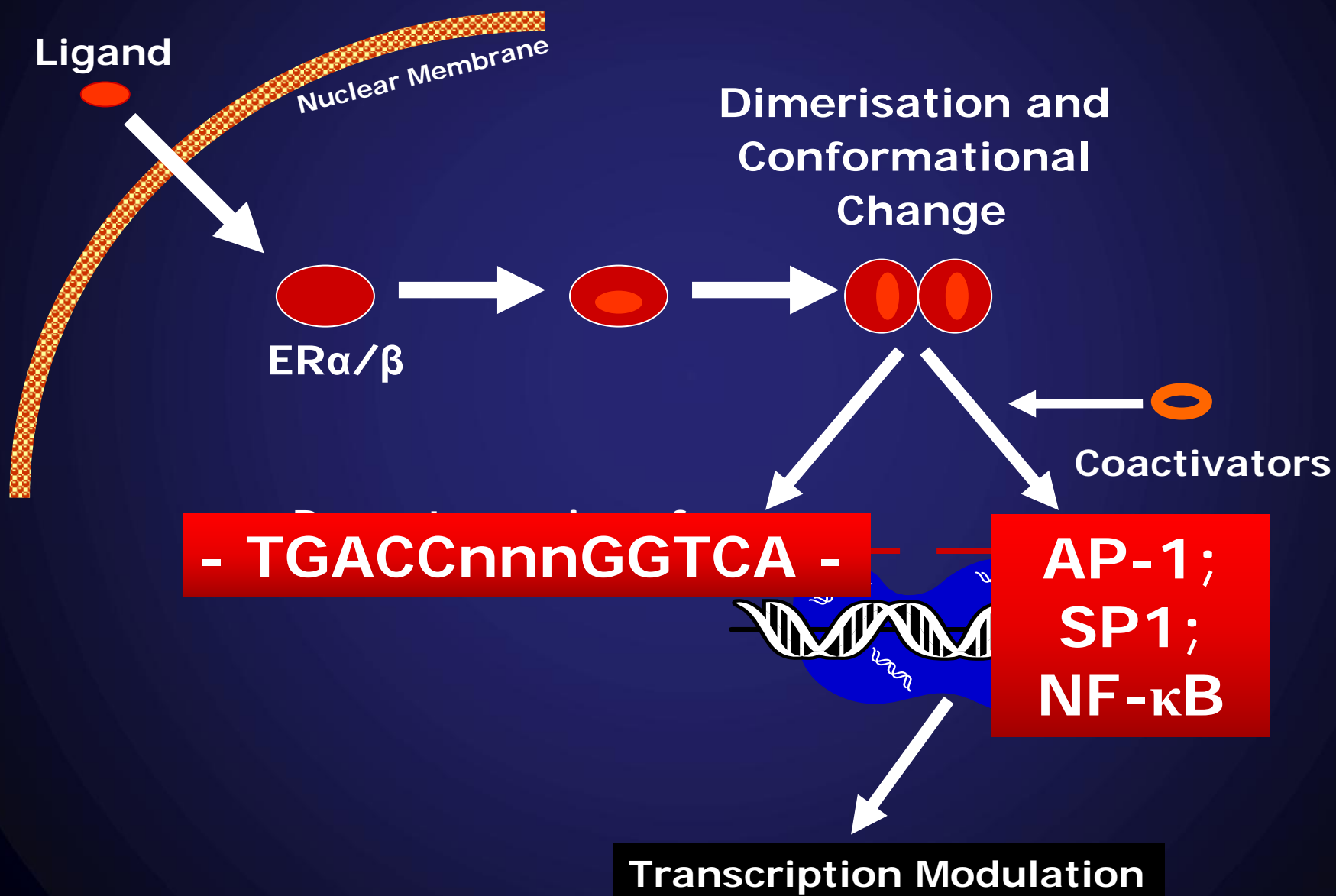
Normalized copies of eNOS
mRNA/ μg RNA



