CLONING AND EXPRESSION OF PROTEINS FROM *XYLELLA FASTIDIOSA* POSSIBLY INVOLVED IN REDUCTING PATHWAYS OF OHR

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Ohr ("Organic Hydroperoxide Resistance") is a protein exclusively present in bacteria. We have cloned and expressed several proteins possibly involved in the in vivo reduction of Ohr, such as two thioredoxins (TRXA and TSNC), two thioredoxins reductases (TRR and YBBN) and proteins belonging to the pyruvate dehydrogenase complex: Dihydrolipoamide dehydrogenase (Lpd) and Dihydrolipoamide succynil transferase (SucB). These genes were cloned in pET vectors that include features such as an N-terminal his tag and T7 promoter. To express most of these proteins in the soluble fraction, their genes were transformed into bacterial hosts that have mutation in thioredoxin reductase (*trxB*) and glutathione reductase (*gor*) genes, which enhances disulfide bond formation. Preliminary results indicated that Ohr in the presence of TSNC has a slightly improvement in its peroxidase activity. TRR and Lpd activities were measured by a DTNB reductase assay in the presence of TSNC and lipoamide, respectively. Also, it was seen that Ohr was capable to reduce organic hydroperoxides in the presence of lipoamide and lipoamide dehydrogenase from bovine serum. We intend to test if Ohr is capable to reduce hydroperoxides in the presence of lipoamide system from *X. fastidiosa* that includes SucB, which has lipoamide covalently attached. Enzymatic assays are underway in an attempt to verify the relative efficiency of each system.

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