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Biosensor for selective detection of E. coli in spinach using the strong affinity of derivatized mannose with fimbrial lectin.

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Abstract

Escherichia coli (E. coli) contamination in foods and water resources represents a major threat for human health and the environment. This work exploits the strong affinity of mannose-containing oligosaccharides with the fimbrial lectin of E. coli to design novel biosensors. Modified carbohydrate ligands were synthesized by introducing phenyl residues and aliphatic chains to mannose via reductive amination in order to increase both the affinity and selectivity to E. coli compared to other pathogenic bacteria. The synthesized ligands include p-thiolphenyl aminomannose (PTAM), p-carboxyphenyl aminomannose (PCAM), 1-deoxy-1-aminomannopyranoside (DAMP), glucosamine and low molecular weight chitosan bonded to mercapto undecanoic acid. The structures of the ligands were confirmed using (1)H NMR and 1H, (13)C, COZY NMR, and ESI/MS. The ligands were immobilized onto gold electrodes and SPR surfaces using-mercaptoundecanoic acid with glycine as deactivating agent. Two detection mechanisms were tested: (i) metal-enhanced electrochemical detection (MED) and (ii) label-free surface plasmon resonance (SPR) detection. The introduction of phenyl residues and aliphatic side groups to the mannose-containing oligosaccharides produced extremely high affinity for E. coli with detection limit of 1 cfu/mL. The relative selectivity of these ligands for E. coli, Citrobacter freundii, Staphylococcus epidermidis were 100%, 2.6% and 8.6% respectively. The biosensors were validated using spinach leaves at 3.0 cfu/mL. The work provides a generic biosensor for other pathogenic bacteria by enabling multivalent binding, immediate recognition for pathogens as well as inhibition of bacterial growth.

KEYWORDS: Escherichia coli; FimH lectin; MED; Mannose-binding protein; SPR

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