Biological Particles (Bacteria and Fungi) in Thunderstorms

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Motivation - Why Study Biogenic Particles?

- <0.1% of cells carried into atmosphere survive (DasSarma, 2018)
- Diseases and airborne illnesses can be carried into the atmosphere via airborne biogenic particles (Burrows et al., 2009).
- Seasonal variations in bacteria and particle concentrations, and local events (e.g. dust storms) lead to bacteria isolates (Harbizadeh, 2019).
- Cloud and climate impacts (aid in heterogeneous freezing of cloud droplets).
- Most studies focus lower portions of troposphere.
- Challenges collecting airborne measurements.

Introduction - Aerosol Particles

- Aerosol Particles range from variety of natural and man made materials
- Fine particles are Aitkin range, have condensation and coagulation of primary particles and chain aggregates, and accumulation range.
- Forms particles into droplets via homogeneous nucleation or condensation growth of nuclei (Whitby, 1977)

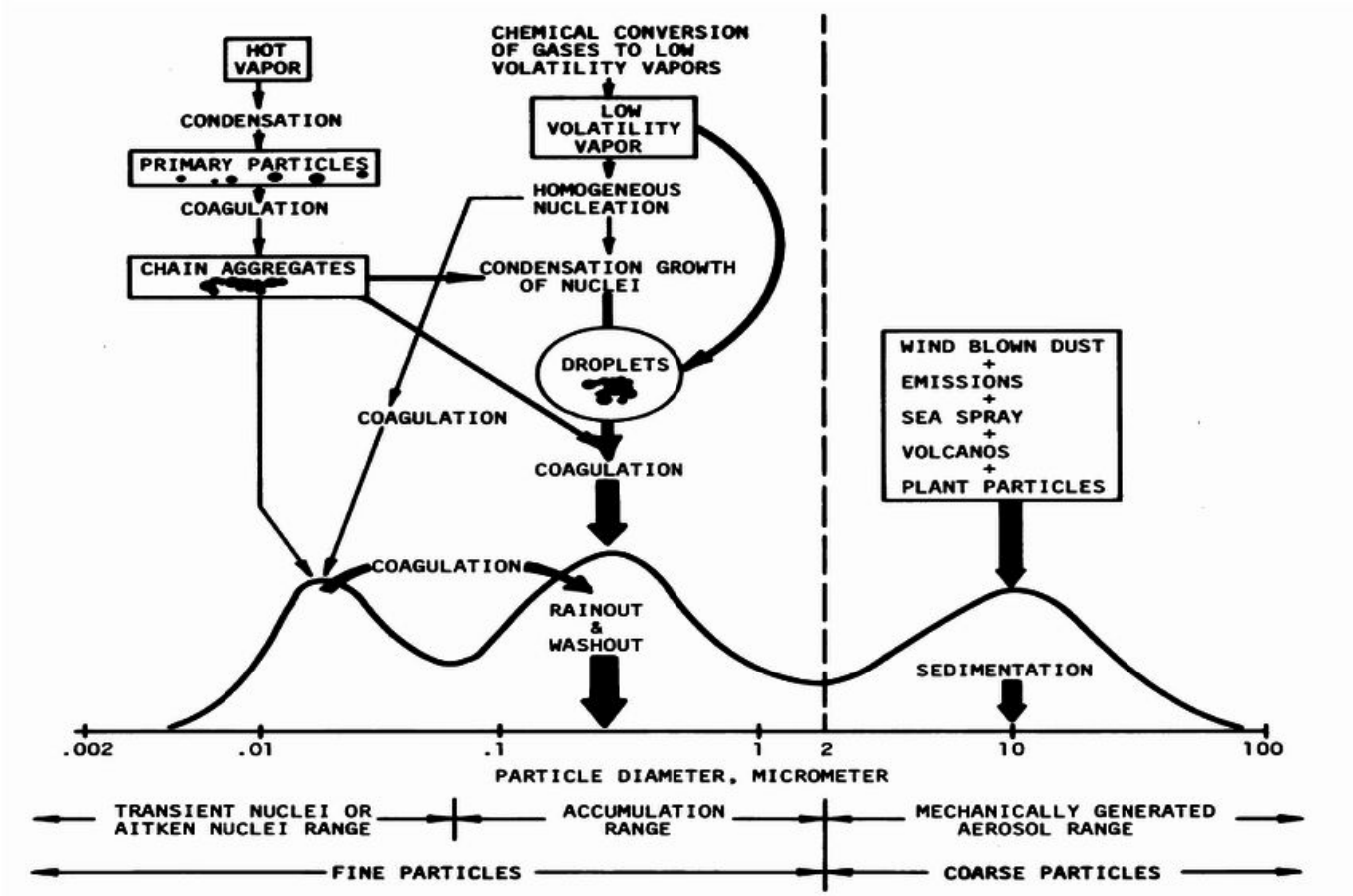
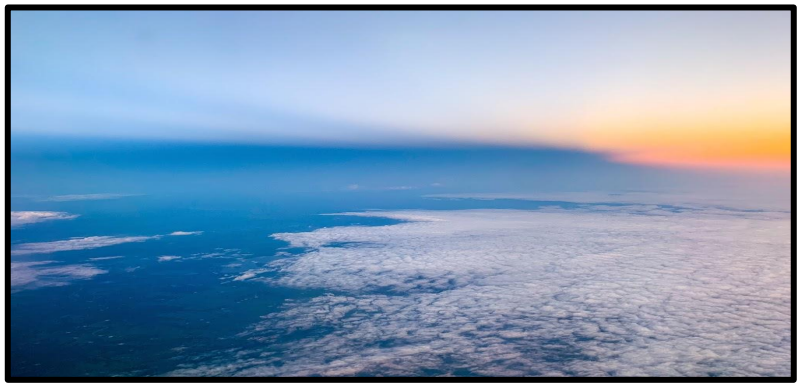


Figure 1: Idealized schematic of the distribution of particle surface area of an atmospheric aerosol (Whitby, 1977).

Introduction - Biogenic Particles

- Biological particles act (dead or alive) as Ice Nuclei Particles (INP) at temperatures less than -10 °C, sometimes activate up to -4 °C (Matthias-Maser et al., 2000).
- Bacteria and fungi - proven to be good INP, found in atmospheric fog, clouds, and rainwater (residency time of several days).
- Atmospheric waters can transport bacteria via lifting from surfaces and mix vertically (Hu et al., 2018).



