A whole cell bio-optode based on immobilized nitrite-degrading microorganism on the acrylic microspheres for visual quantitation of nitrite ion

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Article history:
Received 8 June 2017
Received in revised form 20 August 2017
Accepted 15 September 2017
Available online 18 September 2017

Keywords:
Nitrite
Raoutella planticola
Edible bird’s nest
Optode
Reflectance

A microspheres-based microbial optosensor for NO$_2^-$ ion quantitation was constructed by using immobilized Raoutella planticola (R. planticola), the bacterium expressing NAD(P)H nitrite reductase (NiR) enzyme, which was isolated from local edible bird’s nest (EBN) via microbial technique. The whole cells and the lipophilic Nile Blue chromoionophore (NBC) were physically adsorbed on the self-adhesive photocurable poly(n-butyl acrylate-co-N-acryloxysuccinimide) [poly(nBA-NAS)] microspheres, whilst the reduced co-enzyme NAD(P)H was covalently immobilized on the succinimide-functionalized acrylic microspheres via peptide link to produce a reagentless nitrite biosensing system. As the microbial bio-optode responded to nitrite through colour change from blue to pink, a facile reflectometric approach was adopted to measure reflectance intensity at 639 nm, before and after reaction with nitrite at optimum pH 8. The optosensor could quantify NO$_2^-$ ion concentration within a dynamic linear response range of 0.5–400 mg L$^{-1}$ with a limit of detection (LOD) of 0.2 mg L$^{-1}$. The large surface area to volume ratio of the acrylic microspheres allowed solid-state diffusional mass transfer of the substrate to occur at microbio-optode surface, and an equilibrium response was achieved within 5 min. The practical feasibility of using the bio-optode for nitrite assay in food matrix sample showed good agreement with standard ion chromatography method.

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