Kinetic modelling helps to understand oxidative stress caused by an antioxidant

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Trisomy21 (Down Syndrome)

- Three copies of chromosome 21
- First described 1866 by J. L. Down
- Genetic basis discovered 1959
- Prevalence ca. 1:800
- Mental retardation and characteristic facial features
- Overexpression of the ca. 250 genes of chromosome 21
- Genes with possible relevance for Down Syndrome: SOD, COL6A1, ETS2, CAF1A, CBS, DYRK, CRYA1, GART, IFNAR

Superoxide Dismutase

$$\longrightarrow O_2^{-\bullet} \xrightarrow{\text{SOD}} H_2O_2 \xrightarrow{\text{cat / GPX}} H_2O$$

$$\frac{dO_2^{\bullet-}}{dt} = k_1 - k_2 \cdot SOD \cdot O_2^{\bullet-}$$

$$\frac{dH_2O_2}{dt} = k_2 \cdot SOD \cdot O_2^{\bullet-} - k_3 \cdot cat \cdot H_2O_2$$

$$O_{2,ss}^{\bullet-} = \frac{k_1}{k_2 \cdot SOD}$$

$$H_2O_{2,ss} = \frac{k_1}{k_3 \cdot cat}$$

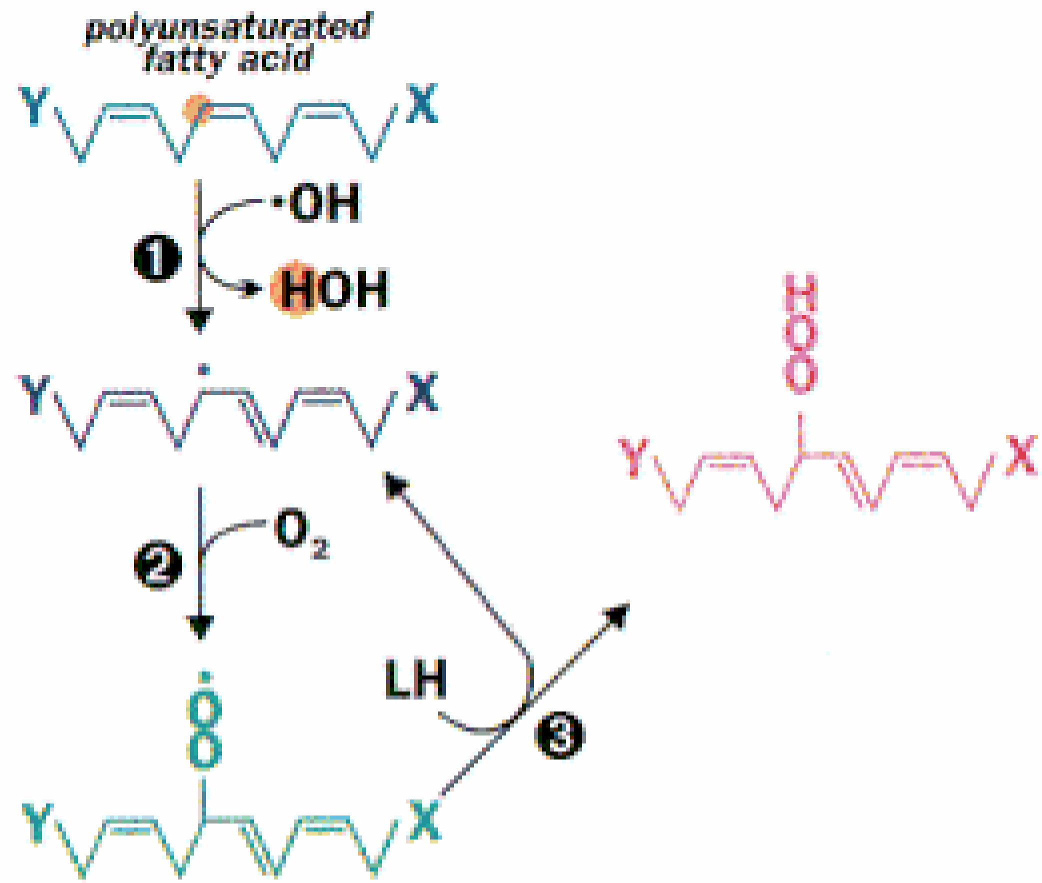
Negative Effects

- Increased lipid peroxidation in Trisomy21
- Increased lipid peroxidation in cell cultures
- Increased mortality in transgenic *D. melanogaster*
- Lipid peroxidation, diminished prostaglandin synthesis and serotonin uptake in transgenic mice.
- Bacteria overexpressing SOD showed increased paraquat sensitivity.

