

**Analysis of combinatorial miRNA treatment to regulate cell cycle and angiogenesis**

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**RESPONSE TO REVIEWERS' COMMENTS**

We thank the reviewers for their time, effort and for their insightful comments. We have revised our manuscript according to their suggestions. Following each of the reviewers' comments (in italics), we present our specific responses.

**Editorial and production comments:**

*Note the change in numbering (only the protocol should be numbered).*

*1. Please include comments in the Table of Materials so its clear which software is used for which analysis step (6.13, 7.33, 9.13).*

**Corrected**

*2. 1.5: 4 mL culture media, as appears to be the case in the video?*

**Corrected. Information added to the text protocol**

*3. 1.9: Steps 2 and 3 are mentioned here, but don't appear to be correct (even in the original numbering).*

**Corrected**

*4. 2: The bench is being sprayed down in the video; is this important for the protocol (e.g., if an RNase inhibitor is being used)? Please mention in the written protocol if so.*

**Added to the protocol**

*5. 2.5: How fast and how long do you centrifuge?*

**Information added**

*6. 2.12: Is this referring to section 8? This appears to be the case in the 7. Section 3: How is analysis done here?*

**This line was removed from the protocol, as it is explained in section 8.**

*8. 7: The order in the video is confusing-rinsing with deionized video is shown after what appears to be incubation in the coupling chamber but is before this step in the written protocol. Please clarify.*

**We added this information at step 7.26, which should better align with the video. There are multiple washing steps in this protocol. We included a clarification that ddH<sub>2</sub>O is the washing media, when not mentioned otherwise.**

*9. 7.32: Which wavelengths?*

**Information added**

*10. 8.5: The bioanalyzer does not appear to be in the Table of Materials.*

**Information added**

*11. 8.7-8.8: Please provide more details or references here.*

**References added**

*12. 8: Pathway analysis is mentioned in the heading, but is not detailed in the protocol itself.*

**The heading was corrected and the protocol section removed. The pathway analysis was part of a service. Indicative figure 5 demonstrates the potential analysis of the RNA -seq relative expression data.**

*13. Results, RNA sequencing...: Do you have any idea of the statistical tests done here?*

**Appropriate references were added.**