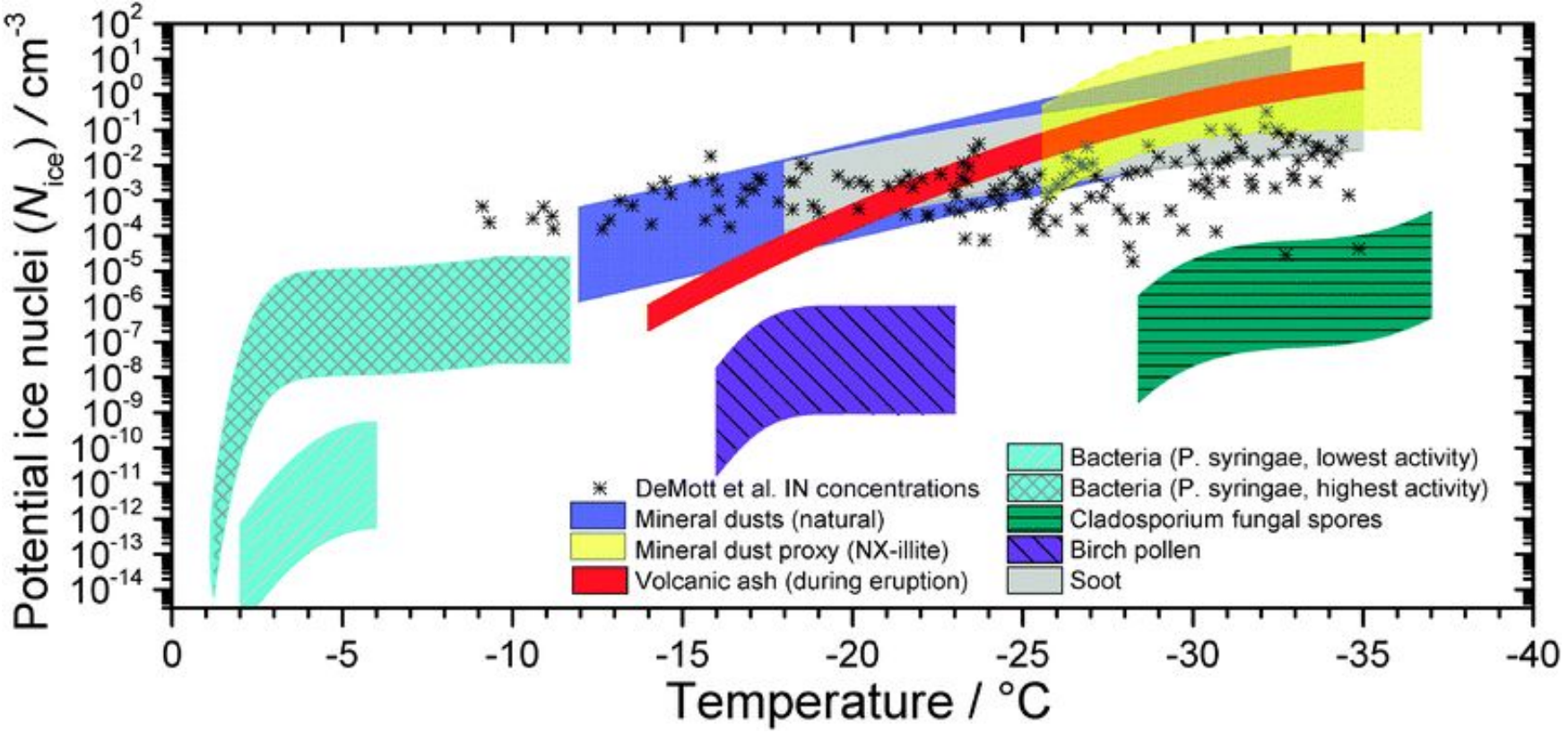


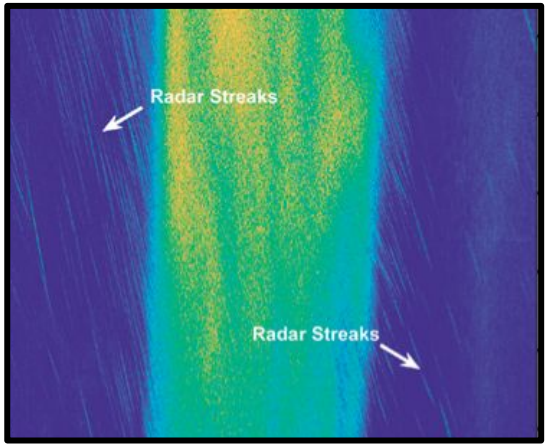
Figure 2: Potential immersion modes of different ice nuclei concentrations as a function of temperature for different atmospheric species (Murray et al., 2012).

Experiment Objectives

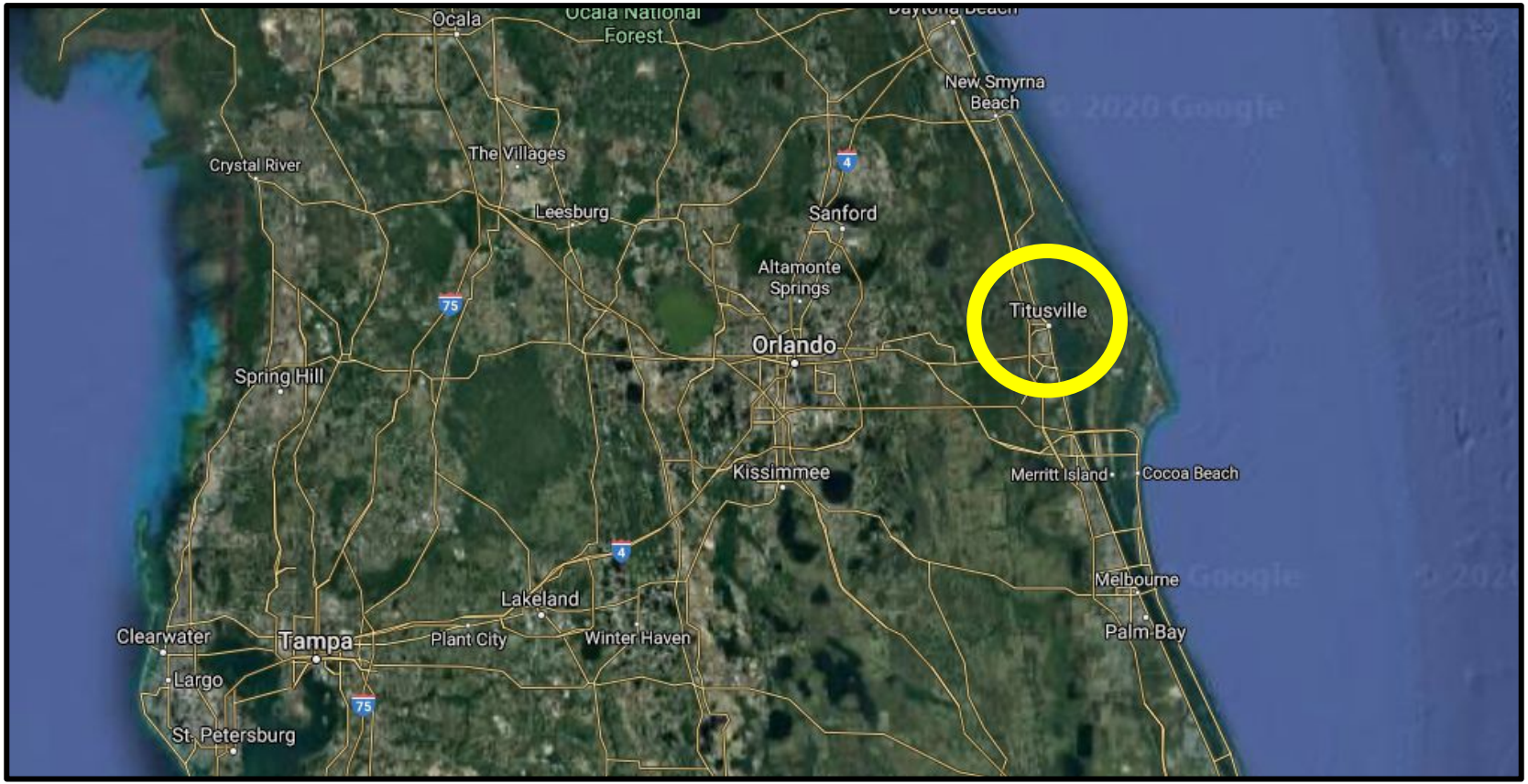
- Determine if we can sample biological particles in the anvils of thunderstorms.
- Use DNA sequencing to determine the kinds of bacteria and fungi sampled, and identify their origins by relating their types to surface locations.

