Hypotensive Effect by Activation of NAD(P)H:quinone oxidoreductase 1 via Modulation of eNOS Activity

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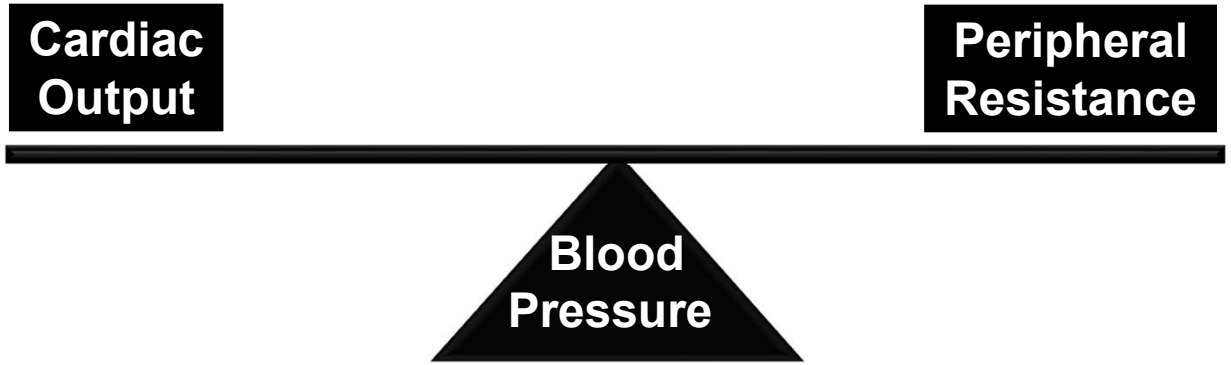
Blood Pressure

What is blood pressure?



Blood pressure regulation mechanism

- √ Baroreceptor & chemoreceptor reflex
- √ Renin-angiotensin system (RAS) & aldosterone
- √ Nitric oxide (NO) synthesized by eNOS (NOSIII)

Hypertension

- Raised blood pressure changes the structure of the arteries.

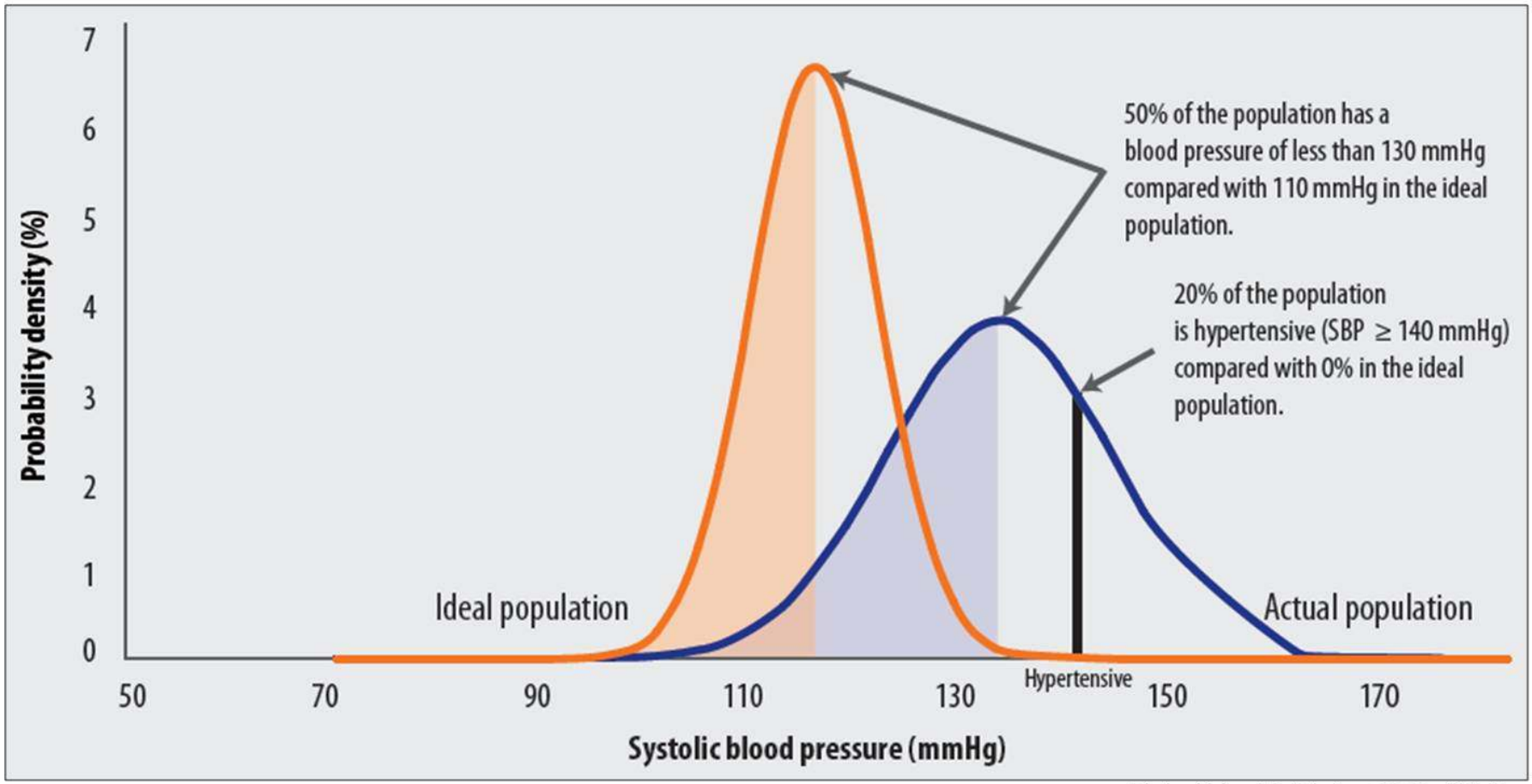
- **Hypertension (High blood pressure)**

