Colorado Space Grant Consortium

University of Northern Colorado
DEMOSAT PROJECT
SUMMER 2015

Effects of Cosmic Radiation on DNA and Quantum Dots

Written by:
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August 13, 2015
Revision D

Revision Log

Revision	Description	Date
A	Conceptual Design Review	4/2015
B	Preliminary Design Review	6/5/2015
C	Critical Design Review	7/3/2015
D	Analysis and Final Report	8/14/2015

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1.0 Mission Overview

Our mission for this launch was to analyze the effects that a high altitude environment has on the structure of DNA and quantum dots. This flight will hopefully show what effects radiation has on the DNA samples and quantum dot samples. The results of the launch and testing will be used in discovering how structural DNA and quantum dots can be used together to create new materials, which could potentially be used for different types of technology in the future, especially for technology that will be used in spaceflight. These ideas come from previous research done with quantum dot based photovoltaic cells (Fedoseyev, A., Turowski, M., Raman, A., Taylor, E., Hubbard, S., Polly, S. , . . Balandin, A. (2009). Space radiation effects modeling and analysis of quantum dot based photovoltaic cells. Nanophotonics and Macrophotonics for Space Environments III.). The testing methods that we will use are gel electrophoresis and UV-vis. These methods are based on research done with radiation and DNA and the methods that other scientists used to gather their data (Swiderek, P. (2006). Fundamental Processes Radiation Damage of DNA. Angew. Chem. Int. Ed. Angewandte Chemie International Edition, 4056-4059.).

2.0 Requirements Flow Down

The top level requirements of the payload deal with the housed samples of DNA and Quantum dots. The payload was required to safely house the samples, which were kept in glass vials, through the entire flight. Level 1 requirements included heating, and data acquisition. Level 2 requirements included data logging and power.

The DNA and Quantum dots were suspended in an aqueous solution that had to remain above 20 °C and under 45 °C. These temperatures were derived from the structural stability of the DNA, 20 °C as a bottom floor as we wanted to ensure the solution didn't freeze, and 45 °C as a roof, because we wanted to keep the temperature below the DNA's melting point. Because the goal of the payload was to analyze how non-UV/VIS radiation interacts with DNA structure, it was vital to limit the number of variables that would affect the DNA's structural stability.

Data acquisition was derived from the various environmental variables we wanted to examine to compare in conjunction with the lab data that would be gathered post flight. Environmental factors we wanted to include were temperatures in various locations of the box, flux in radiation, altitude, and internal box pressure.

Temperature data from the internal box, the Geiger counter enclosure, and the sample enclosure were to be recorded. The internal box temperature

was to be taken, so that we could examine the overall effectiveness of the heating element's ability to heat the entire box. The Geiger counter temperature was taken to examine the heating elements effect on the internal temperature of a pressurized container located non-adjacent to the heating element. The temperature data from the sample enclosure was useful in two regards. The first was to ensure we stayed within the temperature limits dictated by the DNA's melting point, the second was to examine the effectiveness of the heating element to heat an adjacent pressurized enclosure. Flux in radiation was measured with a Geiger counter. The flux in radiation as well as altitude and pressure readings were used to compare with old data to examine variation and changes, as well as to get a better idea of the conditions of the high altitude environment reached during flight for future projects.

Data logging was used to save all of the data acquired through the various sensors, and power was derived based off of expected flight time and the mA/hr our system required to remain active and recording for the full duration of that expected flight time.

3.0 Design

In the Design and Construction of the 2015 University of Northern Colorado DemoSat project the team decided to design a modular box and components that was designed with Solidworks software and constructed using a TazBot 3D printing system. (Image 1) In the modular portion of the box all components necessary for the experiments were mounted using 3D printed brackets and mounts. (Image 2) The modular interior of the box was then enclosed with 1 centimeter (cm) thick piece of foam board. The foam board was wrapped in a reflective Mylar sheet and the interior walls of the foam were covered in 2 cm thick piece of fiberglass insulation. (Image 3) The walls of the box were attached to the modular interior using hot glue and aluminum tape.

Modular Design:

The 3D printed modular interior of the box went through many different renditions before a final working model was completed. The design and construction using the 3D printer required a sharp learning curve of the software and use of the equipment, but in the end we successfully printed and used the entire printed framework, mounts and brackets.

Interior Components:

The vials which contain the Quantum Dots and DNA samples were incased in foam and placed inside a sealed Polyvinyl Chloride (PVC) pipe and mounted using a 3D printed bracket. The Geiger counter was also placed inside a sealed PVC pipe as to maintain atmosphere and prevent arcing of the circuits. The 8 batteries (9 Volt) and electronic components such as the Arduino and heater shared a common 3D printed bracket and mounted opposite the PVC pipes holding the experiments and Geiger counter.

Heating System:

The initial plan for the heating system required the use of several small heating pads that were supposed to line the inside walls of the box. However, after having some issues with using 9 Volt batteries to power the heating pads we transitioned into using a set of 6 1000 Ohm resistors wired in a circuit to heat the interior of the box.

Box	
	ABS Plastic Filament (500 cm) Foam Board (36x24 inches 1 cm thick) Mylar Cloth (132x213 cm) Aluminum Tape (5 cm wide) M3 Bolts and Nuts x6 Fiberglass Insulation
Hardware	
	Geiger Counter (Sparkfun) Micro SD shield (Sparkfun) Temperature Sensor (Sparkfun) FTDI Board (Sparkfun) Altimeter/Pressure Sensor (Sparkfun) Current Sensor (Sparkfun) Voltage Regulator (5V) (Sparkfun) 9V Lithium Batteries X8 1 k ohm resistors x6 Arduino Uno (Sparkfun)

