Title – HPLC Analysis of Amino Acids Formed by Urey-Miller-Type Experiments

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Mentor and Mentor Email - Dr. Michael Korn (mrkorn@liberty.edu)

Student name and email – Isabella Caramante (igcaramante@liberty.edu)

Abstract: Urey-Miller-type experiments use H_2O, CH_4, NH_3, H_2, N_2, CO_2, and other gases to produce amino acids. These gas mixtures were used to simulate the environment that the earth was theorized to have. Five main amino acids produced in these experiments are glycine, alanine, glutamic acid, aspartic acid, and serine. High-Performance Liquid Chromatography (HPLC) is used to separate amino acids. In HPLC, amino acid detection is increased by attaching a chromophore that fluoresces in the UV light range. Chromophores are unsaturated groups, mainly aromatic rings, that absorb and reflect light at a specific wavelength. To allow for visualization with fluorescent detectors, o-phthalaldehyde (OPA) with 3-mercaptopropionic acid (MPA) is used to derivatize the amino acids. To separate racemic mixtures of amino acids, the amino acids are reacted with OPA and N-acetyl-L-cysteine (NAC) to produce two diastereomers that can be detected. The HPLC protocol, provided by the manufacturer (Agilent), was followed. For the experiment, the five amino acids mentioned above have been run individually and together as mixtures. Based on polarity, different amino acids pass through the column at different rates. The results of the different signal intensities and the time it takes for each amino to pass through the column have been overlaid to identify which signals are which amino acids.

By varying amino acid concentration, the results are used to relate absorption intensity with concentration. The goal of this research project is to create an amino acid standard curve through effective separation techniques, to relate absorbance intensity with time for each of the five amino acids, and to use the standard curve to determine amino acid composition of unknown mixtures. Urey-Miller-type experiments will be conducted, and then amino acid composition, concentration, and optical purity will be tested on these unknown mixtures.