  √ **Systolic blood pressure (SBP) >140 mmHg**

  √ **Diastolic blood pressure (DBP) > 90 mmHg**

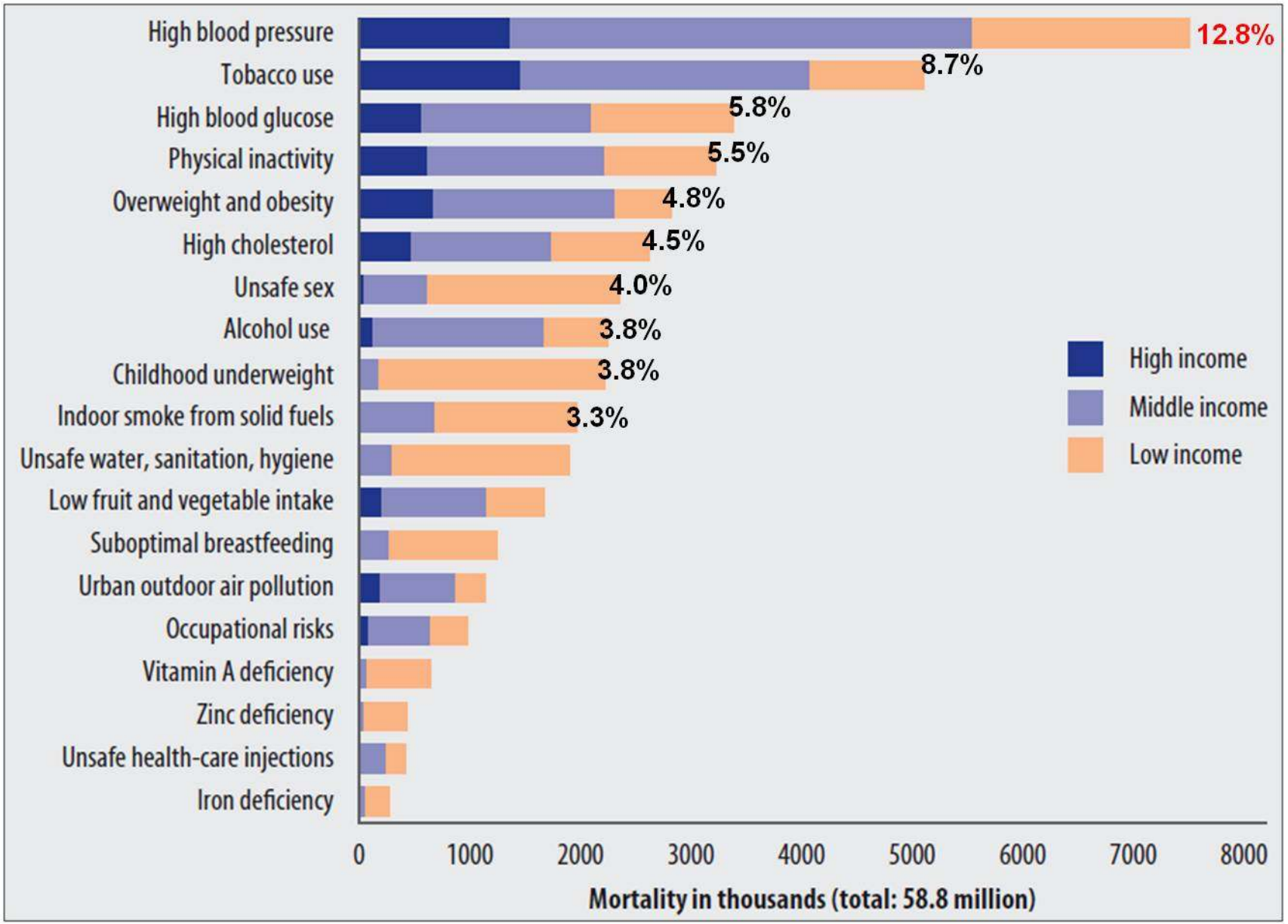
- Raised blood pressure changes the structure of the arteries.
- As a result, risks of stroke, heart disease, kidney failure and other diseases increase, not only in people with hypertension but also in those with average, or even below-average, blood pressure.
- Diet – especially too much salt – alcohol, lack of exercise and obesity all raise blood pressure.

An observed population distribution of SBP



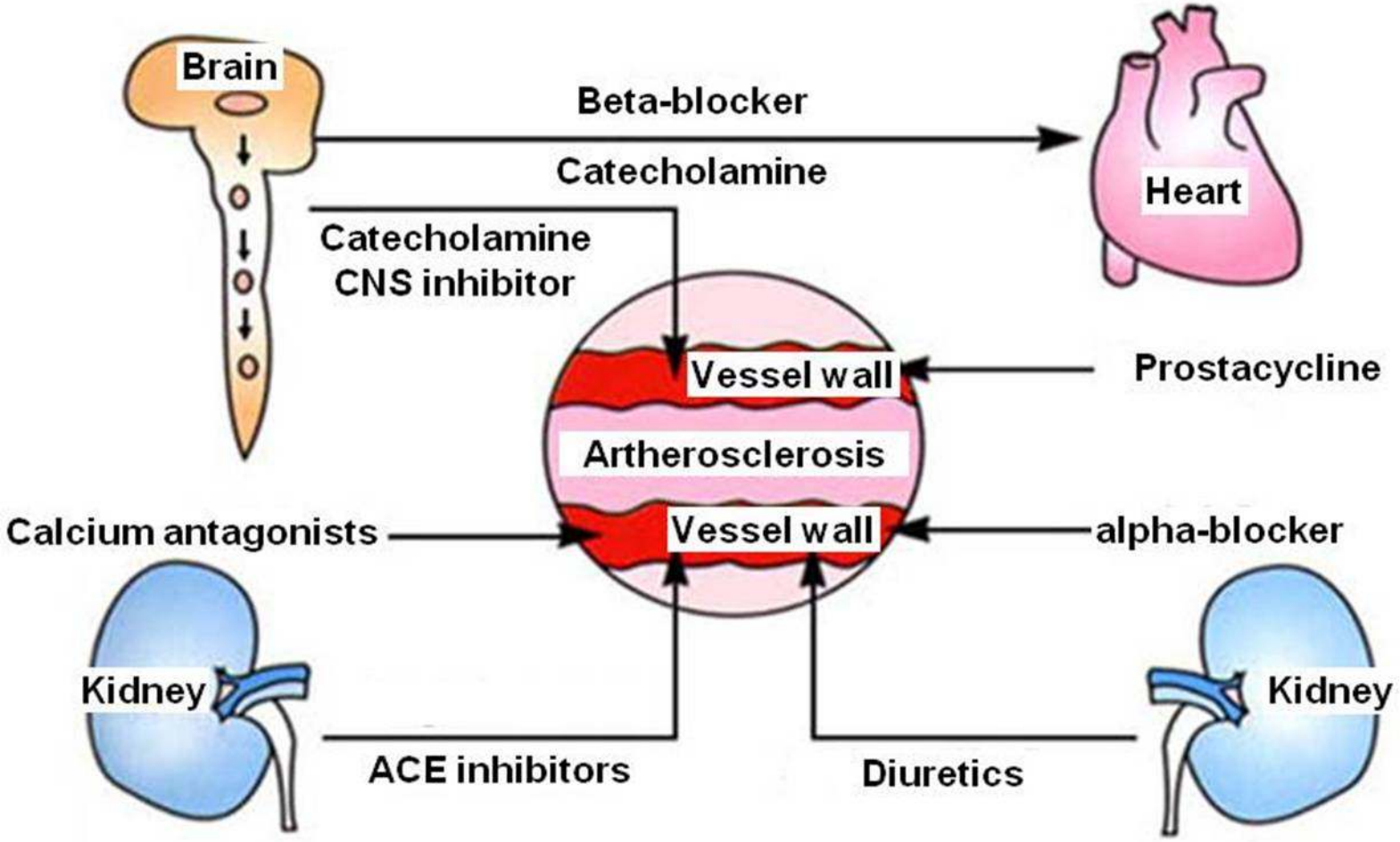
Global Health Risks, 2009, WHO.

Deaths attributed to 19 leading risk factors



Global Health Risks, 2009, WHO.

