

**Title** An electrochemical DNA sensor for the detection of ATP synthase gene expression in fresh-cut pineapple

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**Keyword** DNA sensor; ATP synthase; fresh-cut

### **Abstract**

ATP synthase is an indispensable enzyme in ATP production in living cells. Any cell deprived of its ATP synthases, does not survive. In order to monitoring the deterioration of cells in the fresh-cut pineapple tissue using ATP synthase as an indicator. A technique for the semi-quantitative determination of ATP synthase gene expression in fresh-cut pineapple based on electrochemical biosensor was developed. This technique employed RT-PCR amplification of a specific domain corresponding to a target ATP synthase gene and a common housekeeping 18S rRNA gene. Signals of gene expression was measured based on a phenomenon of DNA aggregation induced by Hoechst 33258 in conjunction with changes in anodic current peaks measured via a carbon screen printed electrode on linear sweep voltammetry. Anodic current peaks of the resulting cDNA products from fresh-cut tissues during storage were between 1.12-2.13 A. Semi-quantitative analysis for the level of ATP synthase gene expression was measured using the comparative ratio between copy numbers of ATP synthase gene and a housekeeping 18S rRNA. When this was used to monitoring the cells deterioration in fresh-cut pineapple, it was revealed that expression levels of ATP synthase were down to 10% of the originals upon deterioration and this was in correspondence with the results obtained using gel visualisation of RT-PCR products. The technique provided several merits especially on its rapidity and simplicity it monitoring the freshness based on semi-quantification of ATP synthase gene expression.