Image 1

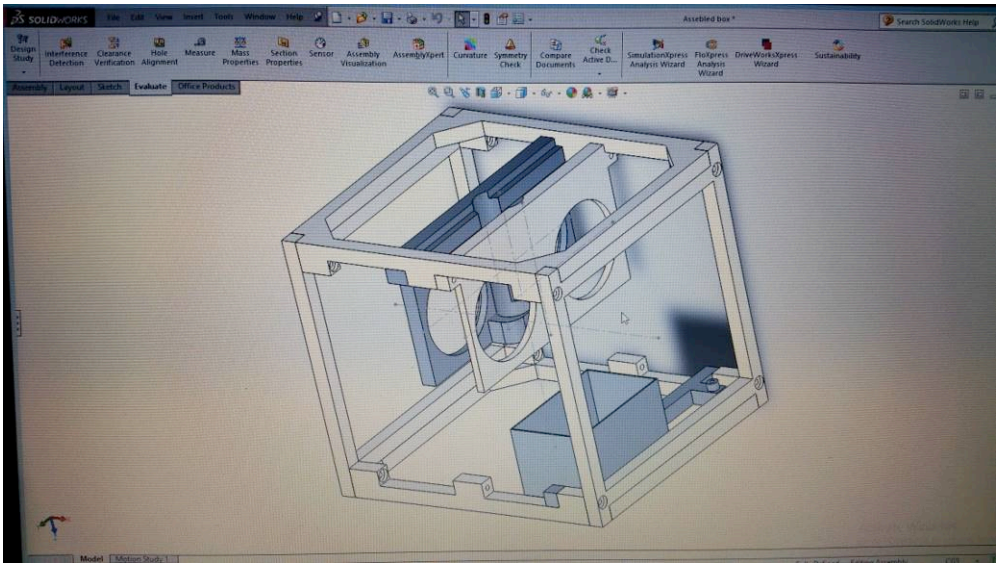


Image 2

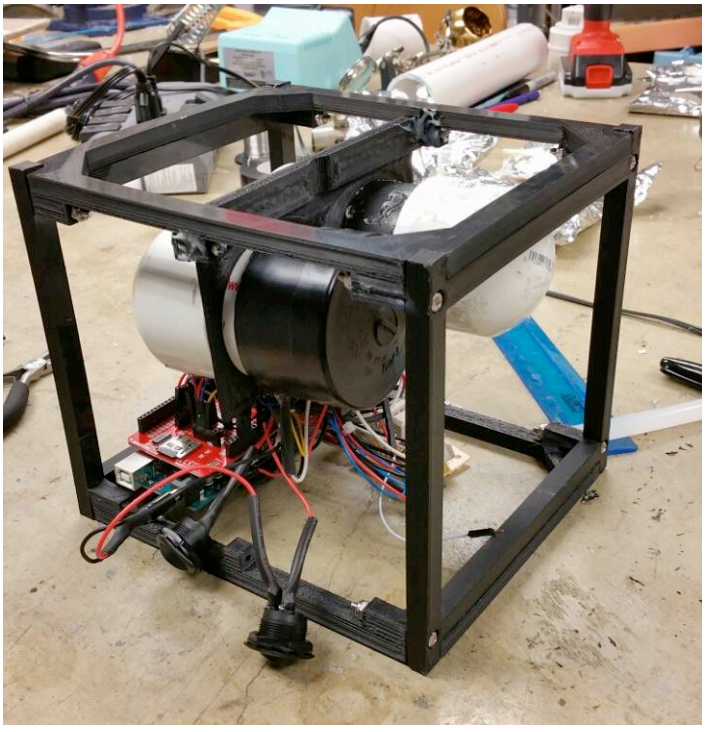
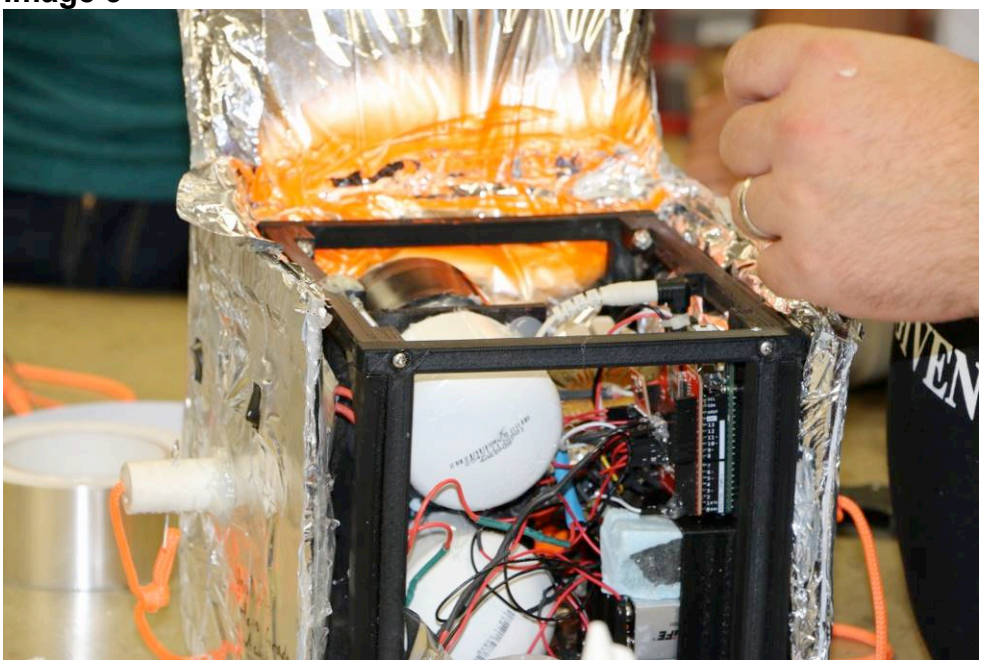


Image 3



4.0 Management

Our team is comprised of 9 students, of which 7 are from the University of Northern Colorado and 2 are from Front Range Community College. Our group includes 2 females and 7 males. All of our college level participants will be juniors in the 2015/2016 school year. These 13 students were split into 5 sub-systems (Quantum & DNA, Thermal, Electronics, Structural engineering and FSI payload).

Project Manager: Ryan Fabian

Qd & DNA: Zachary Blocker, Alexandra Briggs, Steven Diaz

Structural: Andrew Bragg, Kyle Morman, Ryan Angstead

Thermal: Abigail Robertson, Kyle Morman, Andrew Bragg

Electronics: Arick Swietzer

FSI Payload: Josh Seerdorf, Grace VanderWoude, Steven Cromer

The schedule that we initially created was overly optimistic. We failed to consider issues such as the 3-D printer failing and members of the electronics sub-system team not being able to continue to work on the project. These two issues caused us to miss the majority of our scheduled deadlines. However, we have been able to meet all of the Colorado Space Grant deadlines.