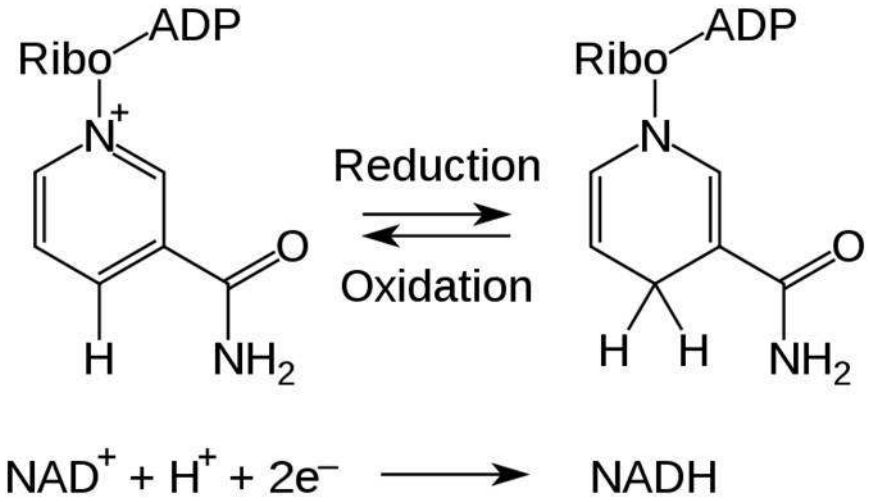
Drugs and targets for hypertension

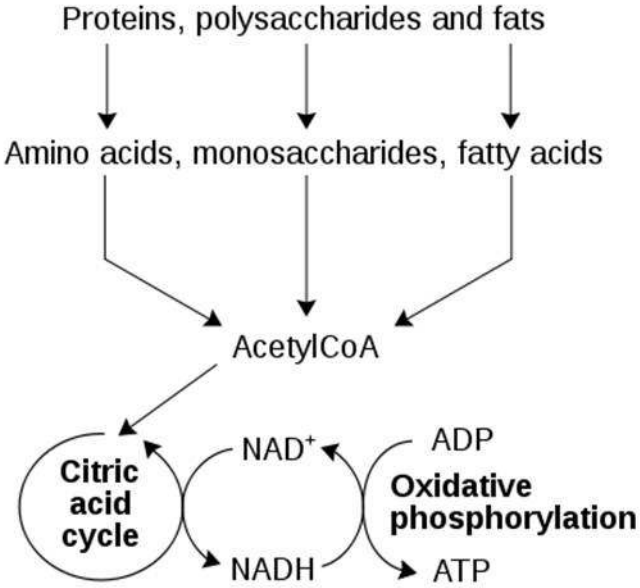


NAD^+ /NADH

Nicontinamide adenine dinucleotide

- Coenzyme involving redox reaction to release energy from nutrient in living cells through NADH oxidation by oxidoreductase (malate-aspartate shuttle).
- As a non-redox roles, NAD^+ is consumed in ADP-ribose transfer reaction
- Recently sirtuin, NAD-dependent deacetylases, is pharmacologically interesting.

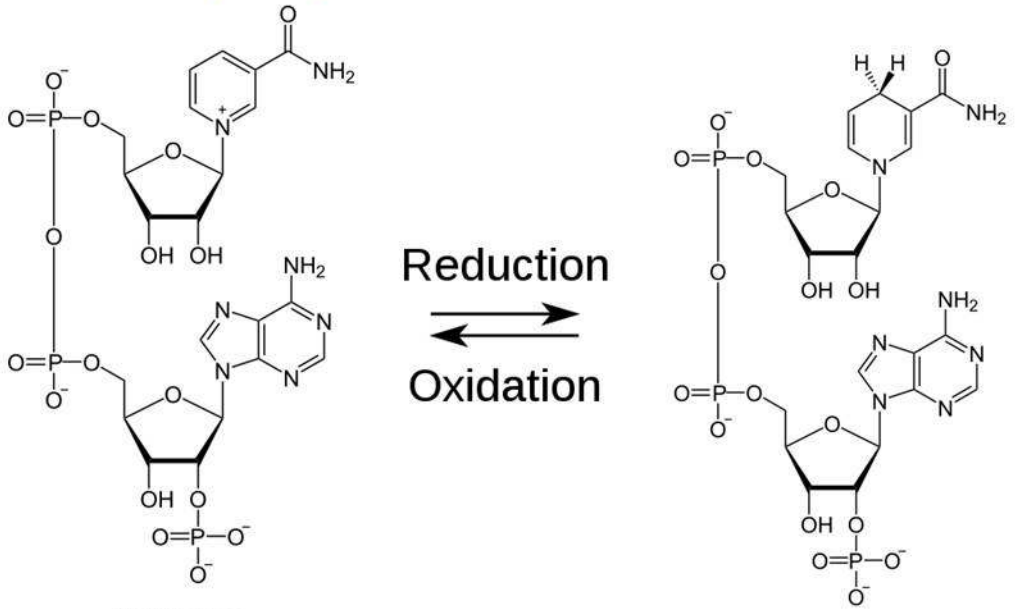




NADP+ /NADPH

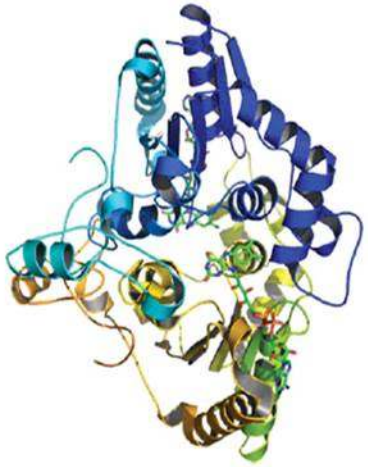
Nicontinamide adenine dinucleotide phosphate

- Coenzyme used in anabolic reactions such as lipid and nucleic acid synthesis, cholesterol synthesis, and fatty acis elongation, which required NADPH as a reducing agent. NADPH provides the reducing equivalents for biosynthetic and the oxidation-reduction involved in protecting against the toxicity of ROS, allowing the regeneration of GSH, and also reducing equivalents for cytochrome P450 hydroxylation of aromatic compounds, steroids, alcohol and drugs.
- The oxidative phase of the pentose phosphate pathway is the major source of NADPH in cells (~60% of the NADPH required)



NADP+ **NADPH**

NAD(P)H:quinone oxidoreductase (NQO)



-NQO1 is a ubiquitous homodimeric flavoenzyme initially identified in 1958.

-NQO encoded by four separate gene loci (Most well characterized members include NQO1 and NQO2)

-NQO1 catalyzes two-electron reduction of various quinones utilizing NAD(P)H as an electron donor.

-NQO1-mediated reduction of quinones to hydroquinones is an important cellular defense mechanism against oxidative stress.

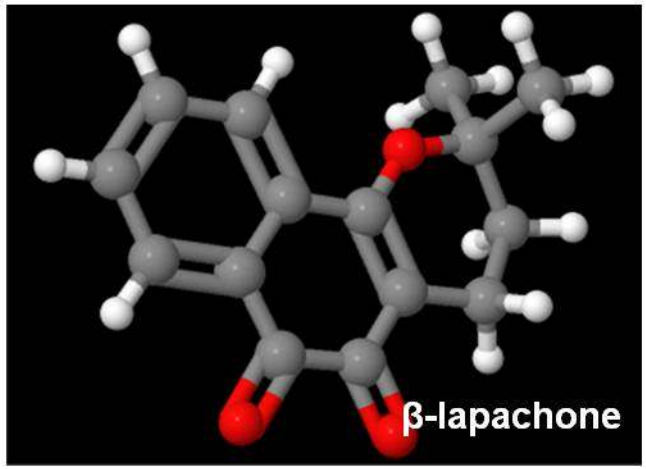
-Studies of NQO1 structure and function have shown that NQO1 is a homodimer that functions via a “ping-pong” mechanism.

