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Determination of free polysaccharide in Vi glycoconjugate vaccine against typhoid fever

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Highlights:

- A novel quantitative method for free Vi determination in glycoconjugates.
- The method works for Vi conjugates with different carrier proteins.
- The method is reproducible and allows Vi quantification in diluted samples.
- An improved quality control method for reliable Vi conjugates characterization.

Abstract

Glycoconjugate vaccines based on the Vi capsular polysaccharide directed against *Salmonella enterica* serovar Typhi are licensed or in development against typhoid fever, an important cause of morbidity and mortality in developing countries. Quantification of free polysaccharide in conjugate vaccines is an important quality control for release, to monitor vaccine stability and to ensure appropriate immune response. However, we found that existing separation methods based on size are not appropriate as free Vi non-specifically binds to unconjugated and conjugated protein. We developed a method based on free Vi separation by Capto Adhere resin and quantification by HPAEC-PAD. The method has been tested for conjugates of Vi derived from *Citrobacter freundii* with different carrier proteins such as CRM_{197}, Tetanus Toxoid and Diphtheria Toxoid.