Event	Summer 2015
Kickoff telecon	May 22
Preliminary Design Review	June 1 - 5
First box design built	June 15
Box testing	June 25
Electrical components testing	June 30
Critical Design Review	June 29 - July 3
Testing of all components	July 15th
Sample standards created	July 15th
Launch Readiness Review	July 27 - 31
LAUNCH!	August 1
Post-launch testing and analysis	August 1-7
Final Reports Due	August 14

5.0 Budget

All DEMOSAT FY15-16 purchases:

2/27/2015	226.00	Nano. & Amorph Mat
3/23/2015	92.90	Edmund Scientifics - Laser
3/18/2015	96.70	Batteries Plus
4/6/2015	500.00	NanoLab - SWNT Suspensions
5/15/2015	15.45	Amazon - Corona Dope/Demosat
5/15/2015	106.03	Amazon - Misc Demosat

5/18/2019	156.72	Sigma-Aldrich/Vials & Cups for Demosat
5/20/2015	34.00	Subway - 5/20 Demosat Food
5/27/2015	45.05	Blackjack Pizza - 5/27 Demosat Food
5/27/2015	261.80	Sparkfun - Demosat Misc.
5/27/2015	127.01	Sparkfun - Demosat Geiger
6/1/2015	49.77	Sparkfun - Demosat Heating Pads
6/1/2015	244.60	IDT - DNA Duplexes
6/3/2015	40.48	Papa John's - 6/3 Demosat Food
6/3/2015	185.30	Ocean NanoTech - Quantum Dots
6/5/2015	51.02	Amazon - Misc Demosat Organizing Supplies
6/5/2015	36.03	Amazon - Demosat Supplies
6/5/2015	26.75	Amazon - Demosat Cables
6/8/2015	128.00	Novedge - SolidWorks
6/9/2015	57.11	Amazon - Demosat Storage Supplies
6/11/2015	165.36	SparkFun - Arduino/Headers - Demosat
6/11/2015	6.53	SparkFun - Female Headers - Demosat
6/12/2015	31.14	SparkFun - Duck Antenna - Demosat
6/12/2015	27.98	Demosat
6/12/2015	143.13	Home Depot - Demosat Supplies
6/13/2015	32.98	Lowe's - Demosat Supplies
6/10/2015	45.05	Blackjack Pizza - 6/10 Demosat Food
6/17/2015	40.65	Domino's Pizza - 6/17 Demosat Food
6/19/2015	42.30	Parallax - Distance Sensor/Demosat
6/22/2015	113.87	Agarose/Demosat/Blocker
6/24/2015	47.45	Blackjack Pizza - 6/24 Demosat Food
6/30/2015	58.86	Harbor Freight/Misc Demosat Supplies
7/1/2015	49.14	Domino's/Demosat 7/1 Meeting Food
7/1/2015	216.21	Amazon/Misc Demosat/Walch
7/10/2015	53.11	Sparkfun/3D Filament/Demosat
7/13/2015	23.17	Ace Hardware/Demosat Misc
7/13/2015	227.25	Life Technologies Corp./Demosat DNA Supplies
7/14/2015	25.52	Amazon/Tape/Demosat
7/15/2015	4.50	CSU Demosat Visit Parking
7/15/2015	32.76	FSI Students CSU Demosat Visit Mileage
7/18/2015	47.80	McMaster-Carr/Demosat Ultem Sheet
7/18/2015	7.34	Amazon/LED Worklight/Demosat
7/18/2015	60.35	Amazon/Foam Board for Demosat
7/18/2015	18.74	Amazon/Duct Tape for Demosat
7/21/2015	13.96	Radio Shack/Demosat Misc
7/21/2015	40.00	Buy Demosat Goggles from Chem. Dept.

7/22/2015	59.98	Domino's/Demosat 7/22 Meeting Food
7/22/2015	18.69	Domino's/Demosat 7/22 Meeting Food
7/27/2015	50.33	Batteries Plus - Batteries for DemoSAT
7/28/2015	5.99	Ace Hardware/Demosat Misc
7/29/2015	32.31	Fisher Scientific/Pipets for Demosat
8/3/2015	7.99	Batteries Plus - Batteries for DemoSAT
8/4/2015	59.69	Amazon - Demosat Misc Supplies
8/4/2015	2.79	Amazon - Demosat Wire Holder
7/15	3,850.00	Student Demosat Stipends
	8,143.64	**TOTAL DEMOSAT FY15-16**

6.0 Test Plan and Results

Box Testing:

Cold Test (July 28, 2015) 	**Test Setup Overview**: Liquid nitrogen inside a Styrofoam cooler which we placed the payload box into. The external test environment maintained a temperature of -196.15 °C
Objective: Confirm temp. can be maintained between 0 and 32	**Results**: The interior of your payload stayed above 0 °C for the duration of the test.

Data Collection Test (July 27, 2015) 	**Test Setup Overview**: Power on and collect data for 3 hours and compare data to calibrated equipment We used a calibrated Geiger counter, thermometer, current pressure and altitude data to check the accuracy or our electronics. We ran the test for 3 hours and then compared our electronics data to the same collection when using the PC's power.
Objective: Confirm accuracy of data collection	**Results**: Data was collected properly. However, the duration of data collection was not sufficient. Adjustments were made to the battery pack in order to extend collection time in response.
Integrity test (July 27, 2015) 	**Test Setup Overview**: We dropped the box from the top of Ross Hall ~45 feet, we then conducted the whip test, drag test and dropped box down two flights of stairs.
Objective: Confirm structural integrity	**Results**: After the drop test, the box did show some exterior damage. However, the internal components remained intact. During the whip test, drag test and stair test, the box maintained its structural integrity.

