THE OXIDATION OF APOCYNIN CATALYZED BY MYELOPEROXIDASE: PROPOSAL FOR NADPH OXIDASE INHIBITION

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Apocynin, a methoxy-cathecol originally extracted from the root of *Picrorhiza kurroa*, has been used as an efficient inhibitor of the NADPH oxidase complex and its mechanism of inhibition is linked to prior activation through the action of peroxidases. Here we studied the oxidation of apocynin catalyzed by myeloperoxidase (MPO) and activated neutrophils. Apocynin (1mM) was incubated with 0.1μM MPO and hydrogen peroxide or zymosan opsonized activated human neutrophils. The oxidation was studied by UV-Vis and the oxidation products. Identified by LC-MS. We found the apocynin and its oxidation products did not conjugate with glutathione, however apocynin radical was able to oxidize sulfhydryl compounds, as demonstrated by adding glutathione (GSH) during the reaction course. The production of hypochlorous acid by stimulated neutrophils was strongly inhibited by apocynin (IC_{50} of 19μM), but this effect was very limited with purified MPO. In conclusion, we demonstrated for the first time that apocynin is easily oxidized by the catalytic action of MPO, resulting in its dimer and trimer derivatives, the former is also produced by activated neutrophils what reinforces its important role in the mechanism of NADPH oxidase in neutrophils. The reactivity of the apocynin radical with sulfhydryl compounds might be related with its mechanism of inhibition of the NADPH oxidase.

Keywords: apocynin, myeloperoxidase, NADPH oxidase, neutrophil