Explanations

- The reaction of H_2O_2 with CuZnSOD leads to hydroxyl radical generation.
- O_2^{*-} radicals might reduce membrane damage by acting as chain breakers.

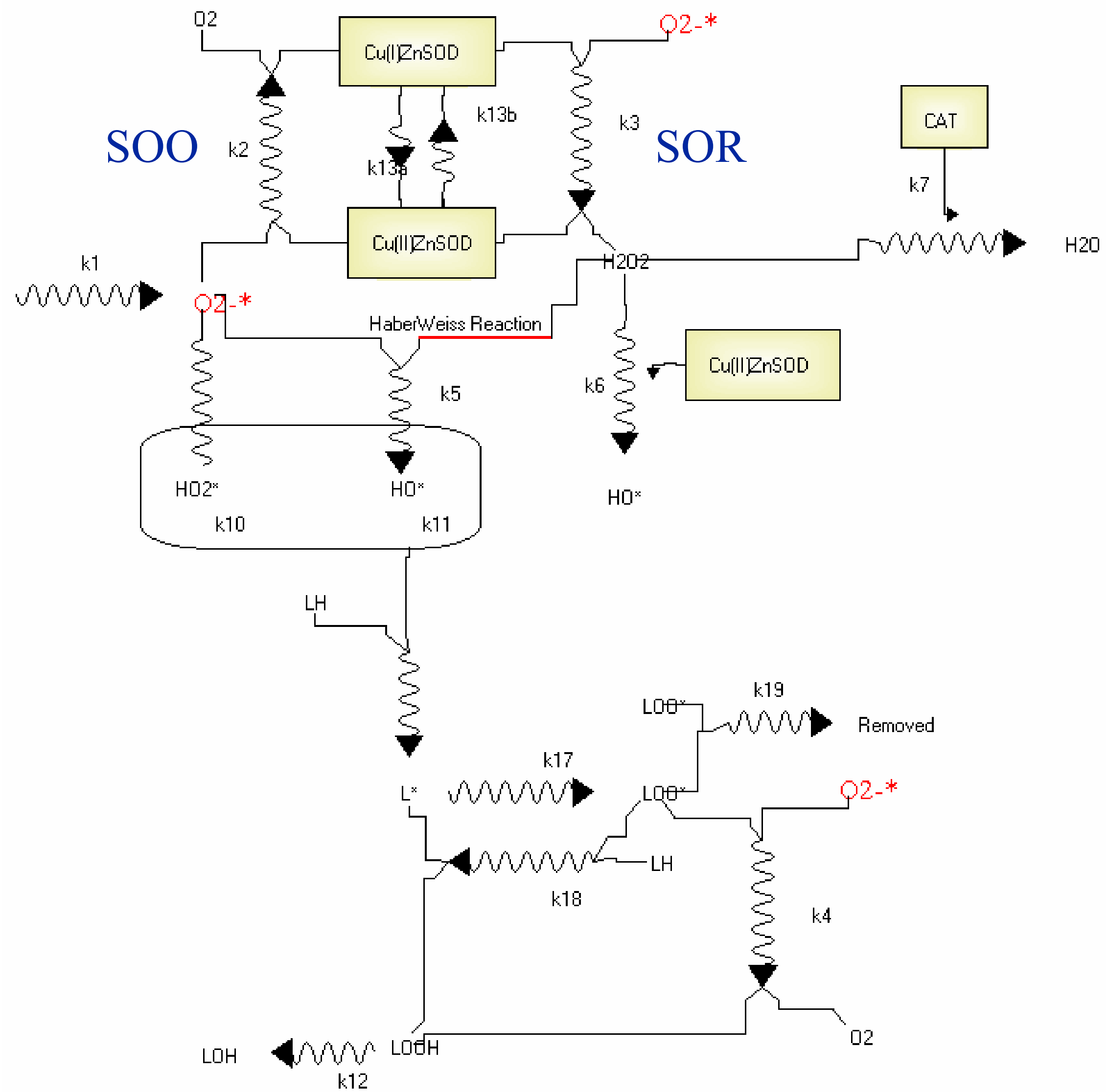
Lipid Peroxidation Chain Reaction



Explanations

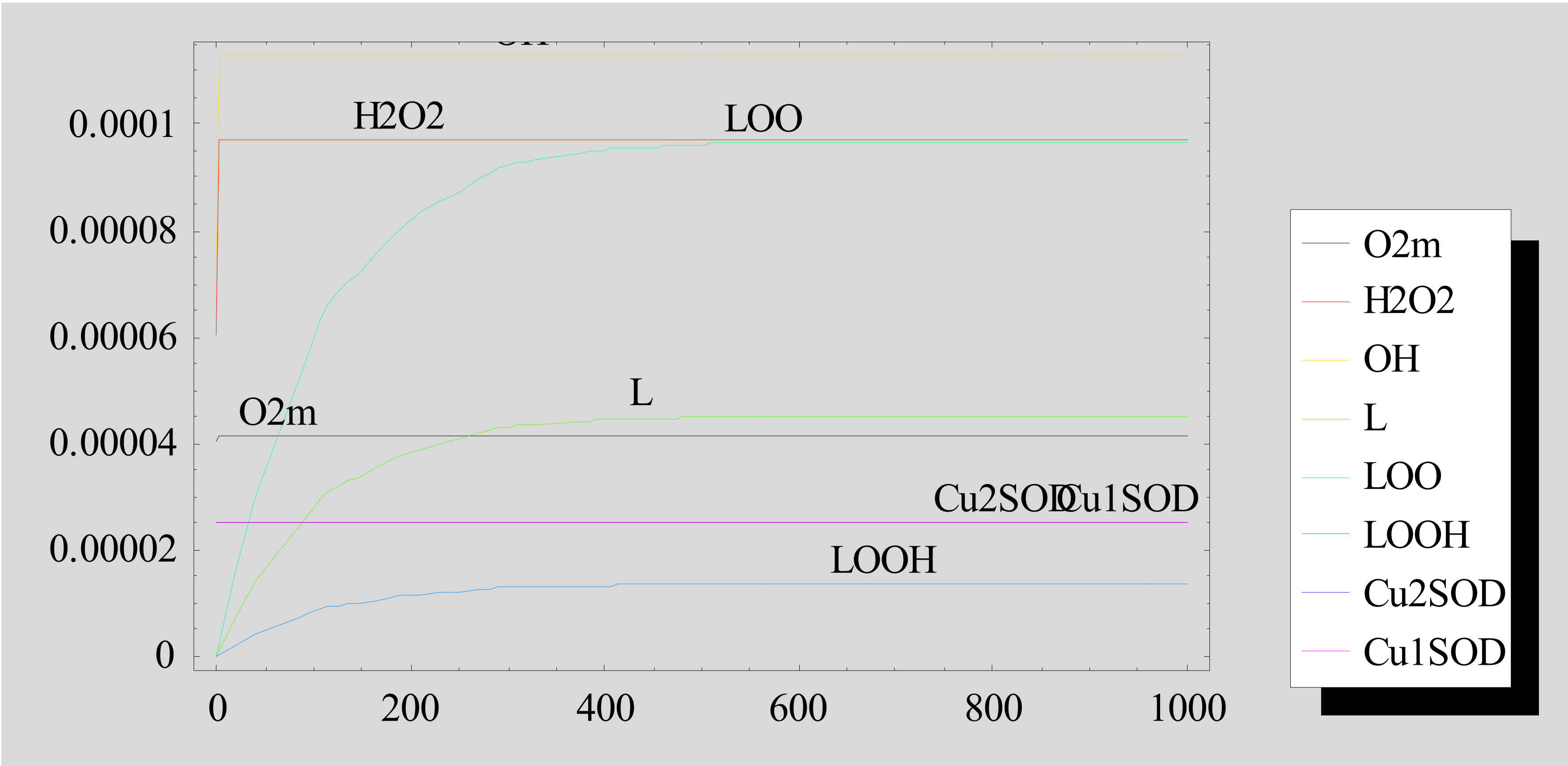
- The reaction of H_2O_2 with CuZnSOD leads to hydroxyl radical generation.
- O_2^{*-} radicals might reduce membrane damage by acting as chain breakers.
- SOD cycles between a reduced and oxidised state. At low O_2^{*-} levels the intermediates might interact with other redox partners and increase the superoxide reductase activity of SOD.

