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Stephanie R. Weldon, PhD
Science Editor
JoVE

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Dear Dr. Weldon

Thank you for inviting us to publish our manuscript “Making the most of FFPE-RNA – optimization for sequencing and analysis of degraded FFPE-RNA samples” in the Journal of Visualized Experiments.

RNA sequencing has emerged as the leading method for gene expression analysis of different types of samples, providing valuable insights into cellular pathways determining disease and treatment outcomes. However, the most widely used method for preserving tissues in clinical settings generates Formalin-Fixed Paraffin-Embedded (FFPE) samples, that, often being highly degraded, fall short of the usual quality requirements for next-generation sequencing. In the attached manuscript, we describe a method for generating good quality data from such suboptimal samples. We list the various steps and precautions for sample quality control, sequencing library preparation, sequencing, and data analysis, that, taken together, increase the chances of reliable gene expression analysis from FFPE-RNA samples. We demonstrate this method using an example dataset of FFPE-RNA samples. We have highlighted parts of the protocol that can be explained better using the visual media.

Given the wide availability of FFPE tissues as samples, this method would be of broad interest to researchers trying to generate good quality gene expression data to understand the molecular mechanisms at work in various diseases.

The contents of this manuscript are original, and all authors of this paper have read and approved the final version.

Thank you for considering this manuscript. We look forward to your comments.

Best Regards,
Monika Mehta, Ph.D. [C]
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