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Evaluation of 2-Pyrazoline as a fluorescent label for quantifying biogenic amines using HPLC-FLD

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Introduction: The design and evaluation of derivatization reagents went a long way bearing comprehensive efforts and researches during the last decades. Many labeling reagents are commercially available for quantifying biogenic amines; however, these reagents show some drawbacks, such as toxicity, lack of sensitivity and selectivity, low solubility, and high cost. Using fluorescent heterocyclic compounds as labels is an outgrowing field in analytical chemistry. Pyrazolines are well known heterocycle were found to be appealing in many research fields due to their fascinating photophysical properties. The applications of these dyes span many areas from photodynamic cancer therapy, organic light emitting diodes, to dye sensitizing solar cell. However, recently it has been evaluated as potential candidates for pre-column derivatization of amino acids and primary alcohols.

The purpose: is to evaluate the adequacy of the newly synthesized pyrazoline, 4-(1-(4-trifluoromethyl)phenyl)-4, 5-dihydro-3-(naphthyl)-1H-pyrazole-5-yl)benzoic acid (PZCF3) as a label for biogenic amines and to develop a precolumn derivatization method for quantifying these analytes in different matrices.

Methodology: PZCF3 was synthesized by the conventional method, and characterized using 1H NMR, IR, HRMS and CHN methods. The photophysical properties including absorption, emission, and lifetime measurements have been studied in different solvents. Some selected biogenic amines were then derivatized by this reagent, and LC-MS was used to assess the produced derivatives. The derivatization procedure and the HPLC conditions were optimized, and the assay of biogenic amines by this method was validated.

Findings: PZCF3 shows excellent photophysical properties including high fluorescence intensity and quantum yield in different solvents. It is used to derivatize amino groups in a simple one step, at low temperature and in short reaction time. The investigated biogenic amines derivatives are stable and show strong well-separated peaks (resolution >1.5) on C18 column using 80% ACN in water. Excellent linear relationships were obtained for four amines in the range 2.5-25 μ mol L-1 (R2 \geq 0.991). The short elution time, feasibility, sensitivity, and reproducibility encourage the use of this dye for quantifying biogenic amines. Therefore, all figures of merit were calculated and the

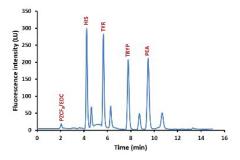


Fig 1: Schematic diagram of PZCF3

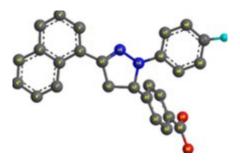


Fig 2: Chromatogram of biogenic amines derivatives

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Biography

Amal Al Sabahi obtained her BSc in science education from Sultan Qaboos University (SQU) in 1996 and MSc in Chemistry from Department of Chemistry, College of Science, SQU in 2003. Recently PhD holder in Chemistry from the same department. Worked as a chemistry teacher for 10 years and as educational researcher for 6 years. Acts as lab instructor in SQU for 3 years. Recent work depends on synthesizing and developing highly fluorescent labelling reagent for use in HPLC.

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