

## A novel probe for detecting two mutations on one RNA

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RNAs play fundamental roles in gene expression and regulation. Moreover, disease-causing mutations in RNAs are related to therapeutic targets. Therefore, methods for detecting, tracking, and visualizing RNA become an essential technology in molecular biology, especially in disease-related fields. Herein, we designed a novel probe set that can simultaneously detect double mutations on one RNA by fluorescence resonance energy transfer (FRET). In our design, DF-probe carrying FAM and dabcyI for detecting a cancer mutation, and T-probe carrying TAMRA for detecting drug resistance mutation in EGFR mRNA. The two probes were designed to be hybridized in part to cause FRET if both of the two mutations were present in the mRNA.

Using these two probes, we measured FRET efficiencies with wild-type and double-mutant RNAs, and we found a significant difference between them (Fig.1). These results demonstrate the feasibility of our probes to monitor the two mutations on one RNA. Thus, we conclude this probe set provides a method for detecting mutations on one RNA via FRET.

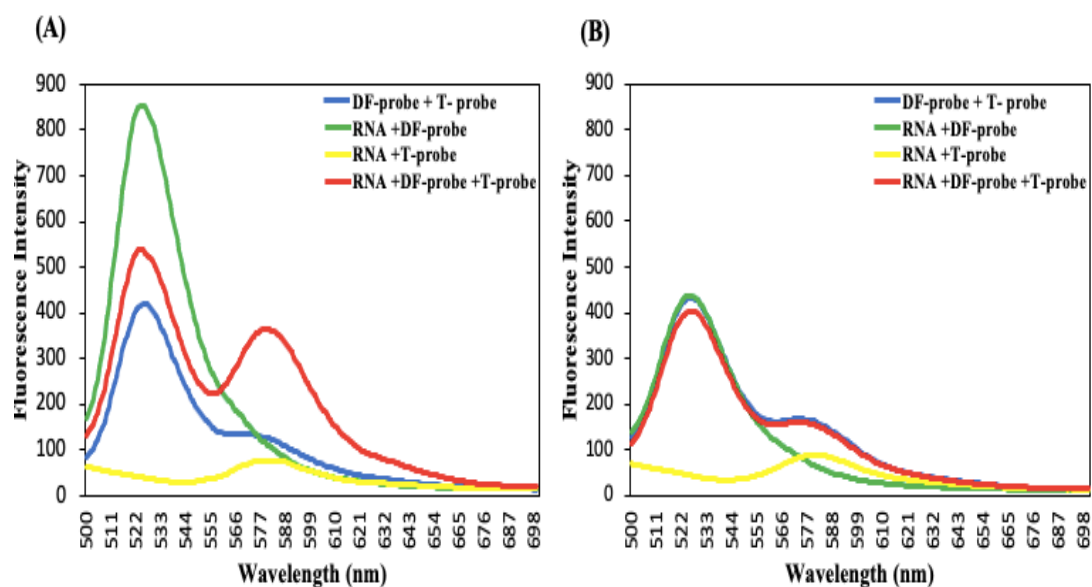


Figure 1. Fluorescence spectra of DF-probe and T-probe, (A) with mutant mRNA and (B) with wild-type mRNA.