CapeEx19 Field Campaign

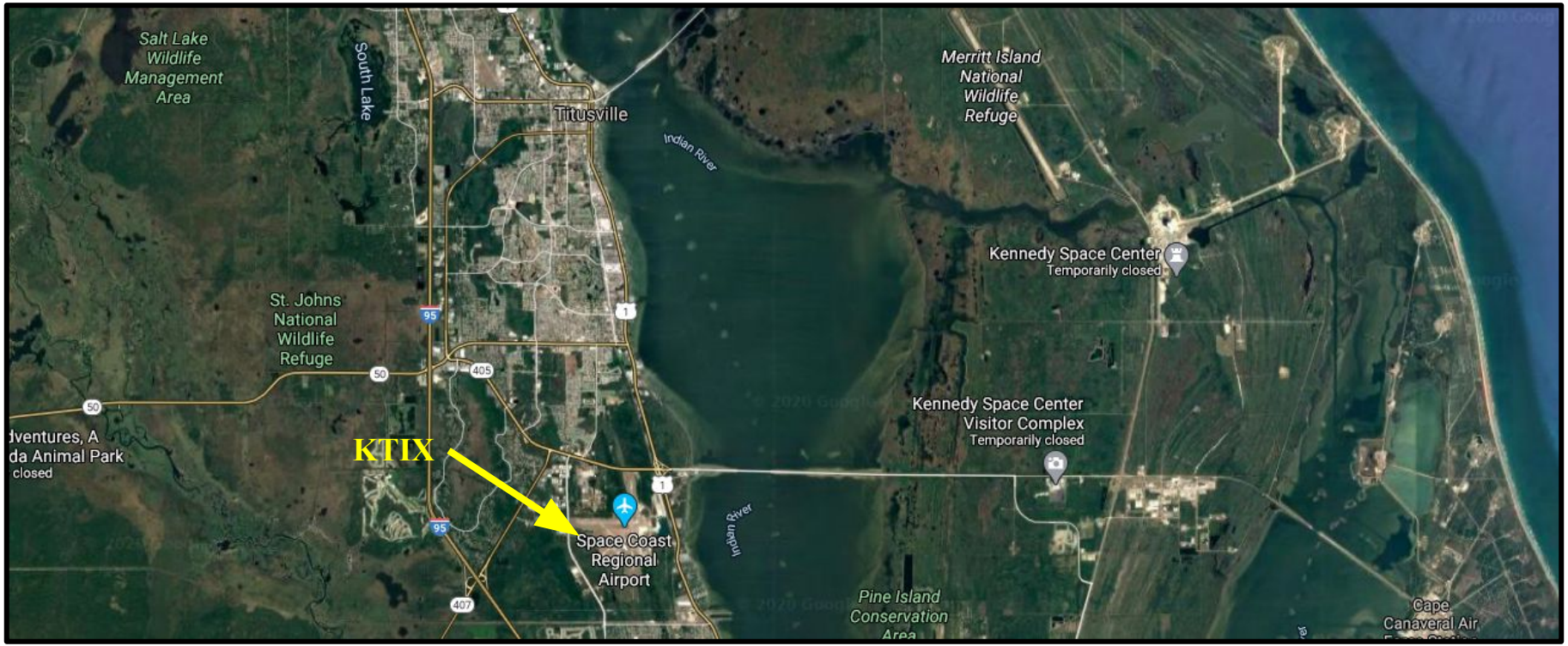
- Titusville, FL July 22nd - August 3rd, 2019.
- Grant from Naval Surface Weather Center to improve understanding of thunderstorms.
- Combines aircraft measurements, observations with US Navy's Mid Course Doppler Radar (MCR) to develop better cloud models.
- Biological particles experiment a side project of the main campaign.



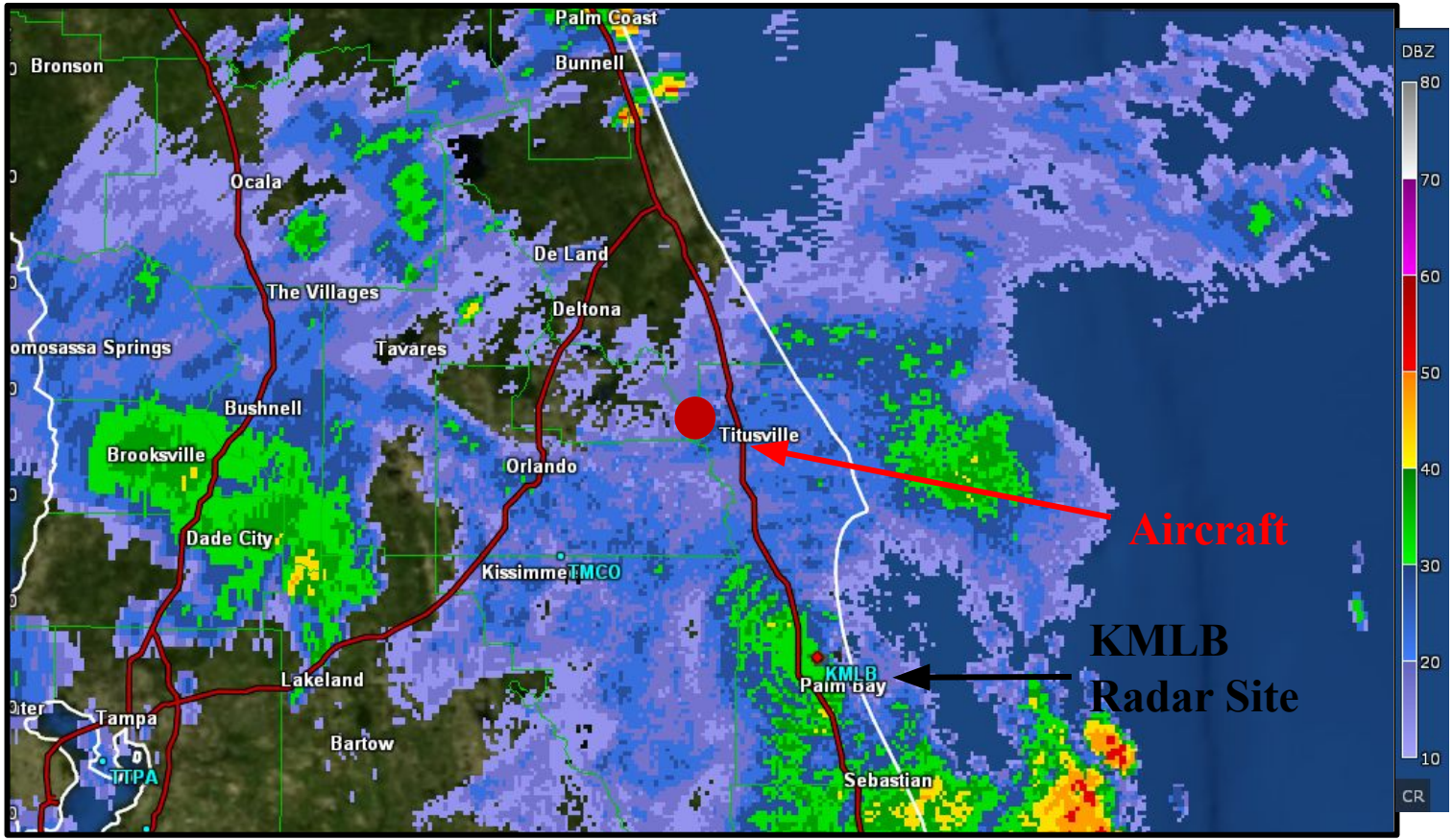
Titusville, FL



Space Coast Regional Airport (KTIX)

