**REACTIVE OXYGEN SPECIES GENERATION BY LUNAR SIMULANTS.** Jasmeet Kaur<sup>1,3</sup>,Martin A. Schoonen<sup>1,3</sup>, Douglas Rickman<sup>2</sup>, <sup>1</sup>Department of Geosciences, Stony Brook University,Stony Brook-11794-2100 ( jasmeet.kaur@stonybrook.edu and martin.schoonen@stonybrook.edu); <sup>2</sup>Earth Science Office, National Aeronautics and Space Administration, Marshal Space Flight Center, Alabama-35812(doug.rickman@nasa.gov); <sup>3</sup>RIS<sup>4</sup>E, Stony Brook University, NY 11794-2100.

**Introduction:** The current interest in human exploration of the Moon and past experiences of Apollo astronauts has rekindled research into the harmful effects of Lunar dust on human health. While the mineralogical composition of lunar regolith has been well documented, other factors, such as partial melting due to space weathering, UV irradiation, and dryness may also contribute to the toxicity of lunar dust. For example, the presence of elemental iron "nano-particles" in agglutinatic material in the respirable size fraction has been recognized as a possible health concern[1]. Building on earlier work on mineral toxicity [2-5], we have started a research program focused on the reactivity of lunar dust in the context of inhalation exposures. As a first step, we have evaluated the generation of Reactive Oxygen Species (ROS) by several Lunar simulants.

**Background**: ROS are chemically reactive molecules containing oxygen and include superoxide (O<sub>2</sub>-), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and hydroxyl radicals (OH). Previous studies have shown that mineral dust generates ROS when dispersed in water [6]. Minerals generate ROS either by surface defects or step-wise reduction of molecular oxygen. In the human body, ROS are produced by various endogenous systems and they play an important role in the normal functioning of cells. However, increased levels of ROS as a result of exposure to mineral dust can lead to oxidative stress, inflammation, genotoxicity (DNA damage) or apoptosis (programmed cell death) [7].

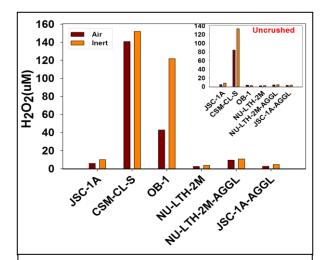
**Methods:** In the present work, we quantified  $H_2O_2$  and 'OH formation for a suite of different lunar simulants upon dispersion in water and simulated lung fluid (SLF). H<sub>2</sub>O<sub>2</sub> was measured using a dedicated electrochemical probe providing real-time data, while 'OH radical formation was determined in separate experiments using a spin trap technique followed by detection using Electron Spin Resonance (ESR) spectroscopy. In all H<sub>2</sub>O<sub>2</sub> detection experiments in water we used EDTA to inhibit the Fenton Reaction, which converts H<sub>2</sub>O<sub>2</sub> to 'OH. If EDTA is not added, H<sub>2</sub>O<sub>2</sub> concentraions are near or below the detection limit, presumably as a result of conversion to 'OH. For ESR technique we used spin trap 5, 5-Dimethyl-1-pyrroline Noxide (DMPO) which forms a stable adduct with the unpaired electron. The intensity of adduct measured using ESR is directly proportional to 'OH concentration and was quantified using hydroxyl 2,2,6,6 tetramethylpiperidine (TEMPOL). We compared the reactivity of these simulants prepared in air and inert atmosphere. We also studied the effect of mechanical stress by hand crushing samples in a mortar and pestle. The effect of mechanical stress by crushing on the production of  $\rm H_2O_2$  was evaluated upto a period of nine days after the treatment.

Sample Descriptions: JSC-1A is a volcanic ash from Merriam Crater, AZ. NU-LHT-2M is a complex mixture [8]. CSM-CL-S is milled from a scoria from +36° 49′ 22″, -104° 9′ 17″. OB-1 is a mixture of anorthosite from the Shawmere Anorthosite, Ontario, plus slag from a smelter near Sudbury, Ontario. \*\*\*\*-AGGL are made from the source simulant by Orbitec Technologies Corporation in a process that converts some of the material to agglutinate particles which contain nanophase-iron. Each of the source simulants were designed and milled to yield a specific, and similar, particle size distribution.

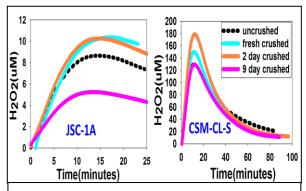
**Results:**  $H_2O_2$  detection experiments dispersed in water showed more  $H_2O_2$  formation with simulants prepared in inert atmosphere than those prepared in air. Fresh crushed samples generated more  $H_2O_2$  than uncrushed samples (see Fig. 1). The reactivity of fresh crushed samples decreased with time. For instance, in inert atmosphere, nine-day crushed JSC-1A generated  $5\mu M$  of  $H_2O_2$  while fresh crushed generated  $10\mu M$  of  $H_2O_2$ . Similarly, nine-day crushed CSM-CL-S generated  $120\mu M$  of  $H_2O_2$  which was similar to the concentration generated by the uncrushed sample while two-day crushed sample generated  $180\mu M$  (Fig. 2). Based on  $H_2O_2$  detection experiments, JSC-1A, CSM-CL-S and OB-1 were found to be the most reactive of all simulants.

ESR results with these three samples showed that fresh crushed JSC-1A and CSM-CL-S generated six times more 'OH concentration compared to the uncrushed samples and fresh crushed OB-1 generated two and a half times more 'OH compared to the uncrushed sample. The high reactivity of fresh crushed samples is likely due to the broken surface bonds. Pre-liminary  $\rm H_2O_2$  detection experiments in SLF showed continued generation of  $\rm H_2O_2$  for a period of about seven hours at a rate of  $8.5\mu M/hr$ . This was in contrast to experiments in DI where  $\rm H_2O_2$  concentration after reaching a peak

value in about 10-20 minutes of reaction starts to decline.



**Figure 1:** Comparison of  $H_2O_2$  peak values of fresh crushed samples in air and  $N_2$  environment. Inset shows the peak values for uncrushed samples. Except for CSM-CL-S notice the low reactivity of uncrushed samples.



**Figure 2:** Effect of mechanical stress on  $H_2O_2$  formation by JSC-1A and CSM-CL-S in inert atmosphere. Notice the decrease in  $H_2O_2$  with increase in treatment time.

**Future work:** In a second phase, the effects of dehydrating simulants in vacuum and irradiation with UV will be evaluated. Also additional experiments will be conducted in simulated lung fluid to more closely match the chemical environment within the human lung.

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