## P28) Poster

# LABELING AMINE-MODIFIED DNA WITH A NOVEL AMINE-REACTIVE COUMARIN: AN EFFICIENT ALTERNATIVE FOR SYNTHESIZING SINGLY FLUORESCENT-LABELED FISH PROBES

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One of the strategies for synthesizing singly fluorescent-labeled oligonucleotide probes for their use in Fluorescence *In Situ* Hybridization (FISH) is the covalent coupling of amine-modified oligonucleotides with amine-reactive fluorescent dyes. Tetrafluorophenyl (TFP) esters are widely used with this aim. In this work the suitability of using a novel coumarin TFP ester for the synthesis of fluorescent FISH probes with a single fluorophore was investigated. The influence of various parameters (namely, previous purification of the 5'-amine modified oligonucleotide, spacer arm length (C6 or C12) and dye/oligo ratio) on: i) the labeling efficiency (dye/oligo molar ratio) and yield of the coupling reaction; and also on: ii) the FISH performance of the resulting labeled probes, was studied. 5'-amine modified EUK516 were used for performing all the labeling reactions and a well-established synthesis and purification protocol was applied. Fluorescent labeled oligo-coumarins with good FISH performance were obtained with the EUK516 that includes a C6 amino spacer arm. The labeling efficiencies (1/1 oligo/dye molar ratio) and the reaction yields obtained with this amino spacer (3-96%; maximal without previous purification of the oligo and using a dye/oligo ratio of 5/1 w/w) revealed that the novel coumarin is an efficient building block for synthesizing FISH probes with a single fluorophore.

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