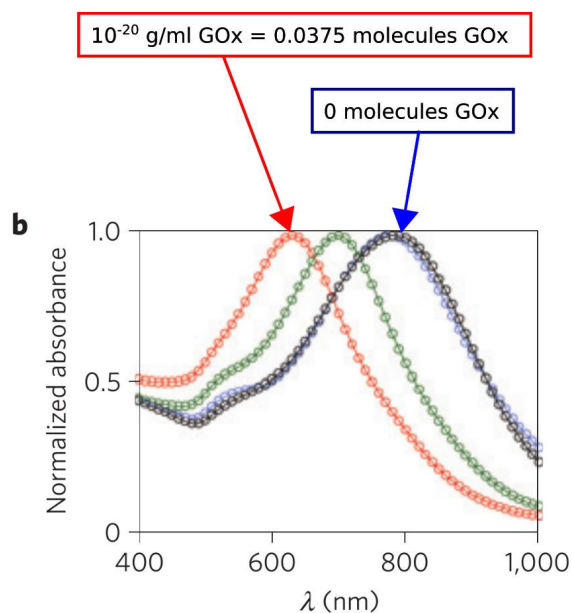


Unknown major signal generator undermines assay interpretation

Abstract

This paper reports a remarkably sensitive enzyme-linked antigen-detection assay. A positive result is detected as a blueshift of the absorption peak of a suspension of functionalised gold nanoparticles. However, identical solutions can produce assay results that differ by the maximum blueshift. The presence of such unexplained variance undermines the utility of the assay.



The problem is apparent in Fig. 2b of the article, reproduced here. A spectral shift equivalent to the maximum signal can be observed between conditions that are reported to differ only in the presence of 10⁻²⁰ g/ml Glucose Oxidase (GOx): the blue curve was obtained in the absence of the enzyme and the red curve in the presence of the indicated amount. Since the

reaction volume is 1 ml and the molecular weight of dimeric Glucose Oxidase is 160 kDa, 10^{-20} g/ml implies only 0.0375 molecules were present for the red curve. It is not credible to assert that a theoretical concentration so far below the single-molecule limit contained any GOx. Therefore, the red and blue curves were obtained under identical conditions, with zero GOx present in both. If the assay can produce both maximum and minimum signals under identical conditions, it is potentially unreliable. Without a convincing explanation of how the different signals could have arisen in Fig. 2b, the assay should not be relied upon.