Sample Testing:

Melting Point Determination (DNA)	**Test Setup Overview**: Samples are loaded into cuvettes and data is collected by a UV/Vis Absorbance spectrometer. The temperature of the samples in slowly increased, allowing for changes in absorbance to be correlated to changes in temperature. Pre and post flight samples will be compared.
Objective: Determine any differences in the temperature in which the dsDNA strands between the control and the flight samples.	
Gel Electrophoresis (DNA and QDs)	**Test Setup Overview**: Samples are placed into a dye-containing agarose gel with multiple wells. Each sample is placed into a well and pulled through the gel by electrical potential through a conductive aqueous buffer. When imaged under UV light, the distance the samples traveled through the gel will be revealed and compared.
Objective: Determine if the flight samples experience any fragmentation due to ionizing radiation.	

7.0 Expected Results

Because the nature of the upper atmospheric radiation expected on the samples, a couple of possible outcomes have been hypothesized. Since the radiation is high in energy, it is possible that the molecule may be severed or destroyed.

Duplexed DNA is held together by hydrogen bonding, which although a somewhat strong intermolecular force, is a weak bond. It can be then assumed that these bonds would be the first broken by the high-energy radiation. Furthermore, it is known that bombarding an organic molecule with electrons with high energy will cause a radical molecule (a molecule with an extra electron) to form. These radicals are highly unstable, so they split into fragments to increase their stability. Thus, another possibility is that the high-energy radiation can cause the molecule to become unstable and cause fragmentation.

A second possibility is that due to the design of the payload, the sample may be completely unaffected by the radiation. The contributing factor to this possibility is the knowledge of the amount of protection the DNA needs. The DNA will be inside glass vials, inside of a PVC pipe, inside of the flight container, which is filled with insulation and covered with Mylar. These items are necessities due to the nature of the DNA. The most observable changes to DNA will be when it is dissolved in a solvent, typically water. To keep the water in liquid form, it will be contained in a PVC pipe that will keep a constant pressure, which, in turn, keeps the boiling point of water 100° C. Furthermore, it is easier to transfer heat if there are molecules present. Because of all this shielding, it may be hard or impossible for any radiation to reach the DNA, which would result in unaffected DNA.

To test the few hypotheses, a few analytical techniques will be used. Two techniques will be explored on the DNA: UV-Vis spectroscopy and Gel Electrophoresis. Both techniques will provide comparative information when sampled along with a controlled sample.

To perform a UV-Vis Spectroscopy, an Agilent 8453 UV-Vis Spectrometer was used. The use of this instrument was gratuitously provided by the Chemistry department at UNC. To gather data from the UV-Vis, a sample is irradiated with light, and its absorptivity is measured. Using a quartz cuvette, the sample is irradiated with wavelengths of light from 200 to 1100nm.

While the sample is irradiated, it is also heated to cause a separation of double stranded DNA (dsDNA).

DNA is known to absorb light at the 260nm range due to the aromaticity of the molecule, so this technique can serve to approximate the Melting Temperature (T_M), the temperature at which the DNA duplexes separate. Due to the change from dsDNA to single stranded DNA (ssDNA), the absorptivity will increase as the concentration of aromatic molecules increases. The spectrometer will then approximate the T_M using the first derivative of the curve created by plotting the absorptivity vs. temperature. Using the first derivative, the program will look for an inflection point on the graph, which will approximately be the T_M of the DNA samples.

The T_M of DNA is relatively constant. It is dependent on the pH of the solution, the salt concentration, and the composition of the DNA. To create a constant T_M, the variables that affect it will be held constant among the control and sample group; thus, if the T_M changes post-flight, we can assume the DNA was affected by the radiation.

The other technique being used to analyze the DNA is Gel Electrophoresis. Gel electrophoresis uses an applied current over a gel mobile phase to move samples through the gel. Molecules with high molecular weight (measured in kDa) have more difficulty traveling through the gel, and as a result, standards can be made through gel electrophoresis in accordance to mass and charge. If the sample is fragmented through the flight, the gel electrophoresis can pick up fragmentation because the fragments will travel further through the gel.

Quantum Dot (QD) testing is currently under review. It may be possible to perform a UV-Vis spectroscopy to measure its absorbance pre and post-flight. However, the molar absorptivity of the QDs being used is unknown, thus performing UV-Vis spectroscopy may prove to be an ineffective way at measuring any changes on the QDs.

8.0 Launch and Recovery

Launch:

The University of Northern Colorado sent a team of eight people to the launch and recovery site of the 2015 Demo Sat project. The team consisted of Ryan Fabian, Steven Diaz, Alex Briggs, Arick Sweitzer, Ryan Angstead, Zach Blocker, Dr. Walsh and Dr. Semak. The launch took place outside

Wiggins, Colorado and all of the team members arrived at the location approximately 1 hour prior to launch. At the time of launch our payload weighed in at 1.963 kilograms, which was slightly over our expanded weight restriction. Arick Sweitzer had the pleasure of holding and sending off our payload at the time of launch. The launch occurred at roughly 06:00 am and went off without a hitch.

Recovery:

Three vehicles went on the recovery mission after launch driven by Ryan Fabian, Ryan Angstead and Dr. Semak. Post launch, the payload traveled south east approximately 6 miles and achieved an altitude of just over 103,000 feet (3 kilometers). Upon landing, after a nearly 2 hour flight, the Payload was recovered in a cattle field at 08:30 am. The impact of the landing had a minimal effect on the structural integrity of the box and components. After retrieval, we waited to recover the experiments and data until the box could be safely opened under controlled conditions in the lab at the University of Northern Colorado. This was to insure the integrity of the electronics and the purity of the experimental samples. After we opened the payload, all experiments and electronics seemed to be intact and no significant damage had occurred. We consider this to be a successful launch and recovery of the 2015 Demo Sat project.

9.0 Results, Analysis, and Conclusions

After performing adequate analysis on the DNA, it can be safely assumed that the second hypothesis stated in Section 7 was correct. This will be explained further throughout this section.

With the use of the UV-Vis spectrometer, the T_M of both strands of DNA was determined. To ensure the T_M of the strands[1], trials were run in triplicate for one strand and duplicate for the other. The T_M for the $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ was determined to be approximately. The T_M for the $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ was determined to be approximately.