β –lapachone (β L)

-β-lapachone (βL; 3,4-dihydro-2,2-dimethyl-2H-naphtho[1,2-b]pyran-5,6-dione) is isolated from the bark of the Lapacho tree.

-βL extract has been used as a folk medicine for centuries and initial studies have demonstrated its ability to inhibit tumor growth.

-βL is a well-known NQO1 substrate, and NQO1 catalyzes the reductive activation of βL which is quinolic chemo-therapeutic compound.

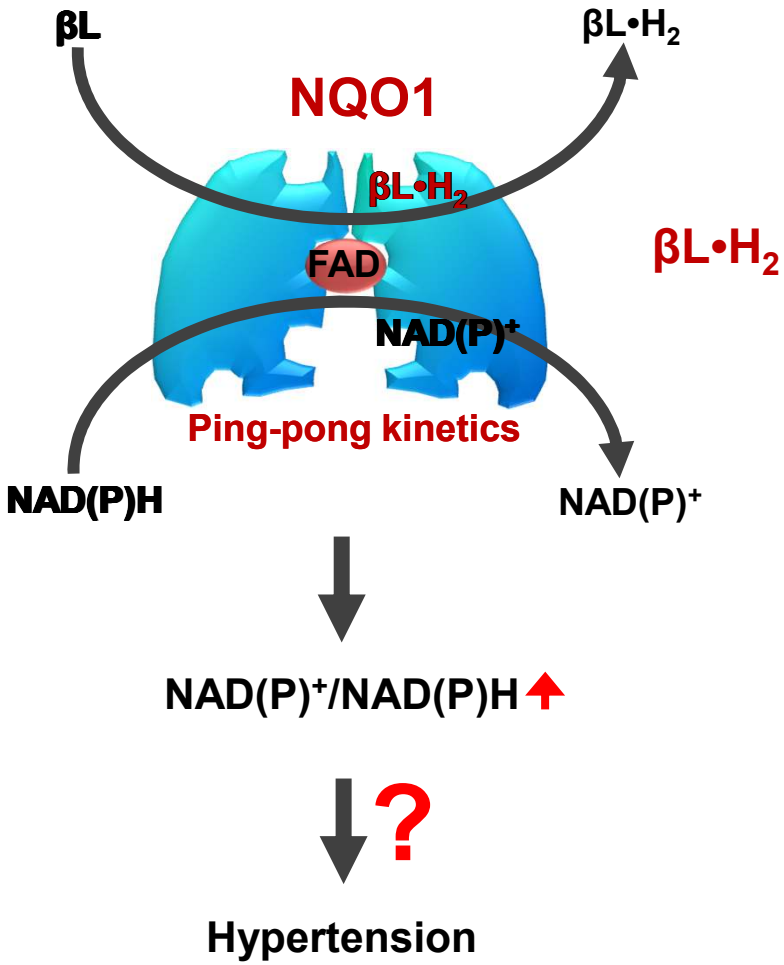


Previously,

- It was reported that cellular $NAD(P)^+/NAD(P)H$ ratio was decreased in several disease conditions, such as diabetes-induced hyperglycemia and skeletal muscle dystrophy.

- Calorie restriction and exercise were reported to lead to an increase in $NAD^+/NADH$ ratio, and have beneficial effects on BP in hypertensive patients during clinical trials.

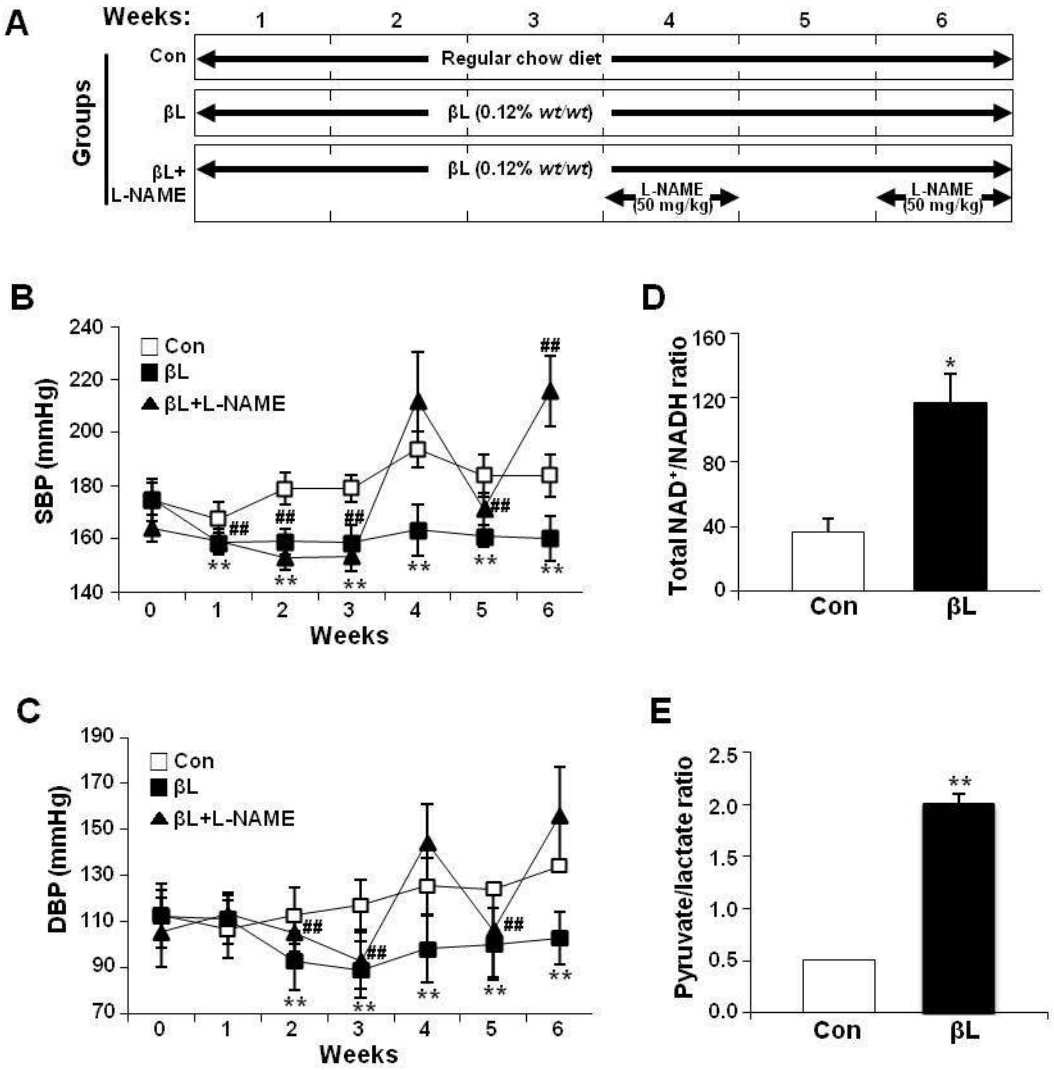
Objective


βL
βL•H2
NQO1
βL•H2
FAD
βL•H2
NAD(P)+
Ping-pong kinetics
NAD(P)H
NAD(P)+
NAD(P)+/NAD(P)H
?
Hypertension