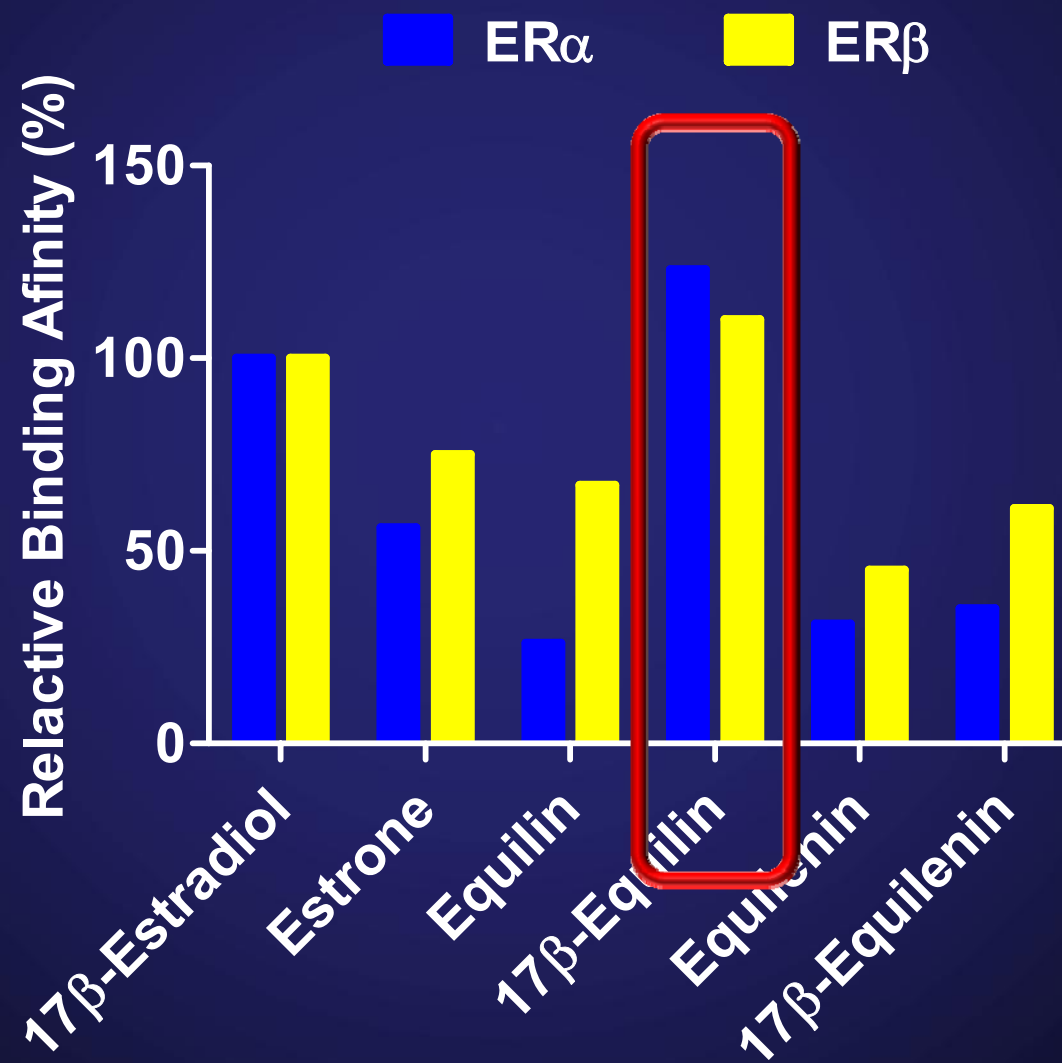
Luciferase Activity
(Fold Increase)



