Horseradish peroxidase embedded in polyacrylamide nanoparticles enables optical detection of reactive oxygen species - DTU Orbit (02/12/2018)

Horseradish peroxidase embedded in polyacrylamide nanoparticles enables optical detection of reactive oxygen species

We have synthesized and characterized new nanometer-sized polyacrylamide particles containing horseradish peroxidase and fluorescent dyes. Proteins and dyes are encapsulated by radical polymerization in inverse microemulsion. The activity of the encapsulated enzyme has been examined and it maintains its ability to catalyze the oxidation of guaiacol with hydrogen peroxide as the electron acceptor, although at a slightly lower rate compared to that of the free enzyme in solution. The embedded enzyme is also capable of catalyzing the peroxidase-oxidase reaction. However, the rate is decreased by a factor of 2-3 compared to that of the free enzyme. The reduced rate is probably due to limitation of diffusion of substrates and products into and out of the particles. The catalytic activity of horseradish peroxidase in the polyacrylamide matrix demonstrates that the particles have pores which are large enough for substrates to enter and products to leave the polymer matrix containing the enzyme. The polymer matrix protects the embedded enzyme from proteolytic digestion, which is demonstrated by treating the particles with a mixture of the two proteases trypsin and proteinase K. The particles allow for quantification of hydrogen peroxide and other reactive oxygen species in microenvironments, and we propose that the particles may find use as nanosensors for use in, e.g., living cells. (C) 2007 Elsevier Inc. All rights reserved.

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