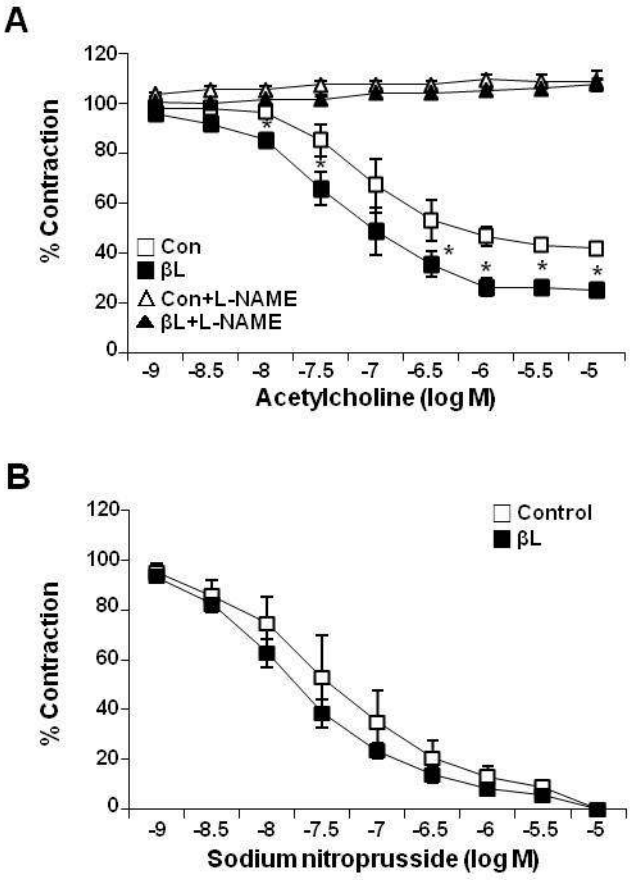
SHR : Spontaneous Hypertensionsive Rat

- In 1963, Dr. Okamoto, Kyoto School of Medicine, from outbred Wistar Kyoto male with marked elevation of blood pressure mated to female with slightly elevated blood pressure. Then, through the brother x sister mating with continued selection for spontaneous hypertension, SHR were established.
- As a animal model for genetic hypertension, SHR are being widely used for the efficacy testing of hypertensive drug development.

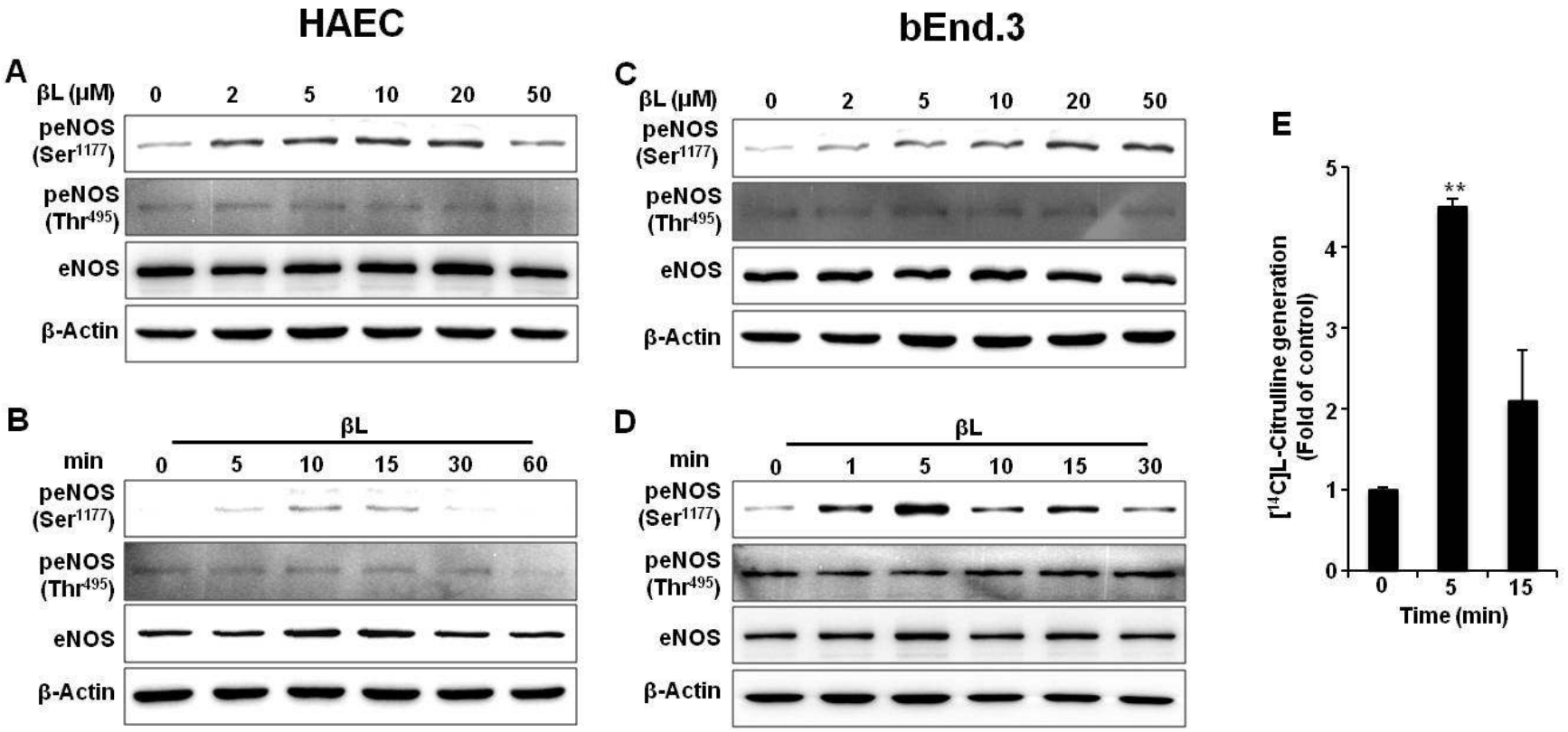
βL reduces blood pressure in SHR and induces NADH oxidation in endothelial cells