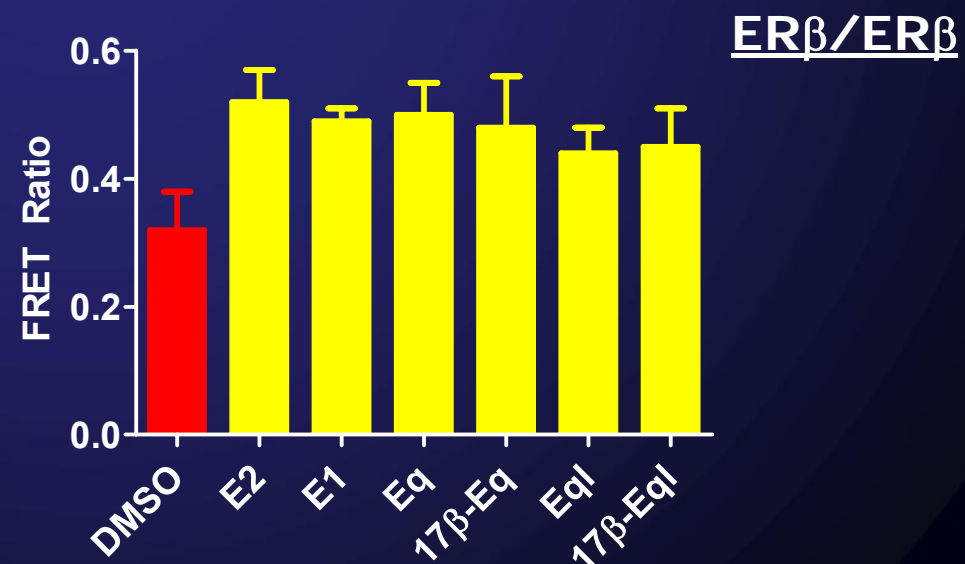
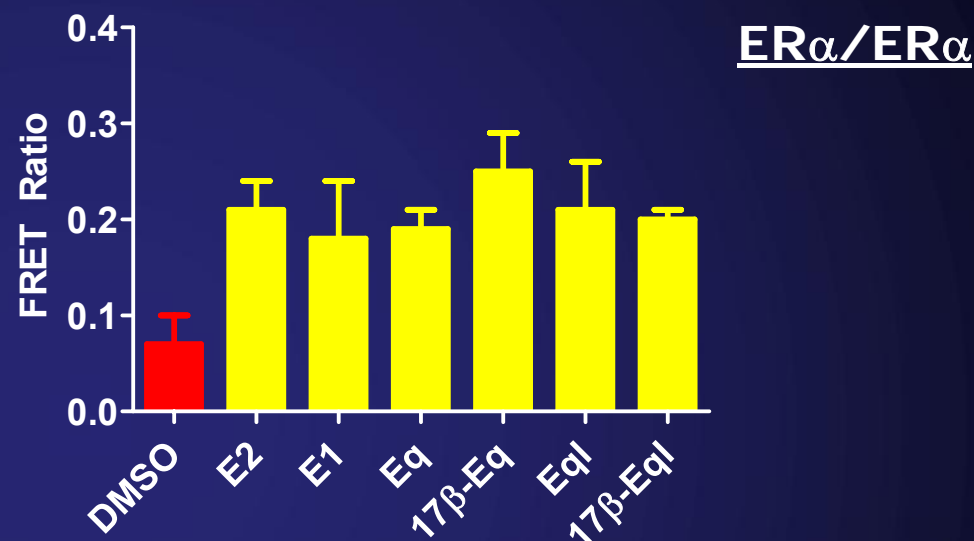
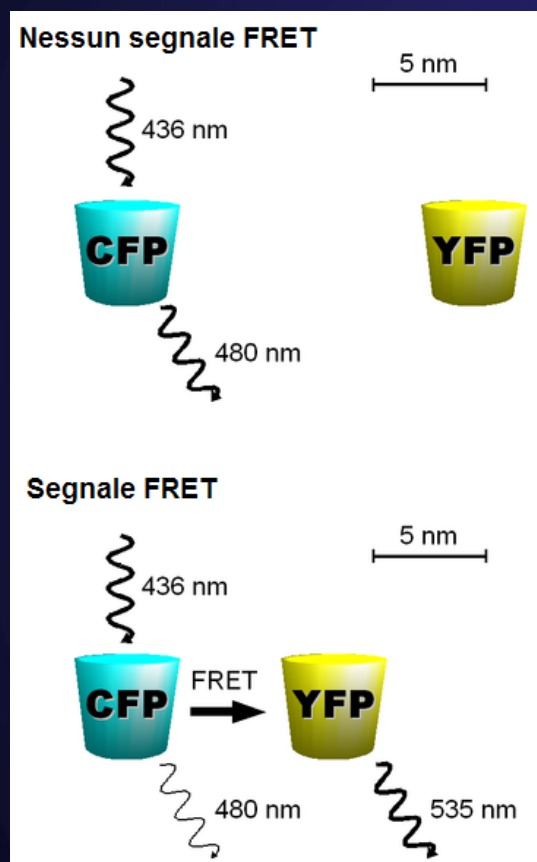
Estrogen Receptor Signaling



