"THIOREDOXIN-DEPENDENT HYDROPEROX IDE PEROXIDASE ACTIV ITY OF BACTERIOFERRITIN CO-MIGRATORY PROTEIN FROM XYLELLA FASTIDIOSA"

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Xylella fastidiosa, a xylem-limited bacterium that causes a range of economically important plant diseases, possesses many antioxidant proteins, including bacterioferritin co-migratory protein or BCP (also referred to Peroxiredoxin Q in their plant homologues). Intending to characterize BCP protein biochemically and structurally, its gene was cloned into an Escherichia coli expression vector (pET15b, Novagen) and the corresponding protein was purified by nickel affinity chromatography. BCP presented thiol-peroxidase activity which depended on the reducing power of a thioredoxin. Only at high concentration (2 mM) DTT supported peroxidase activity that was monitored by ferrous oxidation in xylenol orange assay (FOX). Peroxidase activity was increased 30 fold when thioredoxin (TSNC from X.fastidiosa) was added, suggesting that TSNC might be the BCP biological electron donor. Experiments using NADPH reduction of thioredoxin catalyzed by thioredoxin reductase (Trx and TrxR from X.fastidiosa or E.coli) provided a method to follow continuously this peroxidase activity. Through this assay, we observed that BCP can reduce several hydroperoxides. Kinetic parameters showed that BCP can reduce cumene hydroperoxide and hydrogen peroxide with a better efficiency than tert-butyl hydroperoxide. Non-reducing SDS-PAGE and mutagenesis studies showed that the oxidized form of purified wild type BCP behaves as a monomeric enzyme and suggested no intermolecular disulfide bond being formed during its catalytic cycle. Therefore, BCP probably does not possess 2-Cys typical mechanism. Supported by FAPESP and CNPq.