Reactions



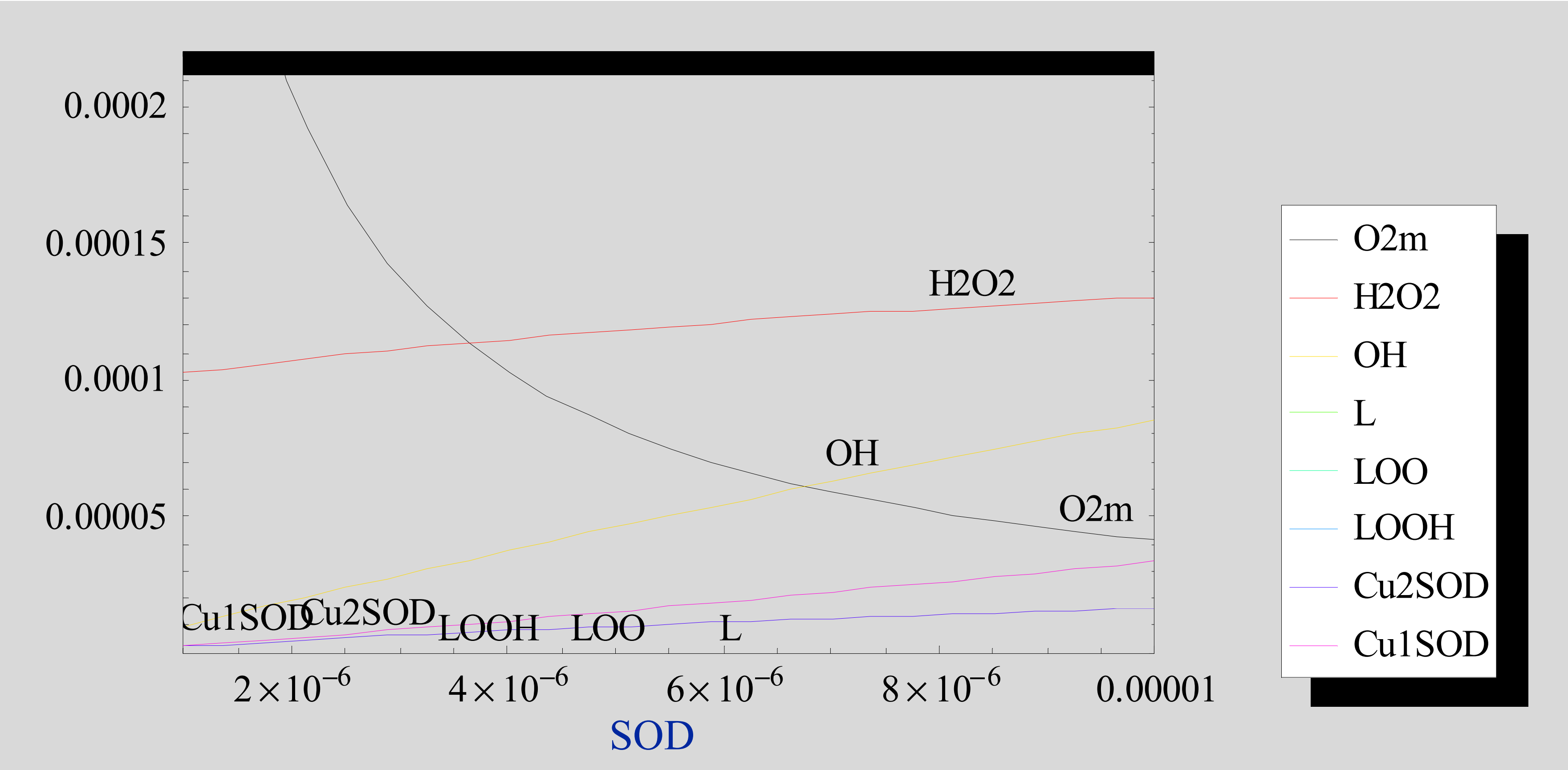
O2
Cu(I)ZnSOD
O2-*
k13b
k3
SOO
k2
SOR
CAT
k13a
k7
Cu(II)ZnSOD
H2O
k1
H2O2
O2-*
HaberWeiss Reaction
Cu(II)ZnSOD
k5
k6
HO2*
HO*
HO*
k10
k11
LH
k19
LOO*
Removed
k17
L*
LOO*
O2-*
LH
k18
k4
O2
LOH
LOOH
k12