Relative Binding Affinity



ER Dimerization – FRET Analysis



eNOS Promoter

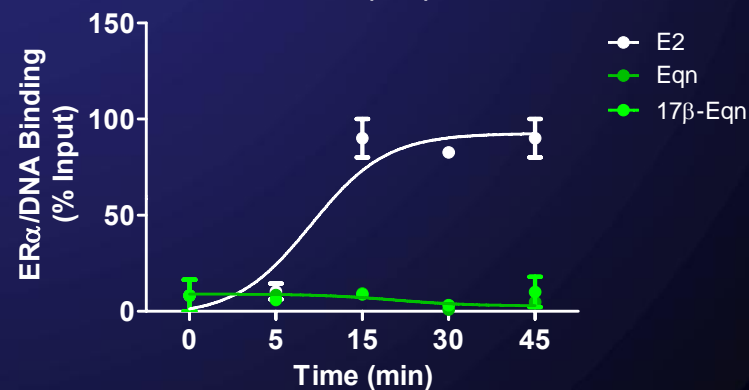
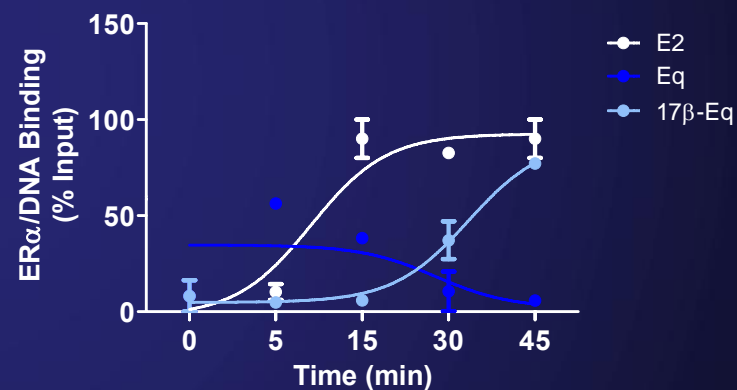
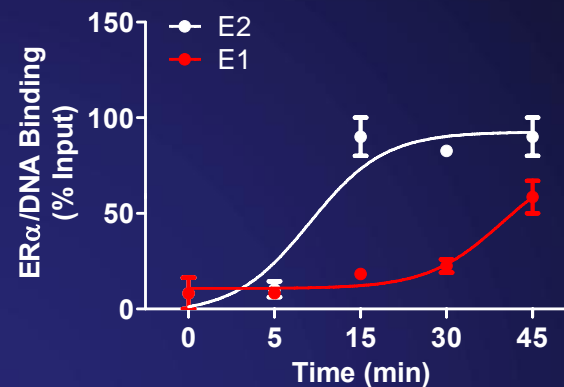
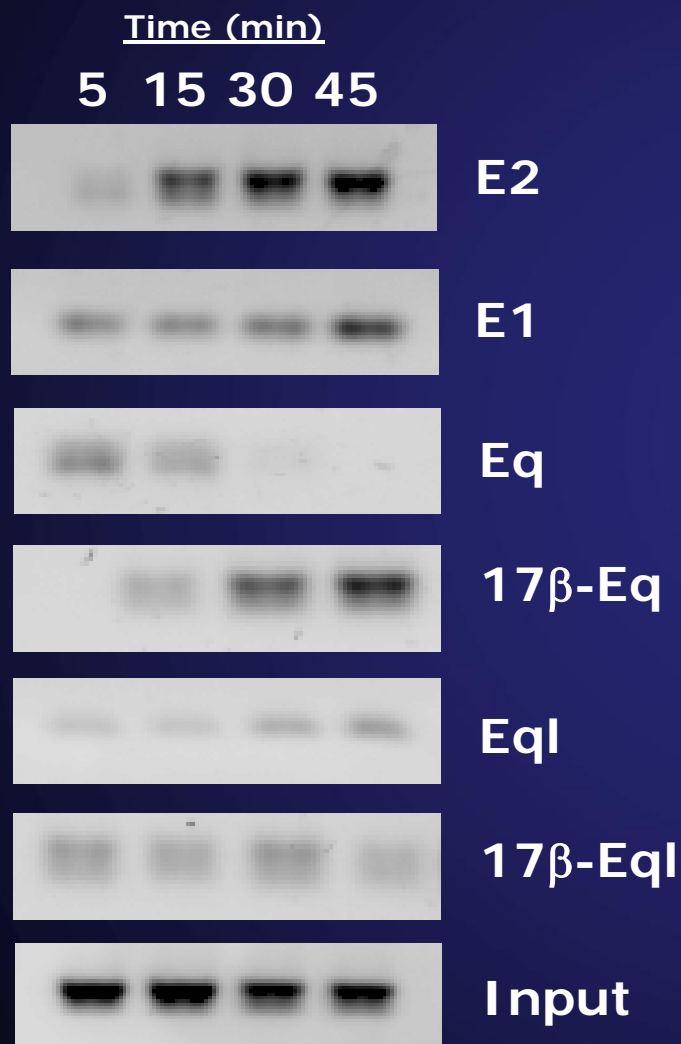
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-1550	TTAACTGGGA	CCTAGGAAAA	TGAGTCATCC	TTGGTCATGC	ACATTTCAAA
-1500	TGGTGGCTTA	ATATGGAAGC	CAGACTTGGG	TTCTGTTGTC	TCCTCCAGCA
-1450	TGGTAGAAGA	TGCCTGAAAA	GTAGGGGCTG	GATCCCATCC	CCTGCCTCAC
-1400	TGGGAAGGCG	AGGTGGTGGG	GTGGGGTGGG	GCCTCAGGCT	TGGGGTCATG
-1350	GGACAAAGCC	CAGGCTGAAT	GCCGCCCTTC	CATCTCCCTC	CTCCTGAGAC
-1300	AGGGGCAGCA	GGGCACACTA	GTGTCCAGGA	GCAGCTTATG	AGGCCCTTC
-1250	ACCCTCCATC	CTCCAAACT	GGCAGACCCC	ACCTTCTTGG	TGTGACCCCA
-1200	GAGCTCTGAG	CACAGCCCGT	TCCTTCCGCC	TGCCGGCCCC	CCACCCAGGC
-1150	CCACCCCAAC	CTTATCCTCC	ACTGCTTTTC	AGAGGAGTCT	GGCCAACACA
-1100	AATCCTCTTG	TTTGTTCGTC	TGTCTGTCTG	CTGCTCCTAG	TCTCTGCCTC