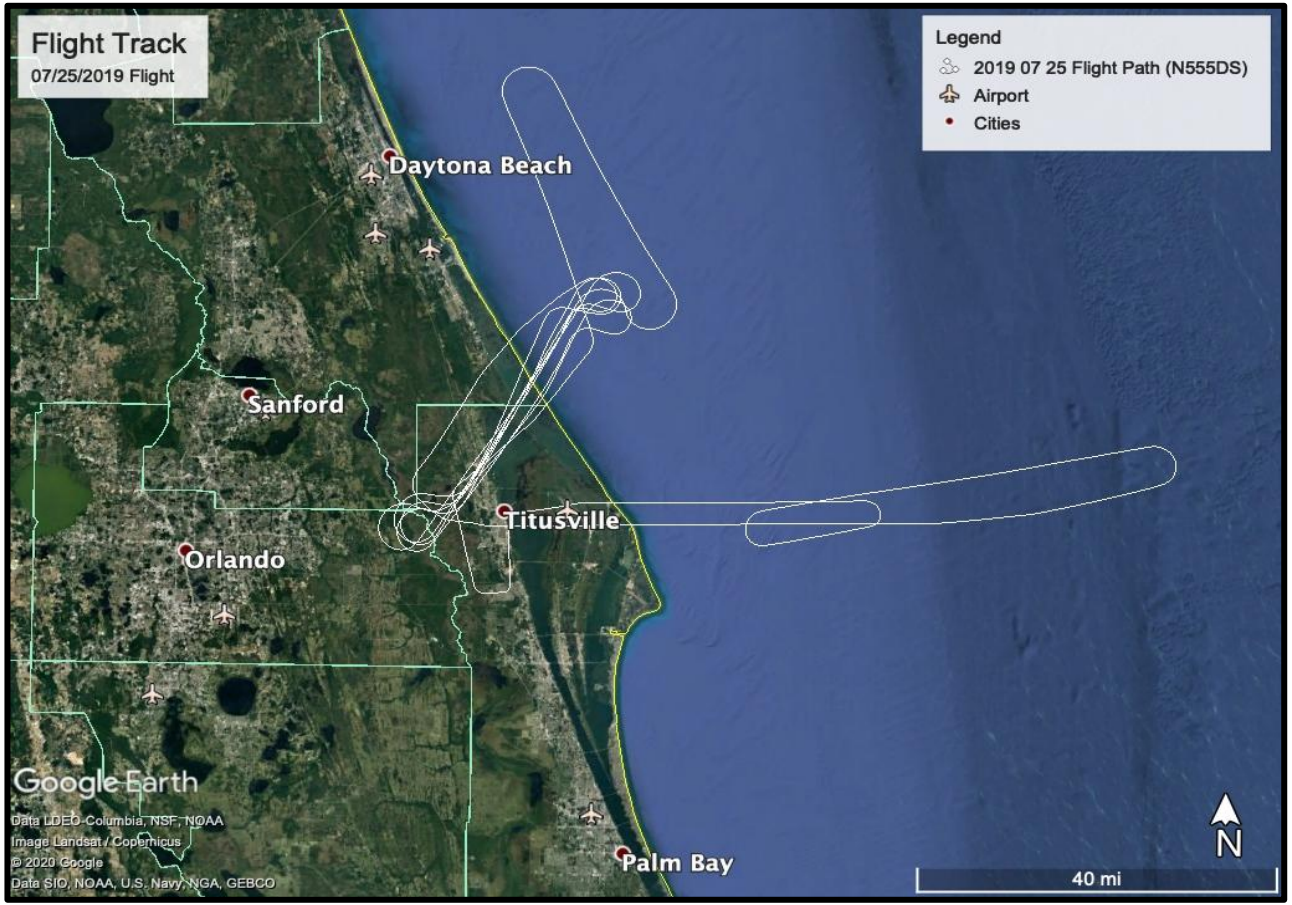


Melbourne Radar (KMLB) - 7/25/19 @18:54:46 UTC



Onboard Citation Research Aircraft

Flight Track Example (7/25/19)



- Takeoff: 18:02:52 UTC
- Landed: 20:14:40 UTC

Methods - Aircraft Instrumentation

- North Dakota Citation Research Jet participated in CapeEx19 Field Campaign in Titusville, FL summer 2019
- Instrument rack held filter holder tied into aircraft's inlet.
- Priority of good airflow, sampling control valves, and prevention of decompression due to pressure changes.

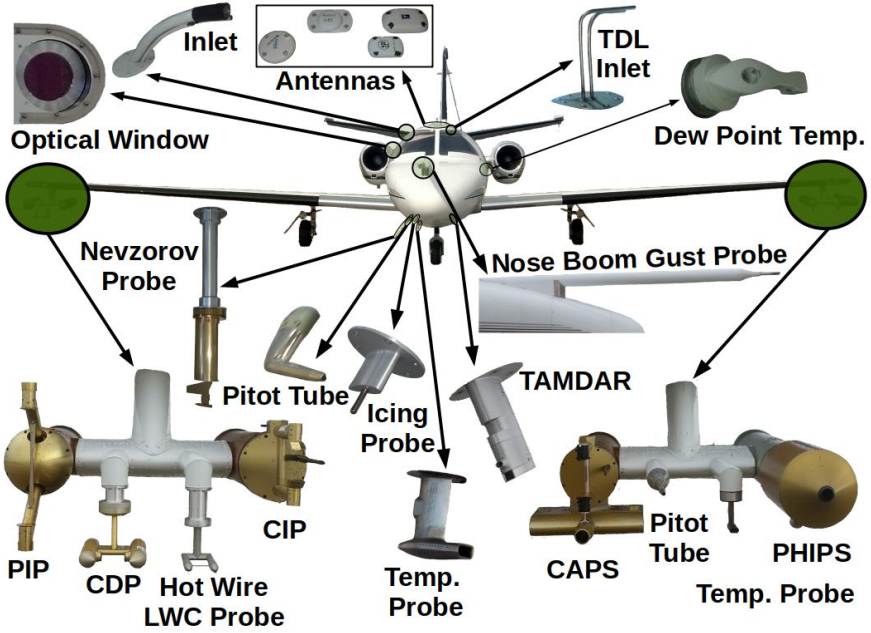
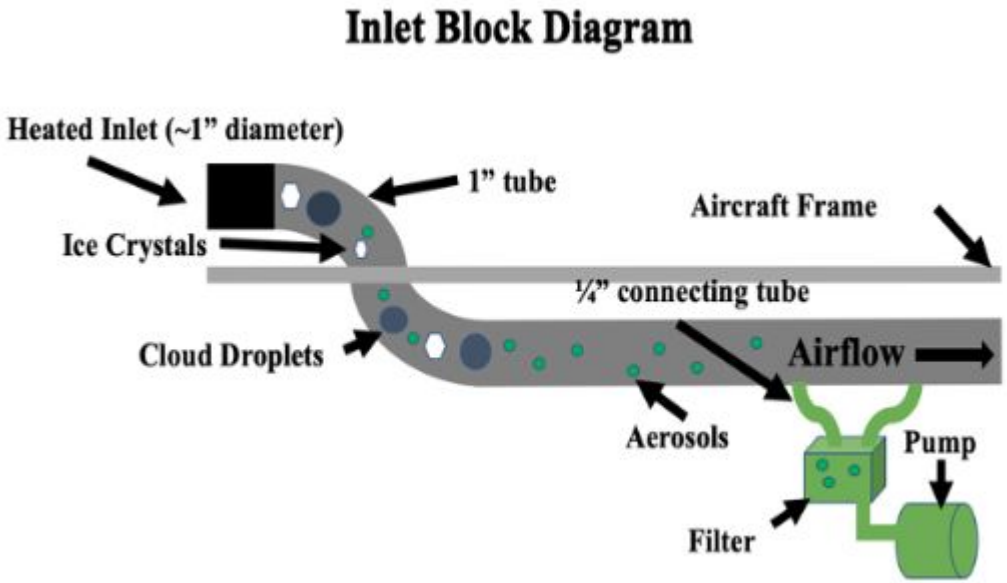


Figure 3: CPC inlet is labeled as "heated inlet".

