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Deciphering zebrafish spectral signatures: insights from Raman spectroscopy

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Abstract

Background: Raman spectroscopy (RS) is a sensitive technique used for gathering chemical and molecular data, resulting in a biochemical fingerprint of the sample. This method is label-free and provides rapid detection [1]. Furthermore, it has proven invaluable in the forensic, biological and medical contexts [2]. It is also suitable for investigations assessing toxic effects of some chemicals, enabling swift acquisition of extensive organism-specific information. Its application to zebrafish serves various purposes, including physiological [3] and toxicological evaluation [4]. However, no base characterization of this organism has been provided in such studies that could be useful for the planning of experiments aiming at diagnosing and follow-up environmental contamination. **Objective:** The present study aimed to characterize the developmental Raman profile of different zebrafish organs, by drawing a baseline analysis of embryos and larvae up to 168 hpf. **Methods:** Different organs or tissues were examined daily from 24 to 168 hours post-fertilization, according to their time window of emergence in the embryo, including the heart, muscle, brain, iris, swim bladder, and melanocytes. Chemometric analysis, employing partial least-squares discriminant analysis (PLS-DA), was employed to characterize the organs and ascertain the contribution of spectral bands to their discrimination. **Results:** A total of 117 spectral bands were identified, with 24 demonstrating robust, systematic representation. The bands were found in the 223 to 3431 cm^{-1} spectral range; most of them were related to amino acids and relevant macromolecules, such nucleic acids, proteins and lipids. Only three bands were found to be common to all recorded organs and tissues. PLS-DA generated distinct spectral fingerprints for each organ, illustrating variations over early development. **Conclusions:** Overall, the work developed provided a clear baseline profile of organs and tissues of zebrafish embryos and larvae, identifying Raman bands of expeditious acquisition and their expected variation over different developmental stages, before and after hatching.

Keywords: chemometric analysis; early development; Raman bands; zebrafish organs

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