-1050	TCCCAGTCTC	TCAGCTTCGG	TTTCTTTCTT	AACTTTCTC	TCAGTCTCTG
-1000	AGGTCTCGAA	ATCACGAGGC	TTCGACCCCT	GTGGACCAGA	TGCCCAGCTA
-950	GTGGCCTTTC	TCCAGCCCCT	CAGATGACAC	AGAACTACAA	ACCCCAGCAT
-900	GCACTCTGGC	CTGAAGTGCC	TGGAGAGTGC	TGGTGTACCC	CACCTGCATT
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-800	CTCTTCCCTG	GCTGGCTGAC	CCTGCCTCAG	CCCTAGTCTC	TCTGCTGACC
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-400	CTGCCCAACC	CTCCAGGGAA	AGGCACACAG	GGGTGAGGCC	GAAGGCCCTT
-350	CCGTCTGGTG	CCACATCACA	GAAGGACCTT	TATGACCCCC	TGGTGGCTCT
-300	ACCCTGCCAC	TCCCCAATGC	CCCAGCCCCC	ATGCTGCAGC	CCCAGGGCTC
-250	TGCTGGACAC	CTGGGCTCCC	ACTTATCAGC	CTCAGTCCTC	ACAGCGGAAC
-200	CCAGGCGTCC	GGCCCCCCAC	CCTTCAGGCC	AGCGGGCGTG	GAGCTGAGGC
-150	TTTAGAGCCT	CCCAGCCGGG	CTTGTTCTCTG	TCCCATTTGTG	TATGGGATAG
-100	GGGCGGGGCG	AGGGCCAGCA	CTGGAGAGCC	CCCTCCCACT	GCCCCCTCCT
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+1	<u>AGCAGAGTGGACGCACAGTA</u>				