Standard Simulation



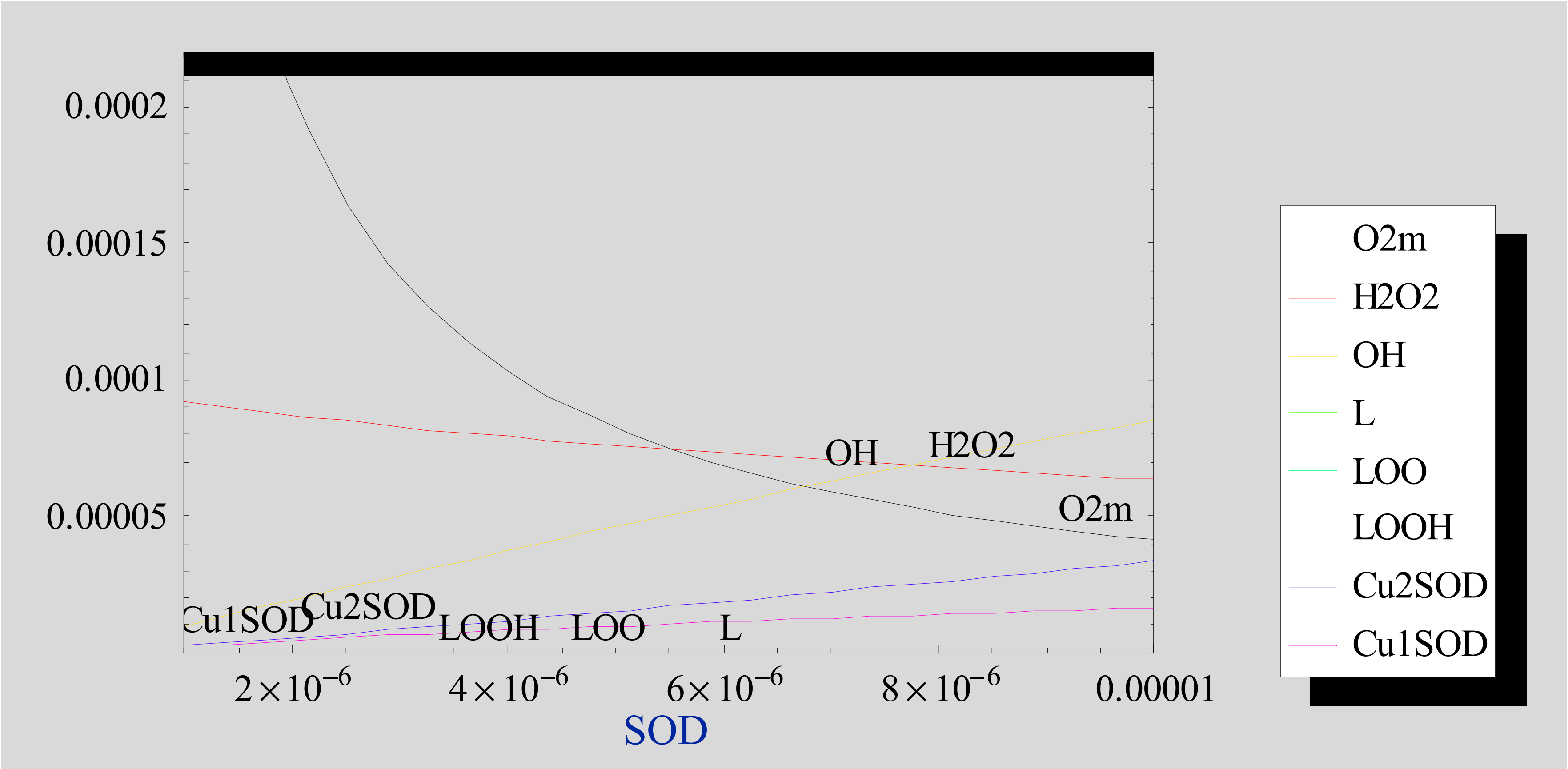
{k1 → $6.6*10^{-7}$, k2 → $1.6*10^{9}$, k3 → $1.6*10^{9}$, k4 → 10^{5}, k5 → $2*10^{4}$,
k6 → 1, k7 → $3.4*10^{7}$, k9 → 10^{6}, k10 → 2000, k11 → $5*10^{8}$, k12 → $10^{7}*10^{-5}$,
k13a → $8700*10^{-6}$, k13b → $8700*10^{-6}$, k17 → $3*10^{8}$, k18 → 14, k19 → $8.8*10^{4}$,
SOD → 10^{-5}, cat → 10^{-5}}