Tests of the irradiated samples from the flight were also performed. The T_M of those samples was also calculated. The T_M for the $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ sample was determined to be approximately. The T_M for the $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ sample was determined to be approximately.

[1] It should be noted that all DNA strands are assumed to be duplexed with its complimentary strand unless otherwise stated.

Comparison of the T_M of the controls and the samples, it seems as nothing occurred. Had damage occurred to the DNA, the T_M plots would show differences.

After performing adequate analysis on the DNA, it can be safely assumed that the second hypothesis stated in Section 7 was correct. This will be explained further throughout this section.

With the use of the UV-Vis spectrometer, the T_M of both strands of DNA was determined. To ensure the T_M of the strands[2], trials were run in triplicate for one strand and duplicate for the other. The T_M for the $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ was determined to be approximately 57.50°C. The T_M for the $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ was determined to be approximately 57.70°C.

Tests of the irradiated samples from the flight were also performed. The T_M of those samples was also calculated. The T_M for the $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ sample was determined to be approximately 56.4°C. The T_M for the $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ sample was determined to be approximately 57.70°C.

Comparison of the T_M of the controls and the samples, it seems as nothing occurred. Had damage occurred to the DNA, the T_M plots would show differences. The following four figures demonstrate the similarities between obtained UV-Vis spectra on both the control and the sample groups. Furthermore, had more trials been obtained, a more clear similarity in T_M would be even more apparent.

[2] It should be noted that all DNA strands are assumed to be duplexed with its complimentary strand unless otherwise stated.

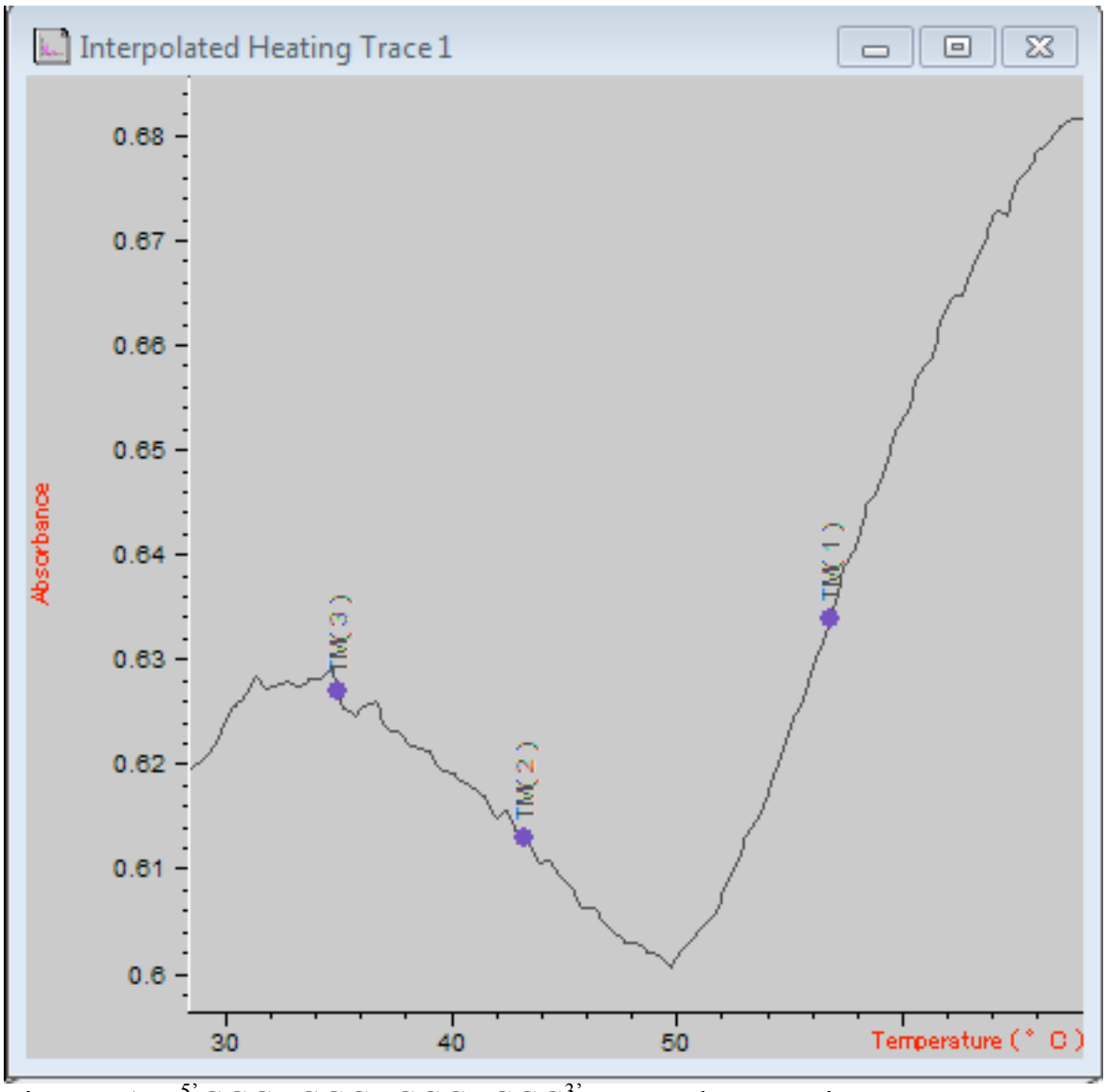


Figure 1. $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ control UV-Vis spectra as obtained August 5th. This graph has a slight strange occurrence with absorbance increasing and decreasing before reaching the true T_M. Causes for this are unknown currently.