 ERE

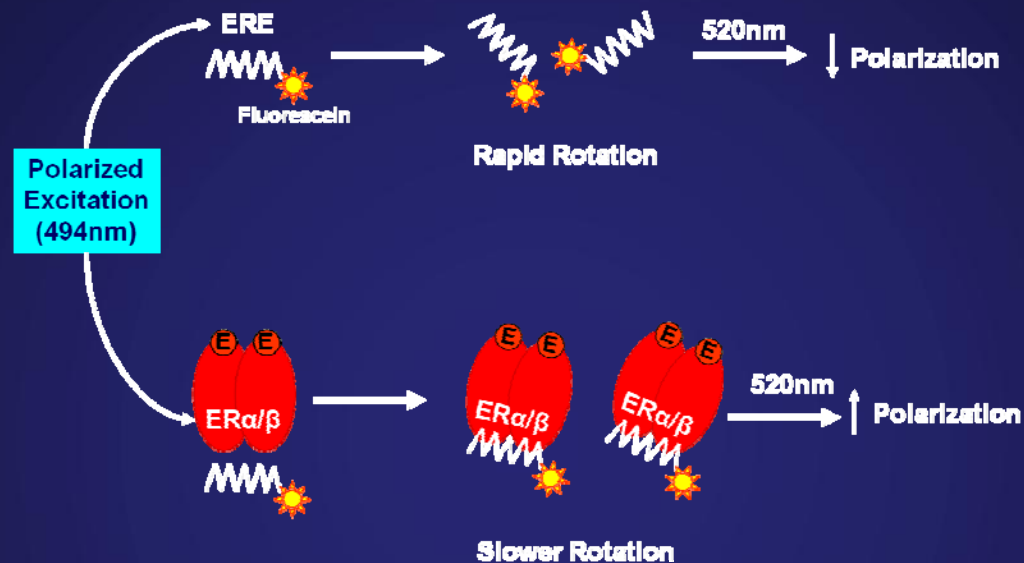
 AP-1

 SP-1

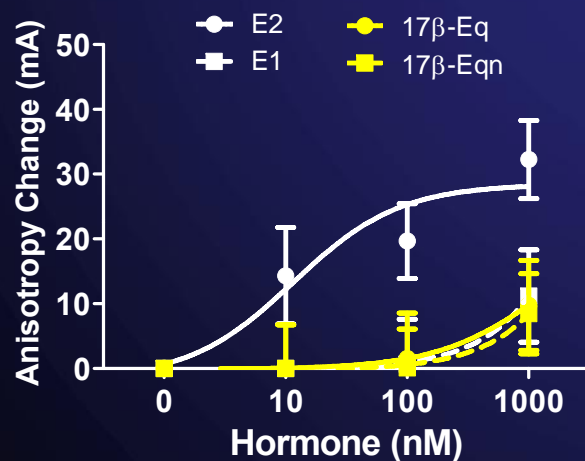
ChIP Analysis for ER α /DNA Interaction



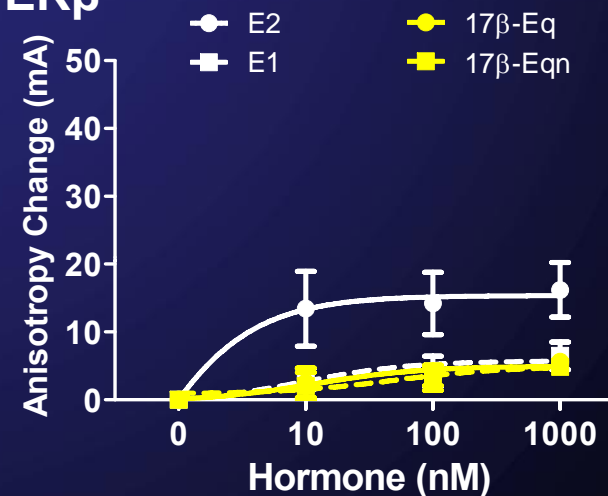
Fluorescence Anisotropy Analysis of $ER\alpha/\beta$ and SP1 Site Interaction



$ER\alpha$



$ER\beta$



Conclusions



✓ Equine estrogens modulate NO production less effectively than naturally occurring estrogens, as a result of their lower ability to activate the ER α -mediated increase of eNOS promoter activity and eNOS transcription.

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