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A New Peroxidase POX_{1b}, biochemical characterization, suitable biosensor for hydrogen peroxide detection in biological samples

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Peroxidases-based biosensors and specially amperometric ones offer an interesting alternative for H_2O_2 detection. In fact, peroxidases (EC: 1.11.1.7) are enzymes that catalyses the oxidation of a variety of organic and inorganic compounds by hydrogen peroxide. Determination of hydrogen peroxide is of practical importance in chemical, clinical, industrial and many other fields. Peroxidases have been used in chemical, biological and clinical as well as many other fields but novel applications in biosensors design are arousing more and more interest. Horseradish peroxidase (HRP) is the most widely used enzyme for biosensors design but new sources of peroxidases are now of great interest. In a previous work, two peroxidase from garlic bulb (*Allium sativum*), designated POX_1 and POX_2 (Marzouki Mdellji S. *et al* 2005) were revealed by native PAGE.

In the present work, we describe the purification and some biochemical properties of a new peroxidase called POX_{1B} and use it as a biosensor to detect peroxides. POX_{1B} seems to be attractive for biosensor design since its apparent Km for H_2O_2 is much lower when immobilized (0.13 mM) than when free (0.56 mM). In addition of its storage and operational stability, POX_{1B} was found to be highly stable vs. temperature since almost 70 % of its activity is conserved at 60°C during 40 min and fool activity is retained after 40 min of incubation at 50°C and 40°C. The optimum pH was around 5 and the optimum temperature was 30°C.

Thus, gelatin was used as a matrix for enzyme immobilization on the gold electrode surface. In order to study the electro-catalytic behavior of the POX_{1B} enzyme electrode towards H_2O_2, cyclic voltammograms (CVs) were used. Amperometric measurements were carried in a three electrode cell. POX_{1B}-based electrode show great potential to be applied in hydrogen peroxide monitoring in biological samples.

Reference:
Marzouki Mdellji Saida, Limam, Farid, Smaali M. Issam, Ulber Roland and Marzouki M. Nejib,
A new thermostable peroxidase of garlic *Allium sativum*: purification, biochemical characterization, immobilization and use in H2O2 detection in milk, *Appl. Biochem. Biotechnol.*, 127/3 (2005) 201-214.