



# CALIFORNIA SCIENCE & ENGINEERING FAIR 2019 PROJECT SUMMARY

<b>Name(s)</b>  <b>Michelle Nazareth</b>	<b>Project Number</b>  <b>S0517</b>
<b>Project Title</b>  <b>RNA Regulation: Identifying and Preventing AMP Depurination in Early Life RNA Polymerization</b>	
<p style="text-align: center;"><b>Abstract</b></p> <p><b>Objectives</b> The first forms of life on Earth 4 billion years ago used RNA as a catalyst and way to store genetic information, but it is not known how the first RNA synthesized before life began, since there were no enzymes. RNA bases underwent repeated wet-dry cycles in volcanic conditions, which caused polymerization. However, adenosine monophosphate (AMP) often underwent depurination, meaning the adenine separated from the phosphate group, and deteriorated the nucleotide. My objective was to identify and prohibit AMP depurination in prebiotic conditions in the development of RNA, using lipid and adenine.</p> <p><b>Methods</b> Dilute phosphoric acid (H<sub>3</sub>PO<sub>4</sub>), AMP, adenine, and lysophosphatidylcholine (LPC) were used. Volcanic conditions were simulated by evaporating and re-hydrating AMP solutions in three thirty minute wet-dry cycles on a 85 degree Celsius hot plate. Either 10 mM adenine or 10 mM LPC was added to the mixtures. After cycle completion, samples were separated on silicic acid TLC plates using a 1.0 M lithium chloride solvent and illuminated with 254 nm UV light. AMP spots were scraped, hydrated, briefly sonicated, and centrifuged. Small aliquots were analyzed using a Nanodrop spectrometer, and the 260 nm absorbance value was used in Beer's Law calculations to find the amount of AMP or adenine remaining.</p> <p><b>Results</b> A protective effect of LPC and adenine was observed over three wet-dry cycles. On average, the AMP in the control samples underwent 33% depurination, while the corresponding rate of depurinated adenine increased by 50%. Samples containing LPC had elevated amounts of AMP in most trials. Adding adenine to the AMP solution protected against depurination and increased the amount of AMP by 21% on average. Additionally, through a hypothesis test using t-tests and p-values, I found that the adenine group significantly reduced AMP depurination when compared to the control group.</p> <p><b>Conclusions</b> Depurination was detected with samples containing AMP and H<sub>3</sub>PO<sub>4</sub> in simulated early life volcanic conditions. Adding LPC and adenine to the samples clearly reduced the amount of depurination and increased the AMP present. This suggests that in prebiotic conditions the integrity of AMP was protected in the presence of lipid and adenine.</p>	
<b>Summary Statement</b>  I identified and prevented AMP depurination using LPC and adenine in simulated early life conditions.	
<b>Help Received</b>  I conducted my research independently at the Startup Sandbox lab under the supervision and guidance of Dr. David Deamer. Ms. Cristie Kirlin was my school advisor.	