Aircraft Filter System



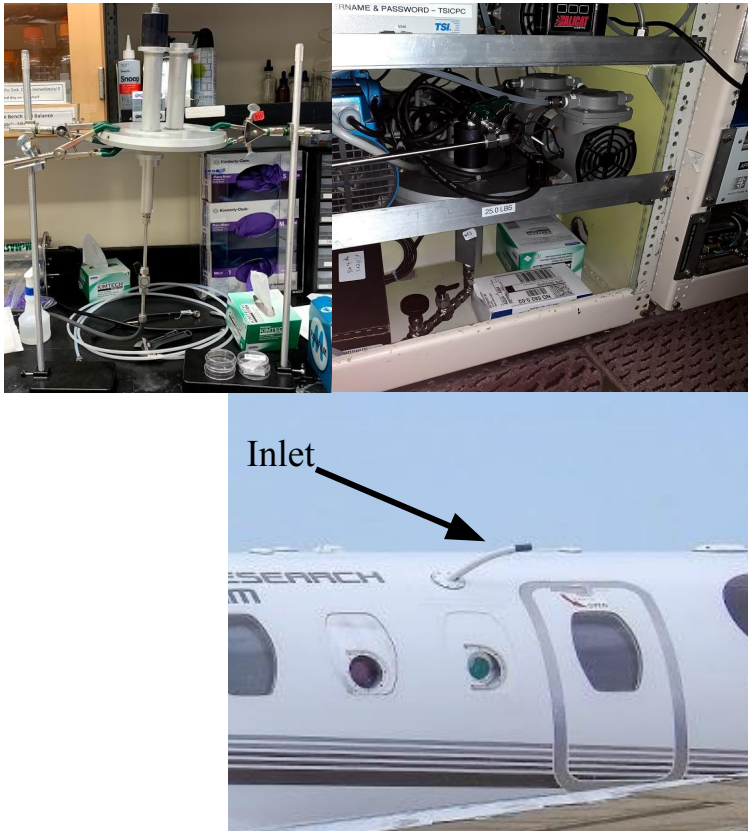
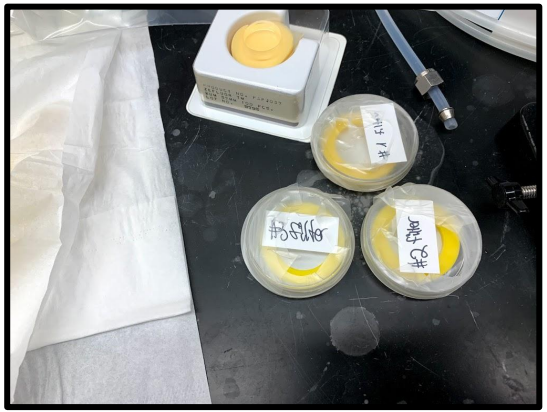


Figure 4: Diagram of filter inlet inside the plane. Real pictures on the right.

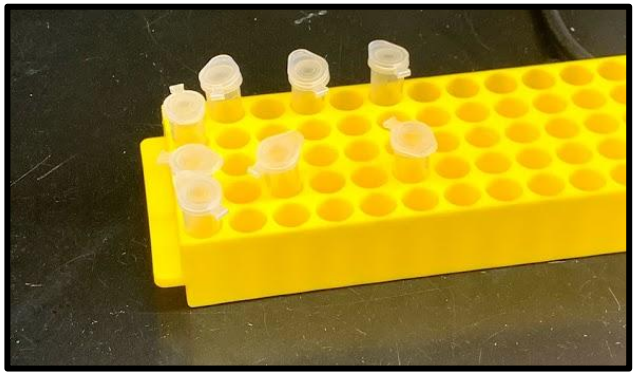
Methods - Pre Experiment Prep

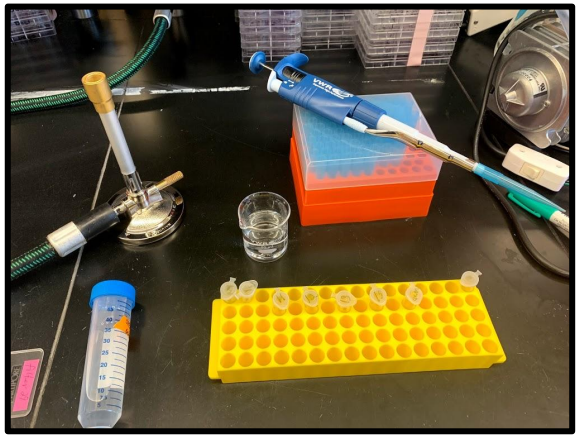
- 37mm tissuquartz filters 0.45 μm pores were sterilized from contaminants via autoclaving (121 °C for 45 min.).
- Placed inside filter holders, sealed in containers.
- Before each flight, filter was taken from container and placed inside filter holder (done carefully with clean gloves and 98% ethanol-based alcohol).
- Filter valves opened when ready for sampling, closed when done
- After landing, filter removed from aircraft, placed into incubation jar



Methods - Post Experiment Prep

- Filters cut up carefully, half of it placed into 2 ml RNA/DNA free tubes with 1 ml of double distilled water. Other half preserved.
- Bunsen burner used to kill outside contaminants.
- Tubes cleaned with ultrasonic bath for 60 min to remove particles from the filter into the water.
- Tubes then stored in -80°C freezer until sequencing via ribosomal internal transcribed spacer sequence ITS for fungi, 16S ribosomal RNA-encoding DNA for bacteria





Overview of Results

- 15 filters used in CapeEx19.
- 10 filters (4 blanks and 6 in flight) were processed for bacteria and fungi sequencing.
- Only 5 filters were completely sequenced.
- Sequencing calculates highest top 10 bacteria/fungi types with highest of concentrations (1000's of bacterias).
    - Clean Filter (background/control test)
    - Filter 11 (Blank Flight) (8/2/19)
    - Filter 12 (Anvil TS40) (8/2/19)
    - Filter 13 (Anvil TS31) (8/3/19)
    - Filter 15 (Multi-Cloud TS38) (8/3/19)

Data Set (Blank Tests)

Blank Filter Tests

Filter	Date	Event	On	Off	Total	Temp Range	RH Range	Pressure Range
#	M/D/Y	Type	UTC	UTC	hrs	°C	%	hPa
Filter 5*	7/28/19	Ground	13:13	16:13	3.0	28 to 31	74 to 91	1020 to 1021
Filter 8*	7/30-7/31/19	Ground	20:19	12:44	15.95	24 to 28	82 to 96	1017 to 1019
Filter 9*	7/31/19	Air	16:30	20:55	5.4	-30 to 30	50 to 105	1000 to 350
Filter 11	8/2/19	Air	13:05	16:17	3.2	N/A	N/A	N/A

Blank Flight Filter

*Filter was cut for testing, bacteria data has not yet been retrieved.

