Exposing Microorganisms in the Stratosphere (E-MIST) C. L. Khodadad[1], and Phillip R. Maloney[2], [1]Sierra Lobo, Inc., Mail-code ESC-24. Kennedy Space Center, FL 32899 christina.l.khodadad-1@nasa.gov; Phillip R. Maloney[2], NASA, Kennedy Space Center, FL 32899 phillip.r.maloney@nasa.gov; Gregory M. Wong[3], [3]Wesleyan University, Middletown, CT 06459 gmwong@wesleyan.edu; and D. J. Smith[4], [4]NASA Ames Research Center, Mail-code SCR, Moffett Field, CA 94035-0001 david.j.smith-3@nasa.gov

Introduction: Microorganisms detected in the upper atmosphere originate from both terrestrial and aquatic ecosystems on Earth, can be dispersed long distances on Earth by prevailing winds, or may reach the troposphere and stratosphere by uplifting forces or atmospheric mixing. Extreme environmental conditions in these systems exert a combination of stressors on these microbes with the potential to generate cell damage or create DNA mutations. Numerous ground studies have illustrated these effects or adaptations and the capacity of microorganisms to adjust and persist [1,2]. However, further studies of survival and adaptation of microorganisms in the upper atmosphere are required to fully understand how microorganisms might survive the harsh environments of space. The goal of this study was to establish baseline data for the E-MISTpayload, to establish microbiological and molecular protocols, and to investigate the response of the target organism, *Bacillus pumilus*, to extreme conditions of Earth's stratosphere. *Bacillus pumilus*, SAFR-032 is a resilient, spore forming bacterial strain isolated from a clean room environment in a NASA spacecraft assembly facility at the Jet Propulsion Laboratory, CA [3].

Materials and Methods: To further study survival and responses of microorganisms in the upper atmosphere a research platform (83.3 cm x 53.3 cm x 25.4 cm; mass 36 kg) designed and built at Kennedy Space Center, FL transported biological samples and environmental sensors to Earth's upper atmosphere to establish baseline data and confirm molecular protocols (Fig 1).

Launched aboard a balloon gondola, the payload traveled to the upper atmosphere where environmental conditions as atmospheric pressure, temperatures, and relative humidity could be recorded along with radiometric data to monitor UV exposure (230 to 400 nm) of the biological organisms. Four skewers each contained up to 10 aluminum coupons each with approximately 1 X 10^6 *B. pumilus* spores. Each skewer rotated 180° exposing the spore containing coupons to the stratosphere. Due to their resiliency, it is suspected that *B. pumilus* spores might survive transport through Earth's upper atmosphere and provide indications of the survivability or response to deep space exposure (e.g. surface of Mars).

Results and Discussion: Baseline data established during the first flight test of the E-MIST payload determined that the experimental coupons containing spores exhibited similar viability suggesting the established payload procedures had no effect on spore survival [4]. Negative controls included in the flight showed no outside contamination during flight or transport [4]. Concerns of whether microorganisms can survive deep space conditions and potentially be transported to other planets such as Mars has long been a concern to scientists. To further this understanding it is necessary to determine if there is a genomic effect that occurs with survival in this extreme environment. A second flight of the E-MIST payload to the upper atmosphere will expose the *Bacillus pumilus* spores to the harsh environment for an extended period of time. Once returned to ground, an investigation into genomic changes will be completed by re-sequencing and comparing the genomic sequences after exposure to the harsh environmental conditions. In addition, ground studies will be conducted in parallel simulating similar environmental conditions (e.g. UV irradiation). It will also provide valuable information for planetary protection as well as terrestrial ecologists regarding the global transport of microorganisms between ecosystems.

References: [1] D.W. Griffin (2007) *Clinical Microbiology Reviews* 20: 459-477. [2] P.N. Polymenakou (2012) *Atmosphere* 3:87-102. [3] M.J. Kempf (2005) *Astrobiology*, 5: 391-405. [4] D. J. Smith et al. (2014) *Gravitational and Space Research, 2,* 70–80.