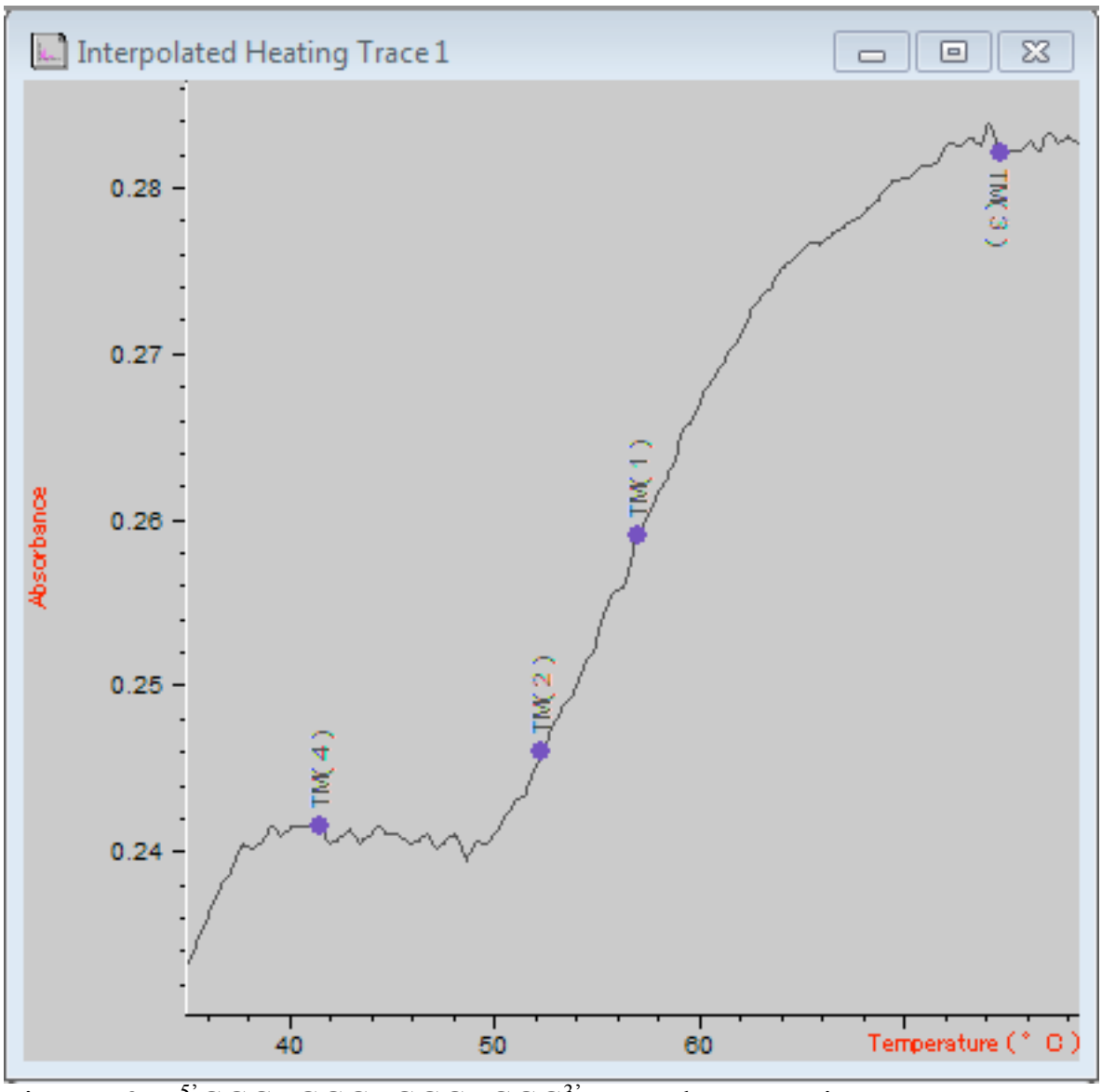


Figure 2. $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ sample UV-Vis spectra as obtained August 7th. This graph has the same sigmoidal shape as apparent in Figure 1. It begins at 50°C and ends near 70°C as does the first graph.