Data Set (Flight Tests)

Flight Filter Tests

Filter	Date	Event	Start	Stop	Total	Temp Range	RH Range	Pressure Range
#	M/D/Y	Type	UTC	UTC	hrs	°C	%	hPa
Filter 1*	7/16/19	Cumulus	00:09	00:26	0.3	-7 to -27	75 to 370	538 to 360
Filter 6*	7/29/19	In Cloud	20:41	21:06	0.4	-46.5 to -54.2	N/A	238 to 206
Filter 10*	8/1/19	High Anvils	20:27	20:46	0.4	-17.5 to -20.5	87 to 109	393 to 377
Filter 12	8/2/19	Anvil	20:09	22:03	2.1	-33 to -57	45 to 160	309 to 286
Filter 13	8/3/19	Anvil	15:26	16:26	1.0	~ -31	25 to 115	300 to 287
Filter 15	8/3/19	Multi-Cloud	21:21	22:46	1.5	-31 to -30	33 to 55	288 to 208

Anvil TS40

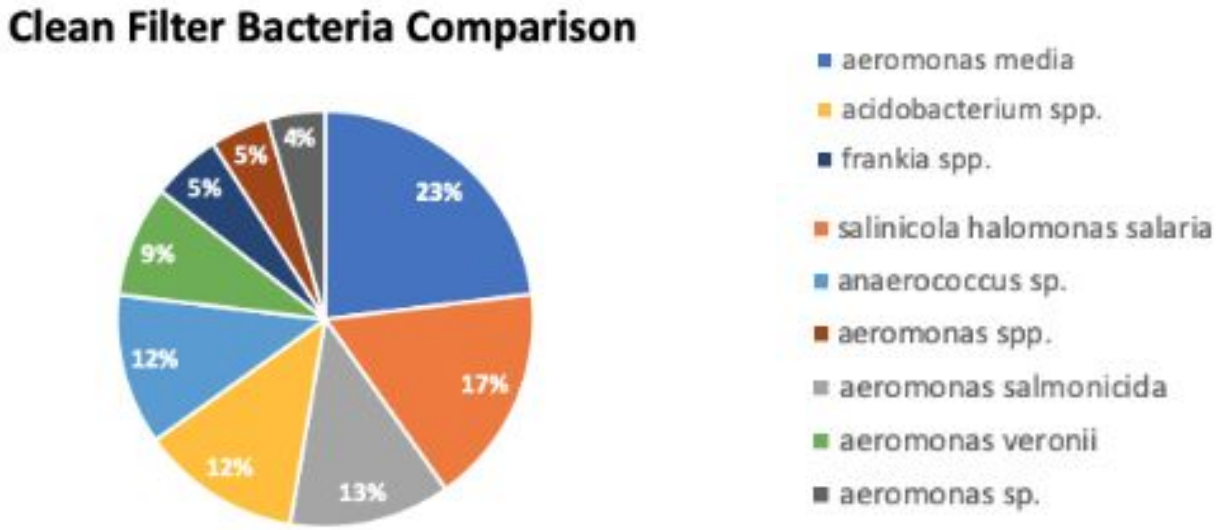
Anvil TS31

Multi-Cloud TS38

*Filter was cut for testing, bacteria data has not yet been retrieved.

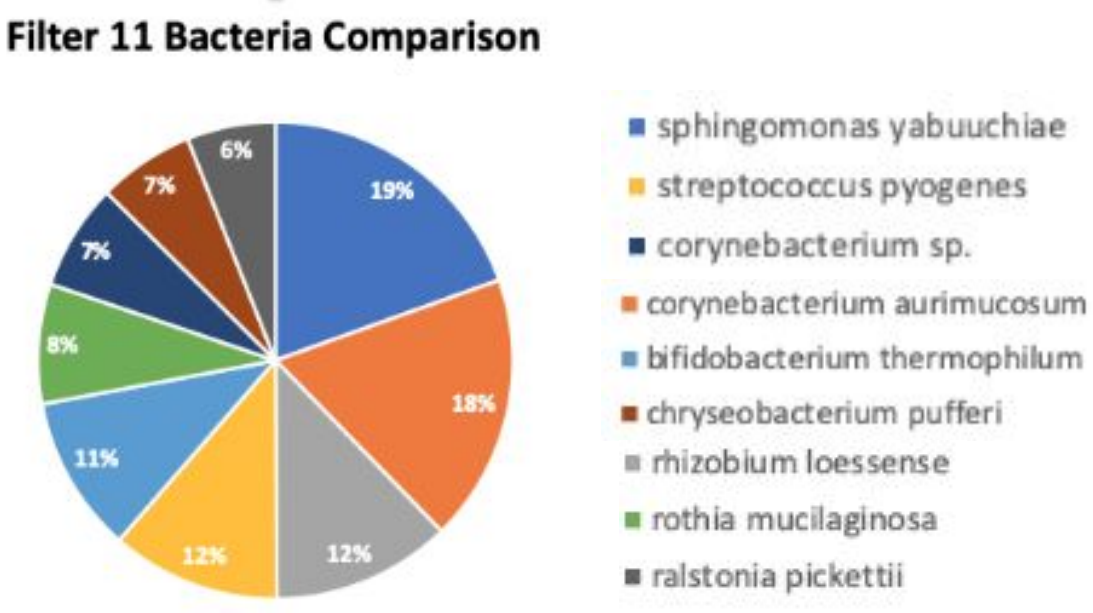
Clean Filter Bacteria Analysis

- Aeromonas media (23%) - not widely researched or known (found on Filter 15).
- Salinicola halomonas (17%) - first isolated from salt water sample.
- Acidobacteria (12%) - found in soils.



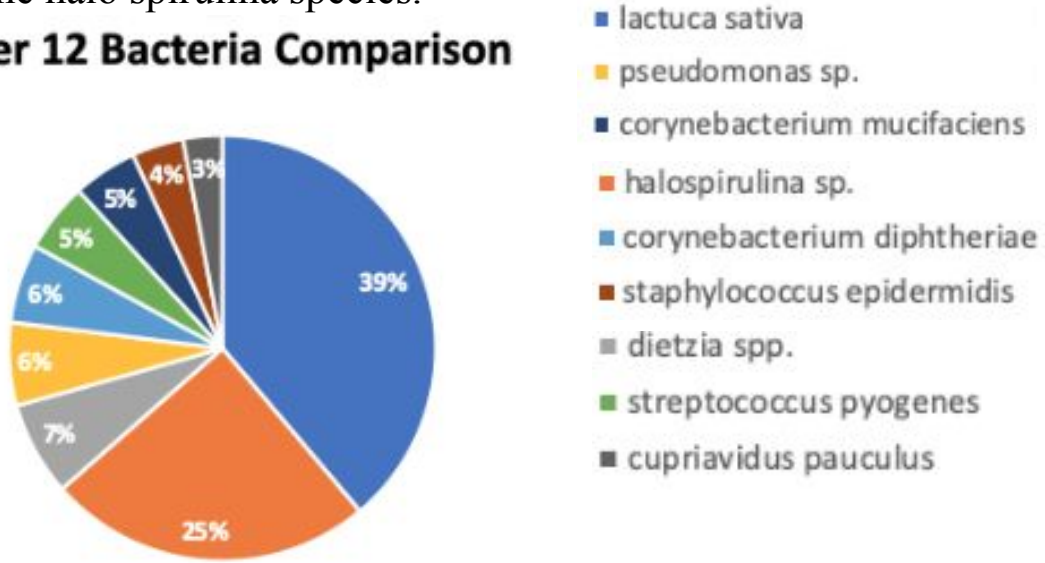
Blank Flight (Filter 11) Bacteria Analysis

- Large amounts of sphingomonas yabucchiae (19%), strictly aerobic bacteria, play a role in treatable human infections.
- Corynebacterium aurimucosum (18%), rothia mucilaginosa (8%) linked to human diseases.
- Ralstonia pickettii (6%) found in moist environments, linked to rare human diseases.