Increasing SOR Activity



k3b = 10*k3a

Increasing SOO Activity

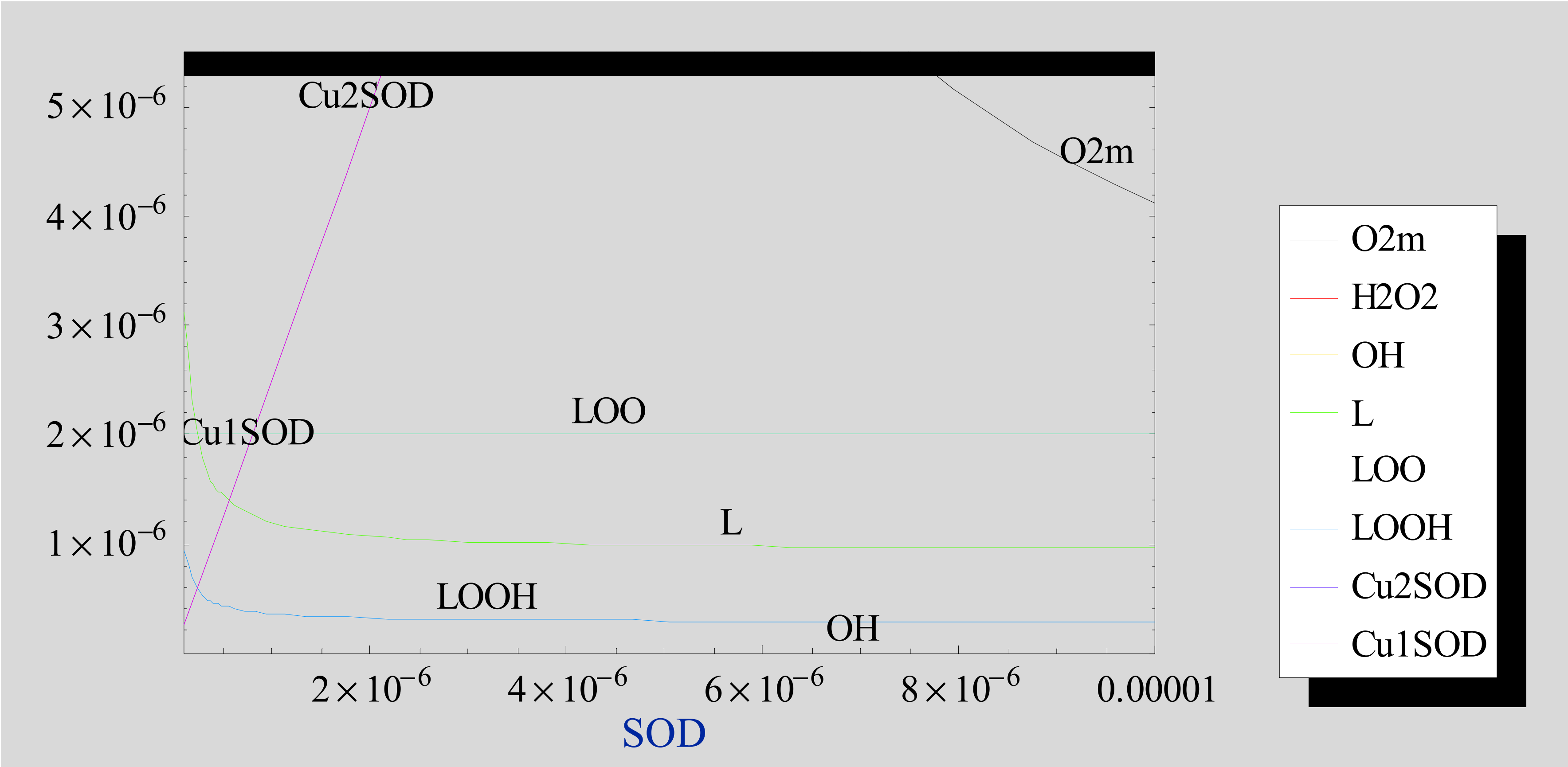


10*k3b = k3a

SOO/SOR Results

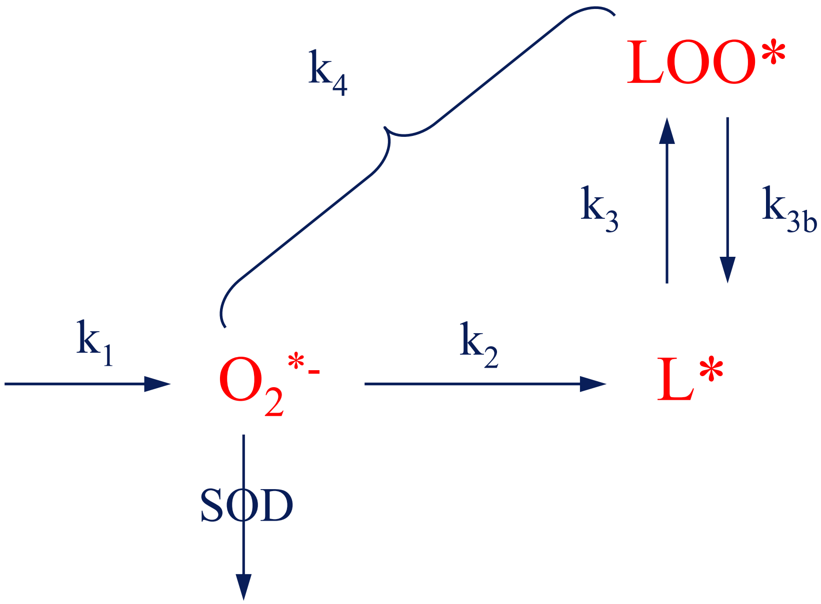
- In principle the SOO/SOR idea works.
- However, it is not clear why in experiments SOR activity is always greater than SOO activity.
- Alternative redox partners have only been demonstrated for CuZnSOD, not MnSOD.

Membrane Damage Termination



- LOO* remains constant throughout the entire range
- L* and LOOH increase for small SOD values, but are constant otherwise.
- Why are the concentrations of L* and LOO* not increasing with increasing SOD ?