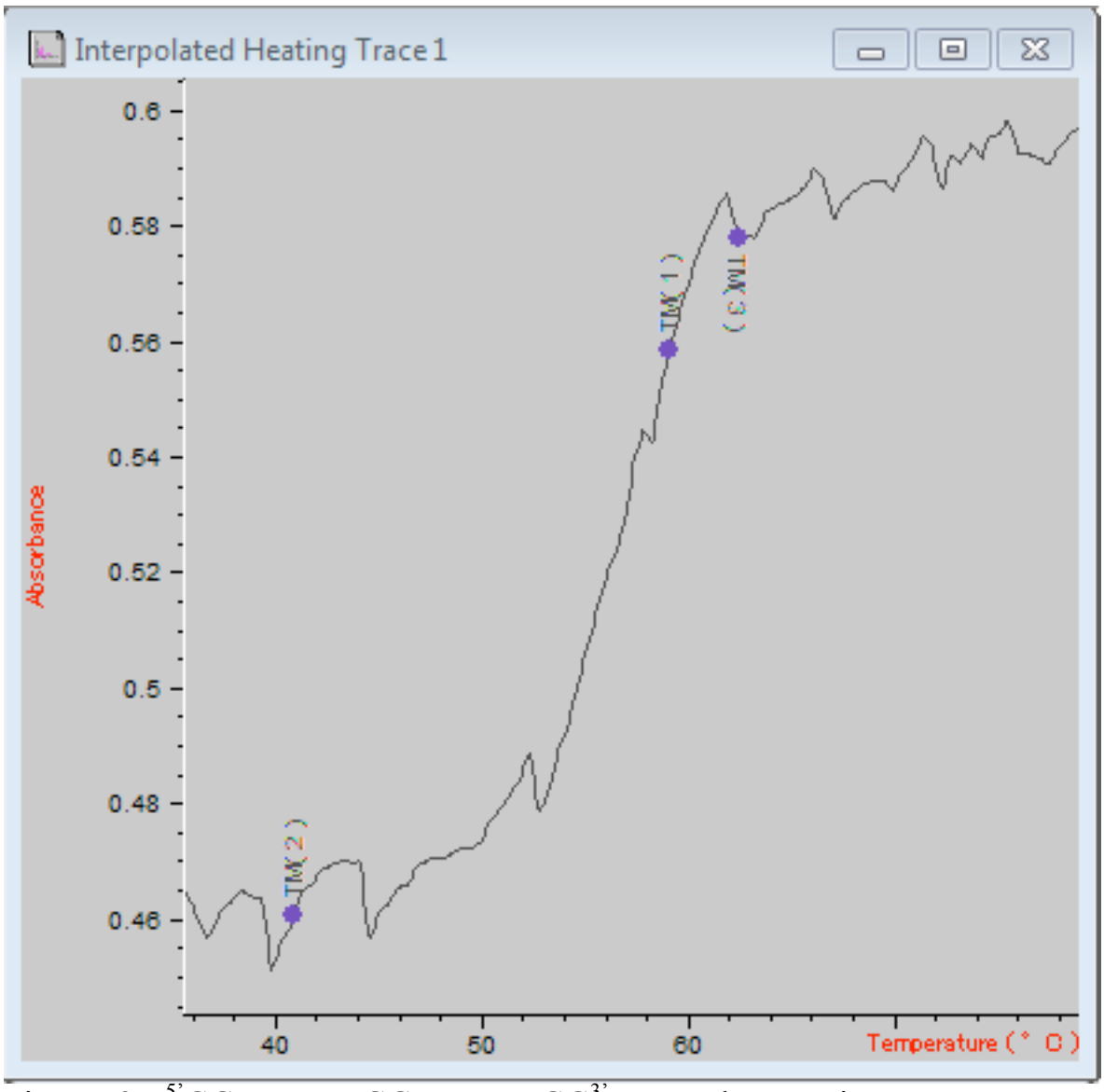


Figure 3. $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ control UV-Vis spectra as obtained August 13th. The T_M algorithm seems slightly mistaken by the inflection point on this graph and its true T_M should be near 57.5°C.

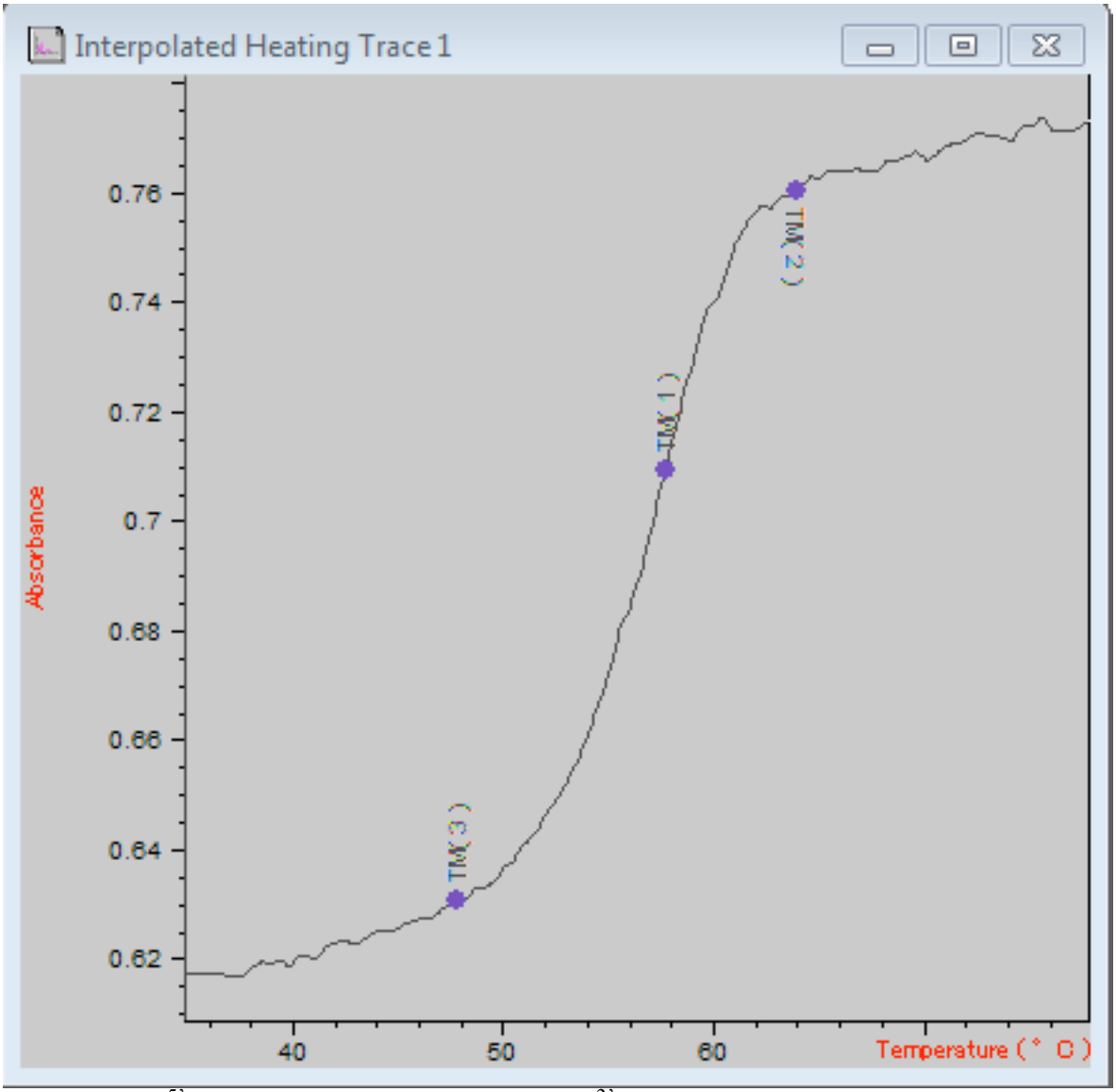


Figure 4. $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ control UV-Vis spectra as obtained August 12th. As with the other graphs, this graph shares the same sigmoidal shape begging around 40°C, much lower than Figures 1 and 2 due to the lesser composition of guanine and cytosine. The shape ends at the same region that Figure 3 does at 62°C.

Viewing the four spectra above, similarities between controls and samples are present in all spectra. Though the T_M seems to be at different temperatures, the same shape of graph occurs at all the same regions. Figure 1 and Figure 2 have the most differences due to varying concentrations of sample and control, which is present in all spectra, and their different temperature values. The first control was run at a temperature range much

lower than all the other samples, thus it has a different starting and ending temperature.

Along with the plots of the absorbance vs. temperature, the gel electrophoresis provides similar conclusions. Had fragmentation occurred along the flight as hypothesized, the gel would show fragmentation along the samples' movement pattern in the gel. As it shows, however, no fragmentation can be observed. Furthermore, samples and controls are completely undistinguishable from each other.

