intensity to enhance fluorescence signal whilst containing the DNA polymerase. In this regard, the inventors have shown that the intensity of light emitted from fluorophores attached to or co-located with the protein molecule within the hotspot can be enhanced relative to the intensity of light emitted from the corresponding fluorophores outside the hotspot. Most significantly, however, is the experimental finding by the inventors that, when DNA polymerase is tethered within the plasmonic hotspot of their nanoantenna in the presence of fluorescently-labelled deoxynucleotide triphosphates (dNTPs) and a DNA template, it is possible to detect the order of incorporation of specific, fluorescently-labelled dNTPs into a growing strand of DNA by the DNA polymerase enzyme during DNA synthesis by detecting the enhanced and unique fluorescence signal emitted from the respective dNTPs upon their incorporation within the plasmonic hotspot. Thus, the inventors have shown that the plasmonic nanoantenna of the disclosure can be used in single molecule DNA sequencing applications.

In one aspect, the present disclosure refers to a method comprising:

a) providing:

i) two plasmonic particles;

ii) a nucleic acid scaffold coupled to the two plasmonic particles; and

iii) a protein, wherein the protein is bound to the nucleic acid scaffold; and

b) with aid of the two plasmonic particles, generating an electrical field at the protein bound to the nucleic acid scaffold.

Accordingly, in one example, the disclosure provides method of sequencing a nucleic acid analyte, comprising:

(I) contacting a nucleic acid polymerase with the nucleic acid analyte and labelled nucleotides for a time and under conditions such that the labelled nucleotides are sequentially incorporated by the polymerase into a polynucleotide having a sequence which is complementary to a polynucleotide sequence of the nucleic acid analyte, wherein the polymerase is positioned within an area of electrical field enhancement; and each labelled nucleotide comprises:

(i) an adenine nucleotide (A), a guanosine nucleotide (G), a thymine nucleotide (T), or a cytosine nucleotide (C),

(ii) a fluorophore, and

(iii) a polyphosphate linker binding the nucleotide to the fluorophore;

wherein the A, G, T and C are each independently linked to a fluorophore via the polyphosphate,

wherein each of labelled nucleotides A, G, T and C has a distinct fluorescence emission signature when the fluorophore to which the respective nucleotide is linked is excited, and

wherein the distinct fluorescence emission signatures of the labelled nucleotides are enhanced upon their incorporation by the nucleic acid polymerase into the sequence which is complementary to a polynucleotide sequence of the nucleic acid analyte; and

(II) detecting the order of enhanced distinct fluorescence emission signatures as the labelled nucleotides are sequentially incorporated by the nucleic acid polymerase into the polynucleotide having a sequence which is complementary to a polynucleotide