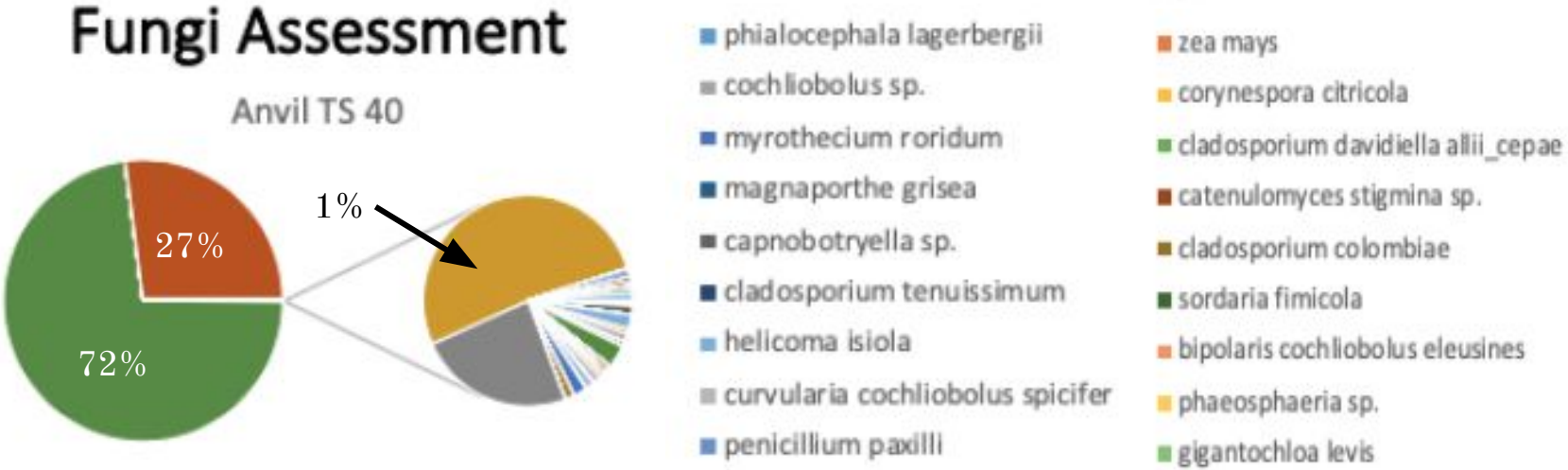
Anvil TS40 (Filter 12) Bacteria Analysis

- Lactuca Sativa (39%) is likely sourced to lettuce seeds.
- Halospirulina (25%) - salt tolerant, not widely known.
- Dietzia (7%) - human and animal pathogen.
- Pseudomonas (6%) - good ice nucleating agent, comes from soil and water.
- Other salt-tolerant bacteria from the halo spirulina species.
- Total sample time: 2.1 hrs.

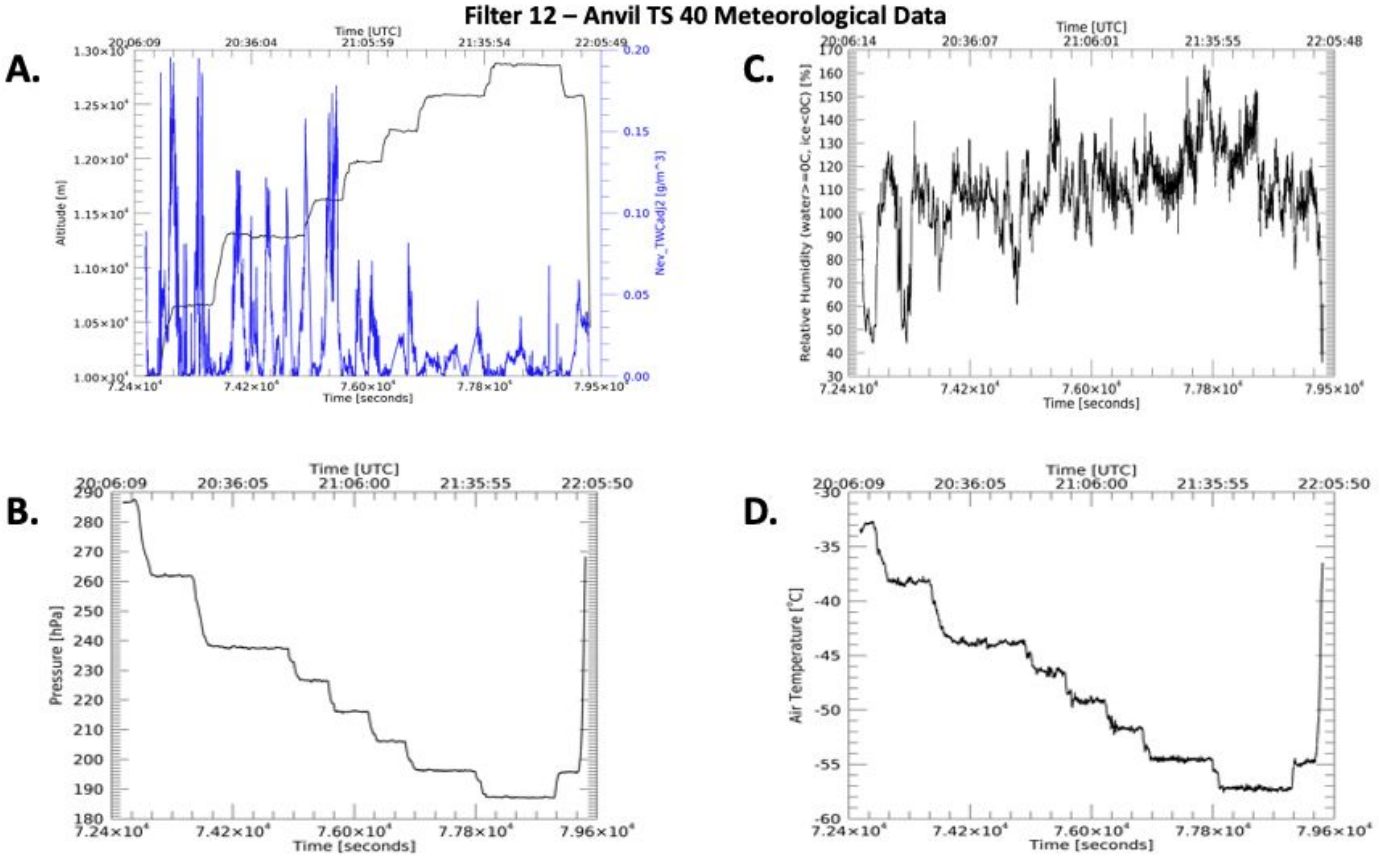


Anvil TS40 Fungi Analysis

- Only filter with significant amounts of fungi.
- Resupinatus trichotis (72%) - known to grow on underside of decaying wood.
- Pichia Kudriavzevii (27%) - found in soil on outside of fruits (McDonald, 2015).