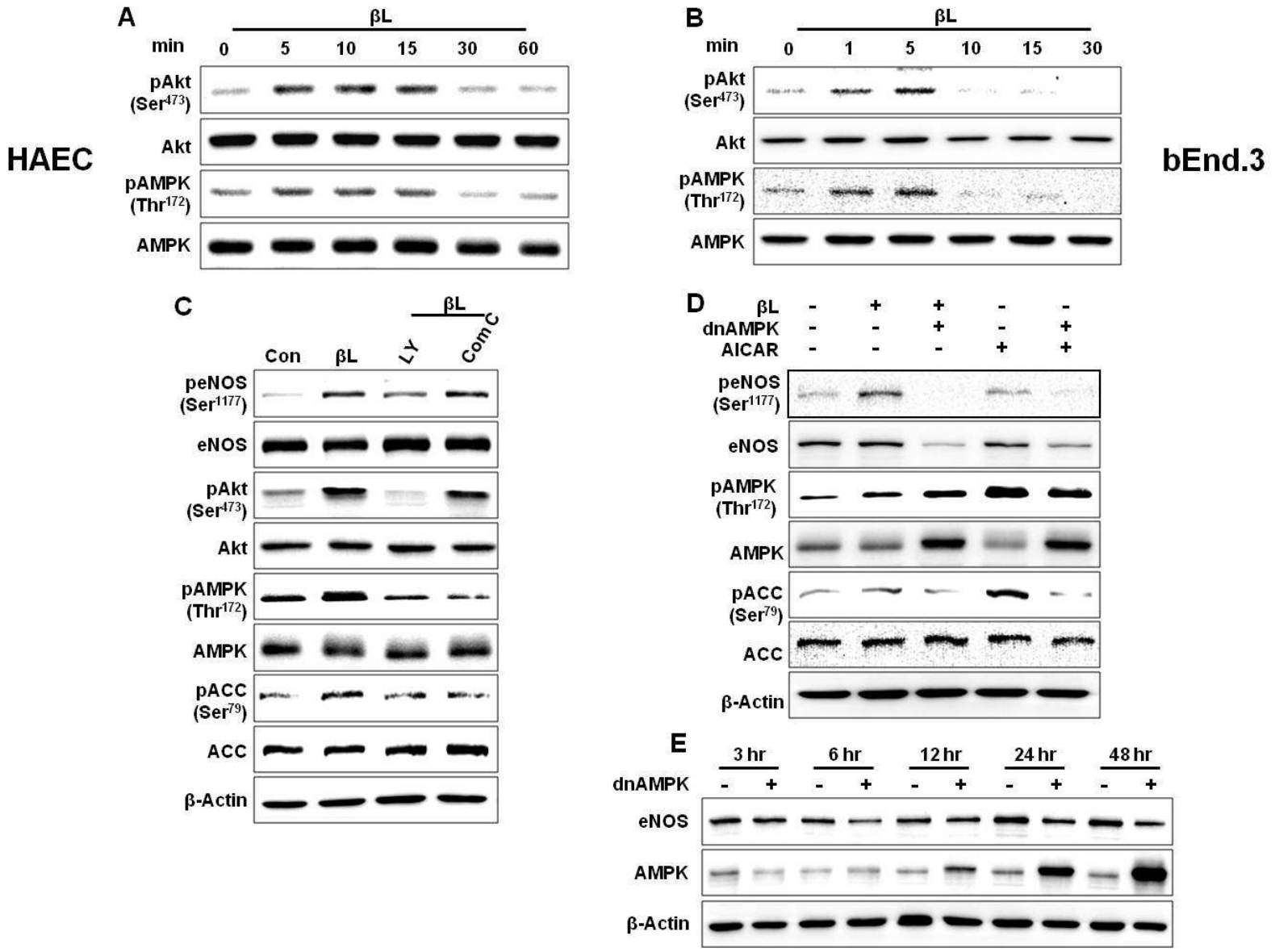
βL regulates endothelium–dependent vasodilatation through eNOS



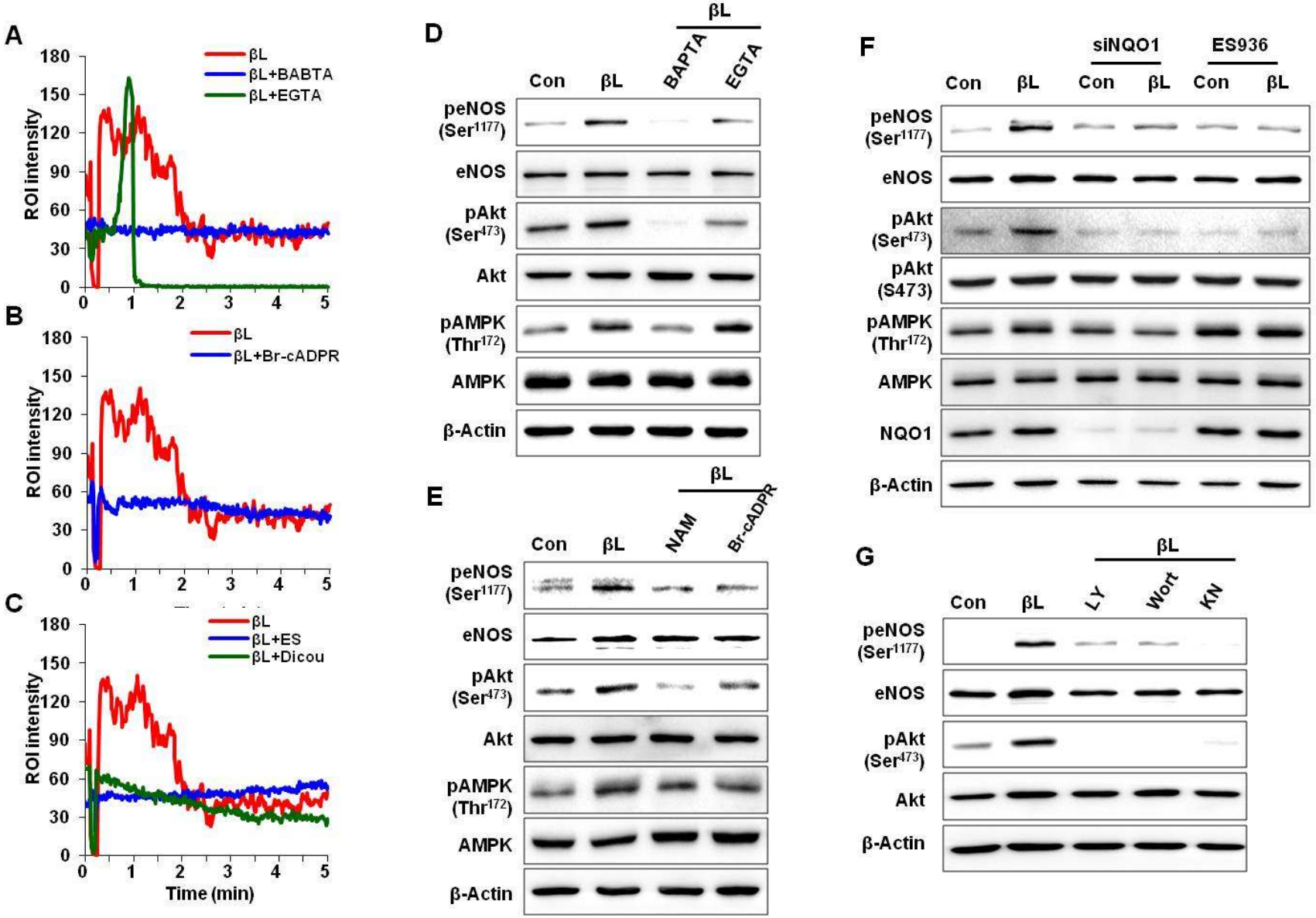
βL increases eNOS activity via ser^{1177} phosphorylation



