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Competitive fluorescent pseudo-immunoassay exploiting molecularly imprinted polymers for the detection of biogenic amines in fish matrix

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Abstract

We developed a competitive fluorescent molecularly imprinted polymer (MIP) assay to detect biogenic amines in fish samples. MIPs synthesized by precipitation polymerization using histamine as template were used in a batch binding assay analogous to competitive fluoroimmunoassays. Introducing a complex sample matrix, such as fish extract, into the assay changes the environment and the binding conditions, therefore the importance of the sample preparation is extensively discussed. Several extraction and purification methods for fish were comprehensively studied, and an optimal clean-up procedure for fish samples using liquid-liquid extraction was developed. The feasibility of the competitive MIP assay was shown in the purified fish extract over a broad histamine range (1 - 430 μ M). The MIP had the highest affinity towards histamine, but recognized also the structurally similar biogenic amines tyramine and tryptamine, as well as spermine and spermidine, providing simultaneous analysis and assessment of the total amount of biogenic amines.

Keywords: Molecularly imprinted polymer, biogenic amines, histamine, fluorescence detection, sample preparation, liquid – liquid extraction

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