Simplified Modell



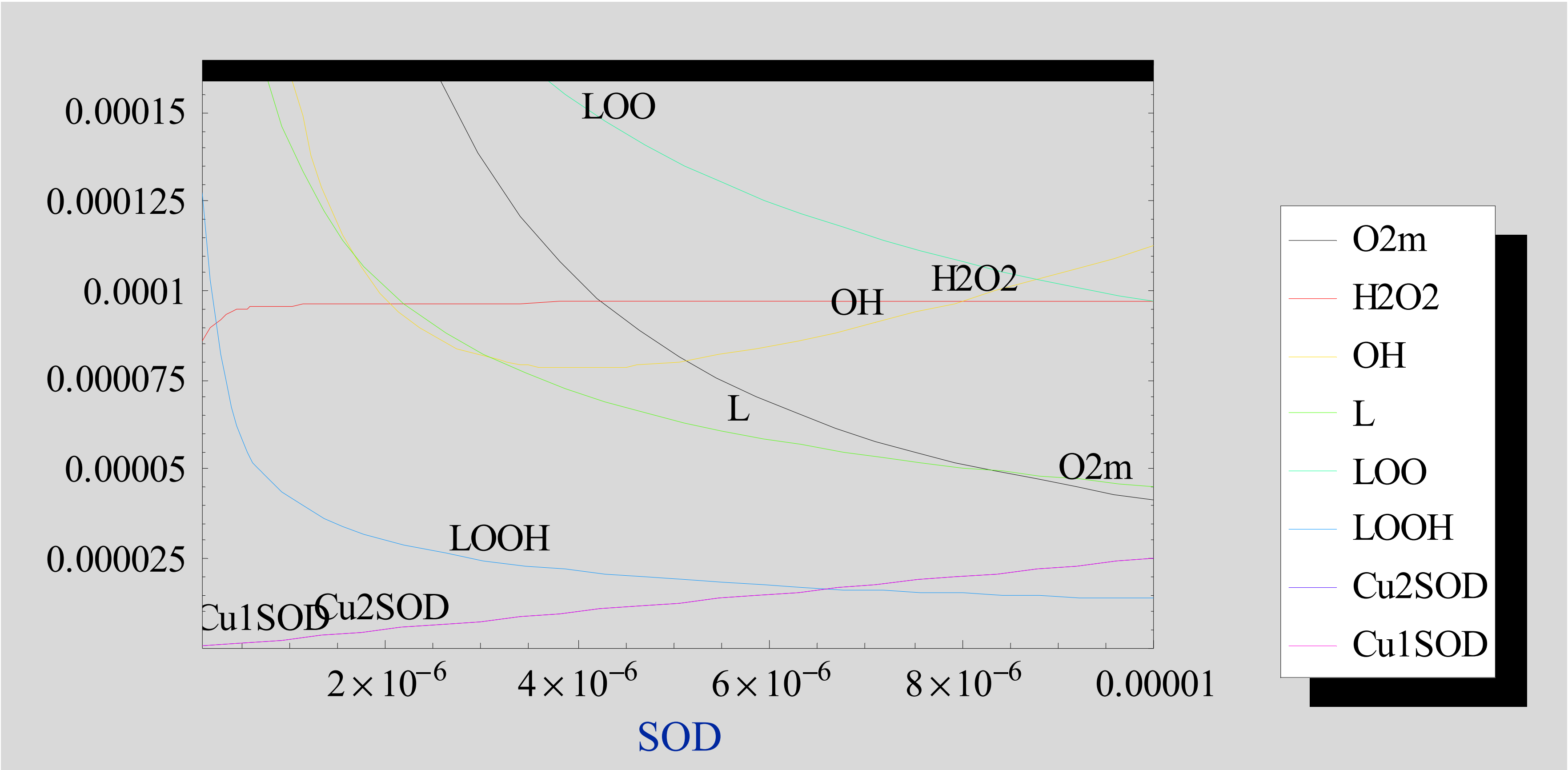
$$O^{\bullet -}_{2,ss} = \frac{k_1}{2k_2 + SOD}$$

$$LOO^{\bullet}_{ss} = \frac{k_2}{k_4}$$

$$L^{\bullet}_{ss} = \frac{k_2 \cdot k_{3b}}{k_3 \cdot k_4} + \frac{k_1 \cdot k_2}{k_3 \cdot (2k_2 + SOD)}$$

- LOO* doesn't change because it is completely independent of SOD and O_2^{*-}. If the concentration of O_2^{*-} changes, membrane damage initiation changes by the same factor as the termination (for a given k_4).
- Without the back reaction k_{3b}, levels of L* should decrease continously with increasing SOD, and this can be confirmed by disabling this reaction in the full modell.
- => Chain breaker idea does not work !!

Alternative Pathway Mechanism



Simulation with full modell and SOO equal to SOR (no effect)

Simplified Modell 2

$$\xrightarrow{k1} O_2^{-\bullet} \xrightarrow{k2 \cdot SOD} H_2O_2 \xrightarrow{k3 \cdot cat} H_2O$$

$$O_2^{-\bullet} \xrightarrow{k_{10}} \text{other reactions}$$

$$O^{\bullet -}_{2,ss} = \frac{k_1}{k_{10} + k_2 \cdot SOD}$$

$$H_2O_{2,ss} = \frac{k_1 \cdot k_2 \cdot SOD}{\left(k_{10} + k_2 \cdot SOD\right) \cdot k_3 \cdot cat}$$

- If there are alternative reaction pathways for the superoxide radical, increasing SOD channels more of the available O_2^{*-} into the H_2O_2 generating branch and can thus increase oxidative stress.
- This principle is independent of the SOD type (CuZnSOD, MnSOD).
- k_{10} does not only represent membrane damage, but ALL other reactions of O_2^{*-} . Therefore k_{10} can be much larger than the value used in the modell and so the resulting effect on H_2O_2 can also be much larger.

Summary

- Overexpression of the antioxidant SOD leads to increased oxidative stress.
- We created a mathematical model to test some possible explanations.
- Only one proposed idea (SOD cycling) works as suggested under certain conditions.
- Studying the model we discovered an additional, more general, mechanism that might account for the increased oxidative stress.