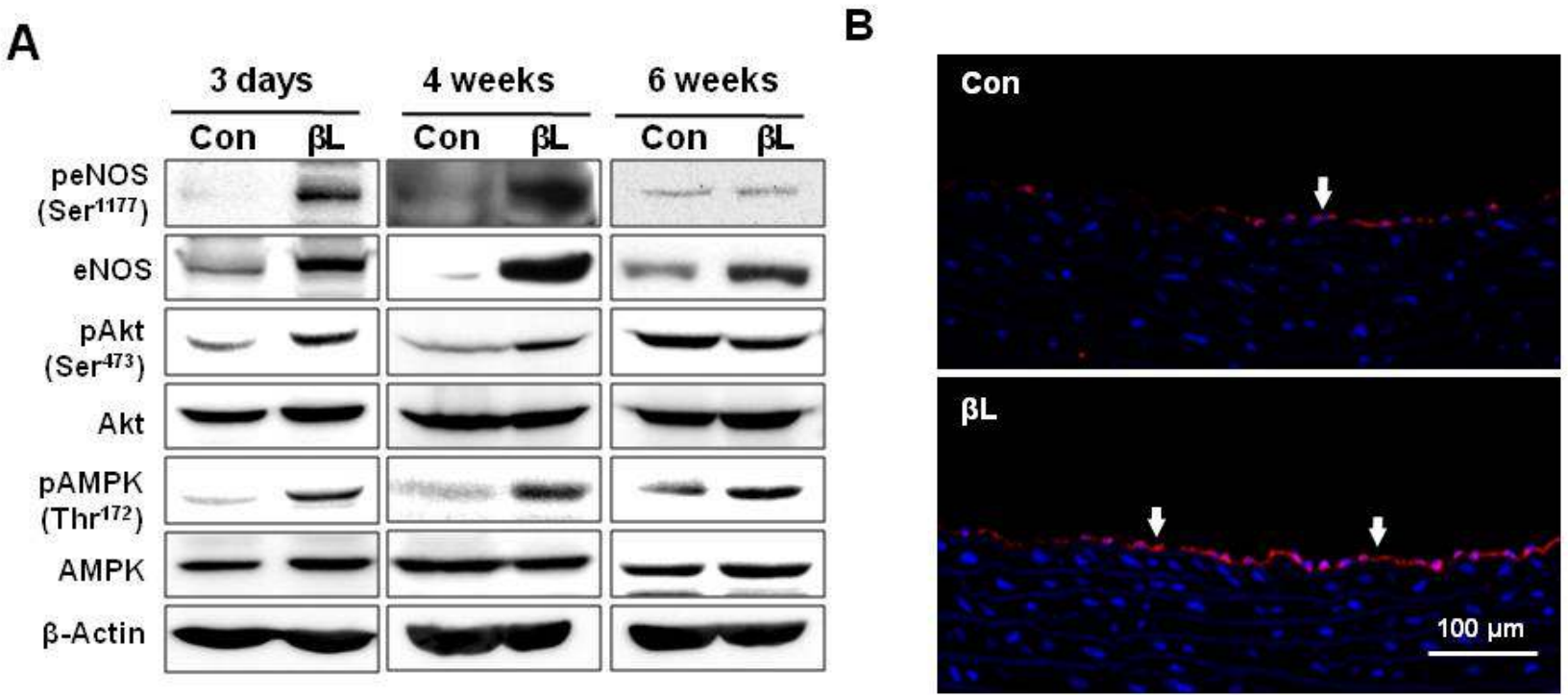
Phosphorylation of Akt and AMPK mediate eNOS activation by βL



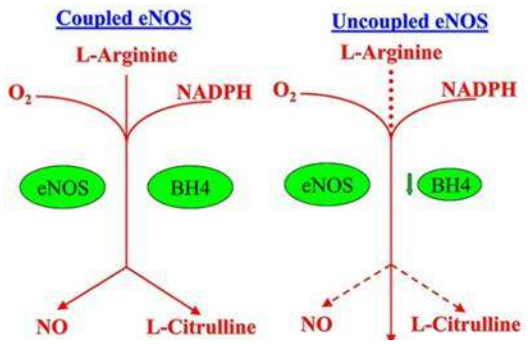
βL–induced increase of $[Ca^{2+}]_i$ activate Akt/AMPK/eNOS via NQO1–dependent mechanism

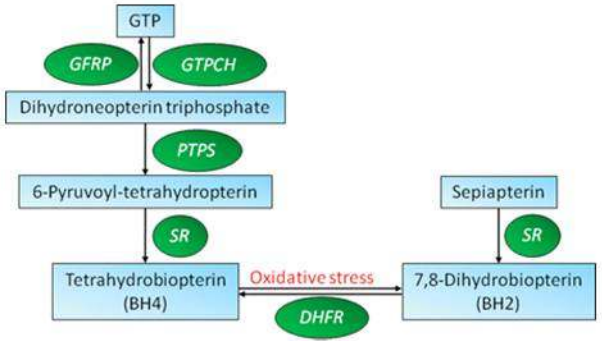


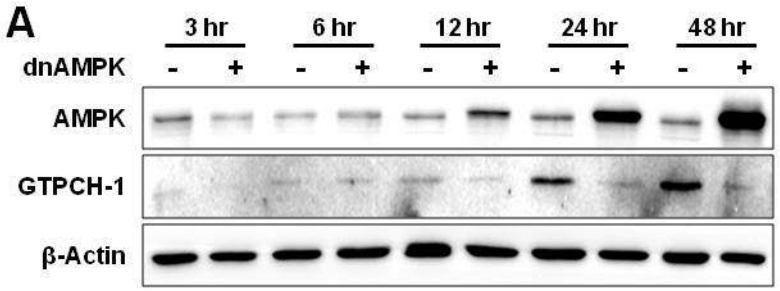
β L activates eNOS in the aorta of SHR

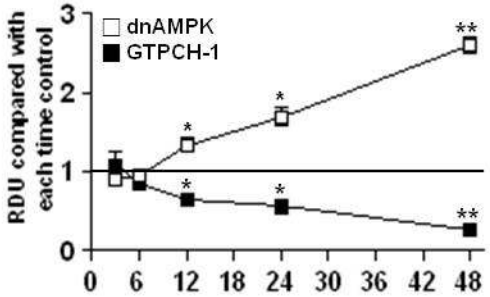


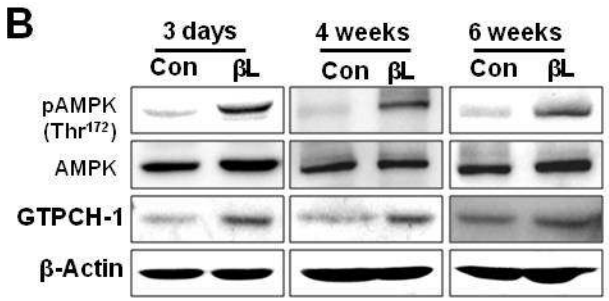
GTPCH−1 level is dependent on AMPK activity and βL inflates GTPCH−1 level in the aorta of SHR by AMPK activation