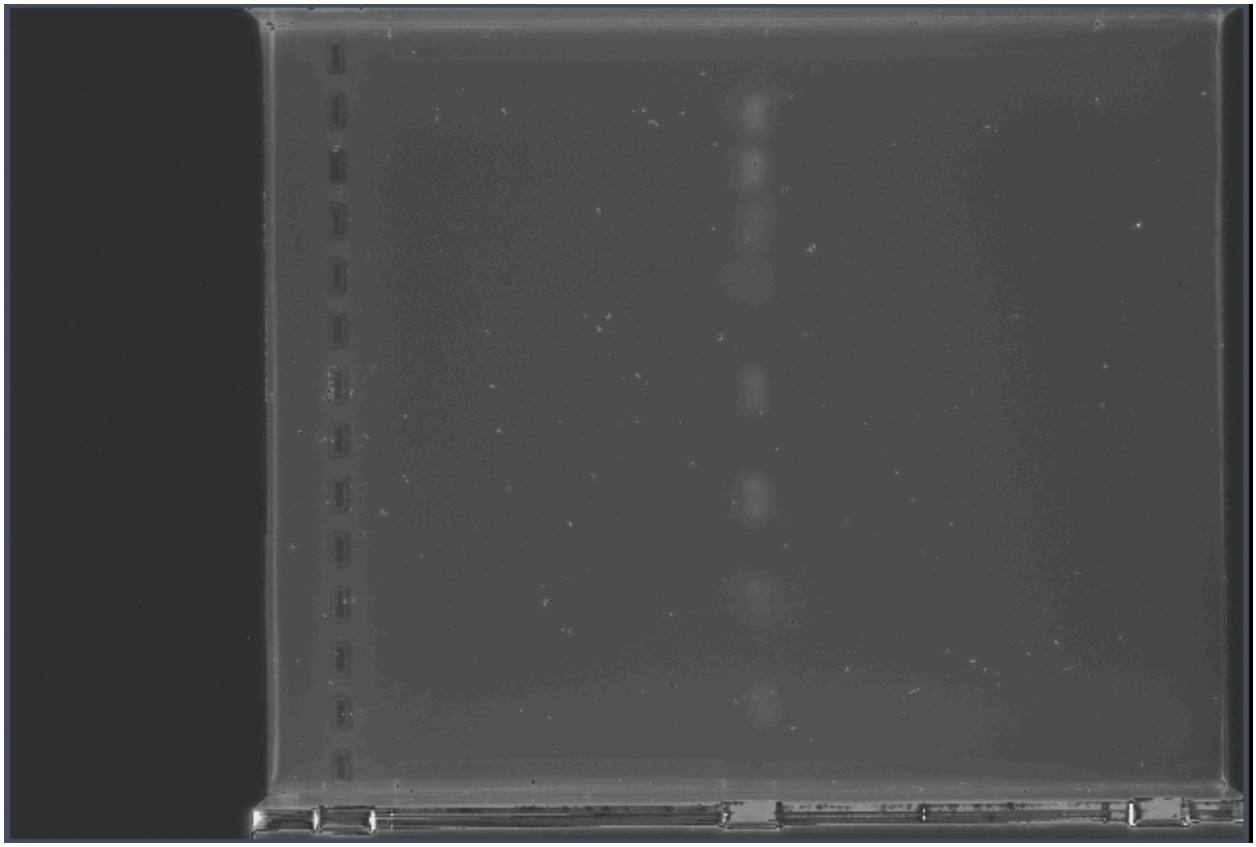


Figure 5. Above is an exposure of a gel electrophoresis performed on August 7th. The marks along the gel are the movement of the controls and samples of both the $^{5'}$GGGTGGGTGGGTGGG$^{3'}$ and the $^{5'}$GGTTTTTTGGTTTTTTGG$^{3'}$ strands.

Using both the UV-Vis spectroscopy and the gel electrophoresis, it is safe to assume that no effects from radiation occurred on the DNA. This in itself is interesting in that the radiation shielding provided, though mostly

unintentional, was adequate enough to properly protect the DNA from upper atmospheric radiation.

Further test are being performed on the QDs, and, as such, no results can be made for them currently.

10.0 Ready for Flight

The payload landed with no structural damage to the internal skeleton, so the box can be used again. While all of the electronics made it through the flight undamaged and can be used again, our team discovered that data was only recorded for the first 10 or so minutes. We are still not positive if it was an electronics problem or a coding problem. The problem has been replicated in the lab multiple times with the same results, but we are having difficulty pinpointing the error, because we have also been able to successfully run the program as intended multiple times. As soon as we figure out the issue in the data logging, the payload would be ready to fly again with the current structure and electronic layout.

There are no special precautions that need to be taken to ensure the payload would be ready to fly again, however our team would probably want to make a new exterior just to ensure that the insulation is as tight as can be.

11.0 Conclusions and Lessons Learned

During this mission, our team has learned how to analyze small scale molecules, which encompass DNA and quantum dots. This analysis also provided us with a knowledge of how to work the instruments needed to test and analyze the samples of DNA and quantum dots. Our team also learned how to set up and run a 3-D printer, which allowed us to create a frame for our payload. Part of the team was able to learn how to produce the hardware and software needed to run the necessary electronics for our payload, such as the Geiger counter. In order to make this mission more successful, our team should look into creating a different set up for the DNA and quantum dot samples in order to have the maximum amount of radiation hitting the samples during the flight. Also, we should look at ways to use less mass in the future, especially in the electronic and structural areas of the payload. If this mission were to be done again, we could research different methods for gaining data on the DNA and quantum dots in order to get a wider range of data and a more precise look at the effects of high altitude radiation on the small scale molecules.

12.0 Message to Next Year

I hope that you are well. When you are beginning your process, please overestimate the amount of time that it will take to accomplish your goals. The estimated time for completion is always way less than it actually is. If you can, add a factor of 3 to whatever your estimates are. Above all, have fun, enjoy the process and appreciate the fine opportunity that our faculty, Colorado Space Grant and UNC have provided us with.