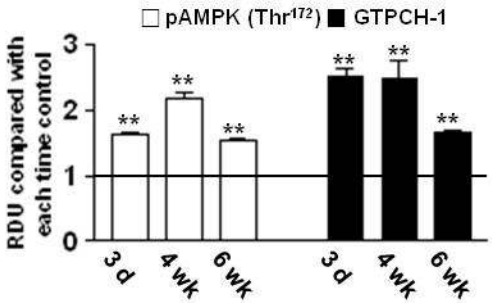




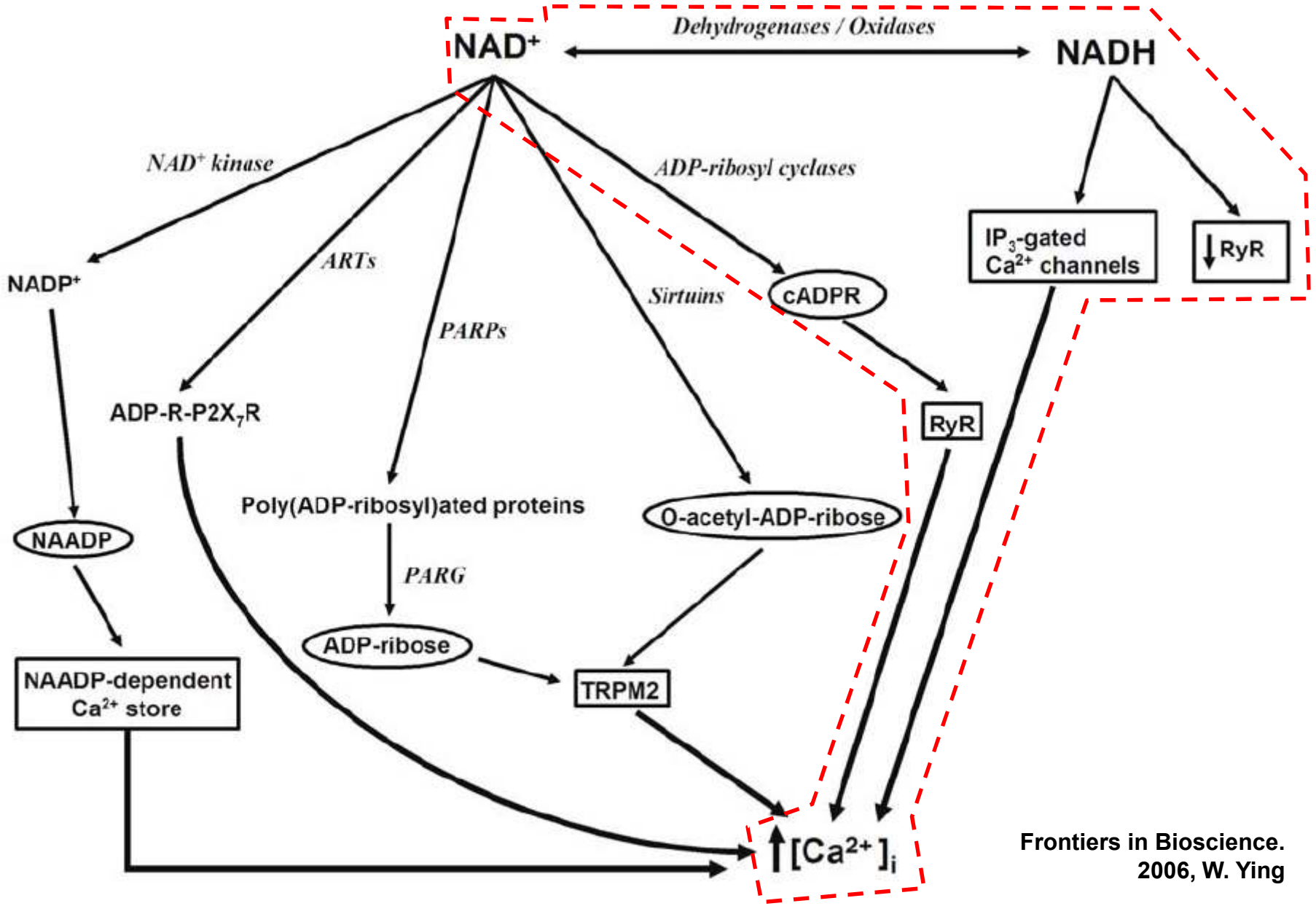








NAD^+ /NADH & Calcium



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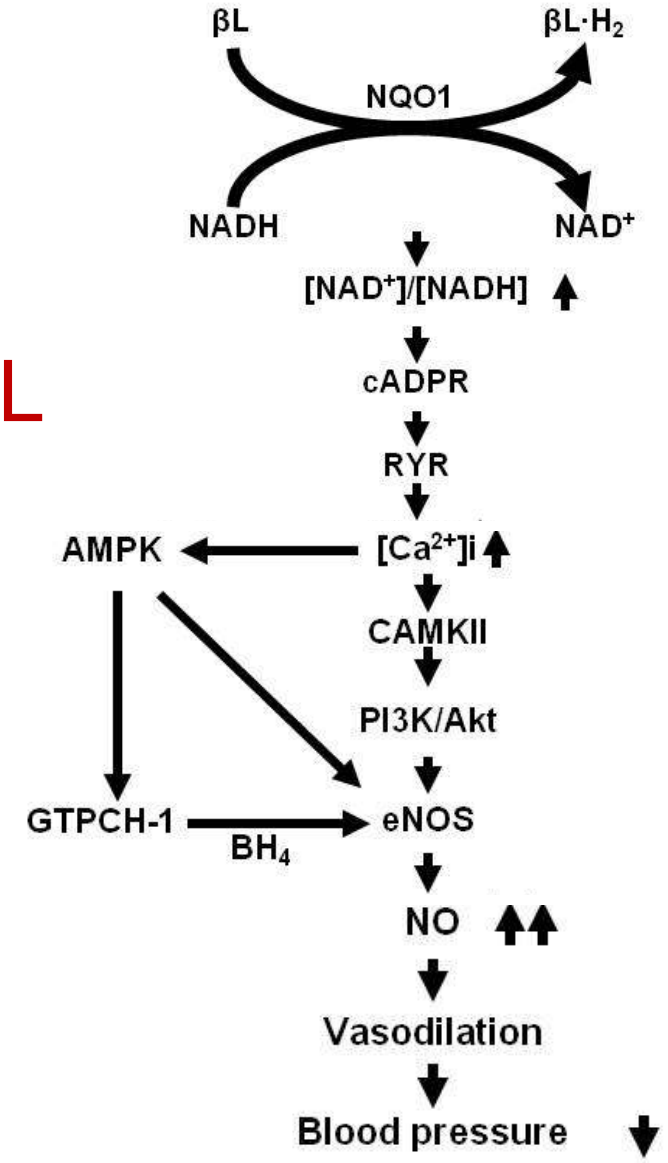
2006, W. Ying

Model for the blood pressure regulation mechanism of βL



Conclusion

- This study is the first to demonstrate that NQO1 activation has hypotensive effect mediated by eNOS activation via modulation of cellular $NAD(P)^+/NAD(P)H$ ratio in rat hypertension models.

- Even though it is unclear why $NAD(P)^+/NAD(P)H$ is decreased in several pathologic conditions, regulation of $NAD(P)^+/NAD(P)H$ ratio by βL via NQO1 activation could be beneficial for improvement of hypertension.

Acknowledgements

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Thank you!