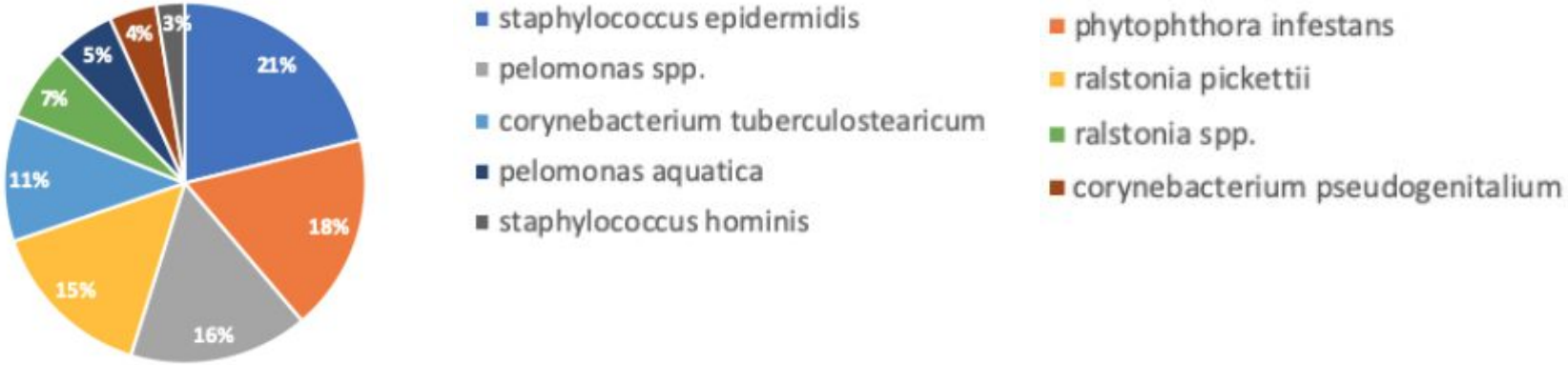
Anvil TS40 Aircraft Data (20:09Z to 22:03Z)



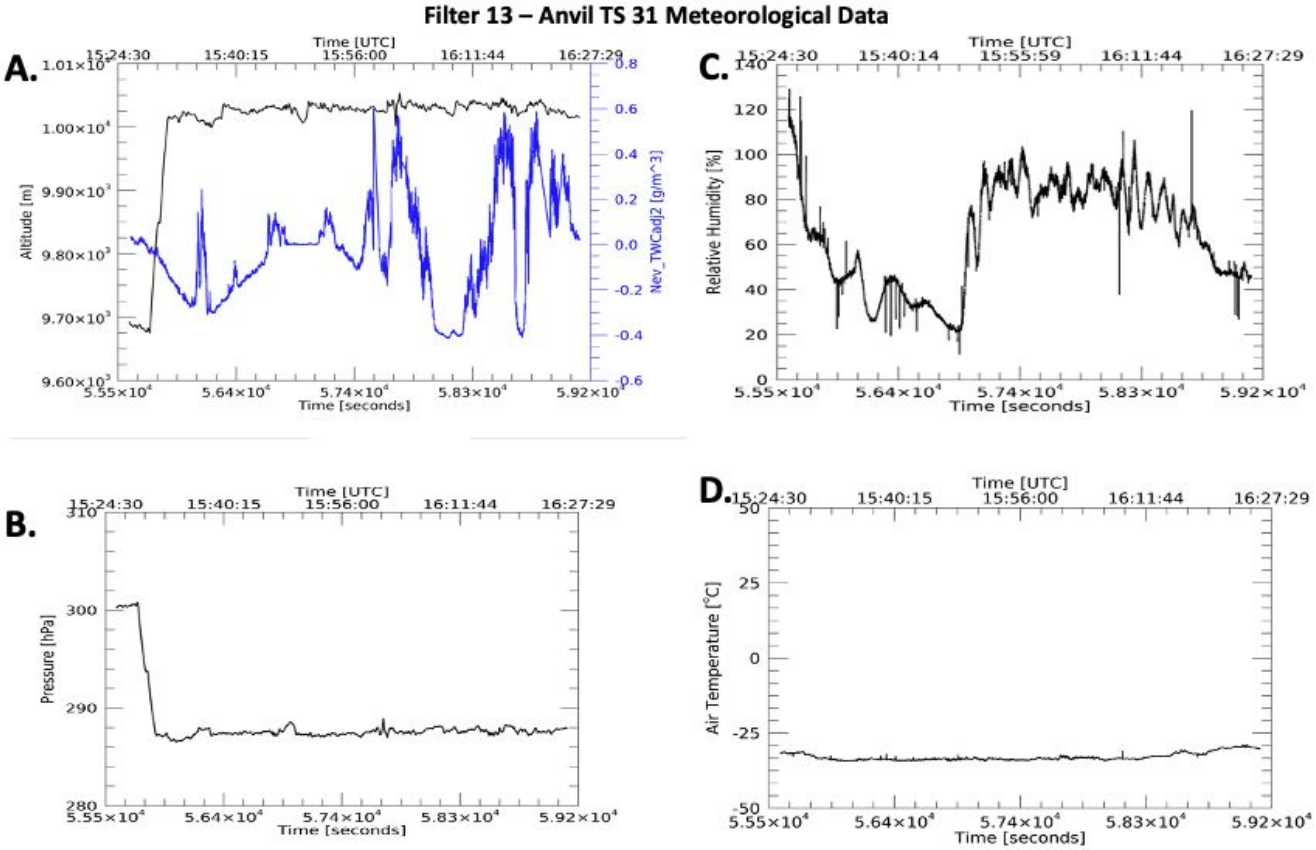
Anvil TS31 (Filter 13) Bacteria Analysis

- Phytophthora (18%) - plant pathogenic fungus, causes potato & tomato disease.
- Staphylococcus epidermidis (21%) - normal human flora from skin.
- Pelomonas (16%) and ralstonia pickettii (15%) are natural water sourced bacteria.
- Total sample time: 1.0 hr.

Filter 13 Bacteria Comparison

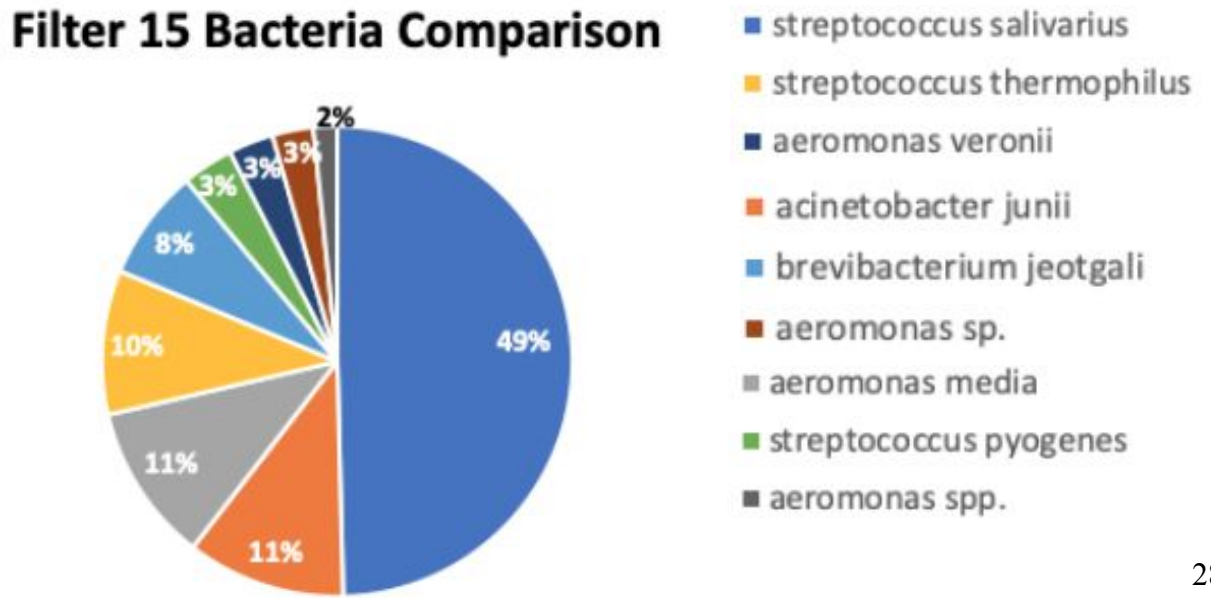


Anvil TS31 Aircraft Data (15:26Z to 16:26Z)

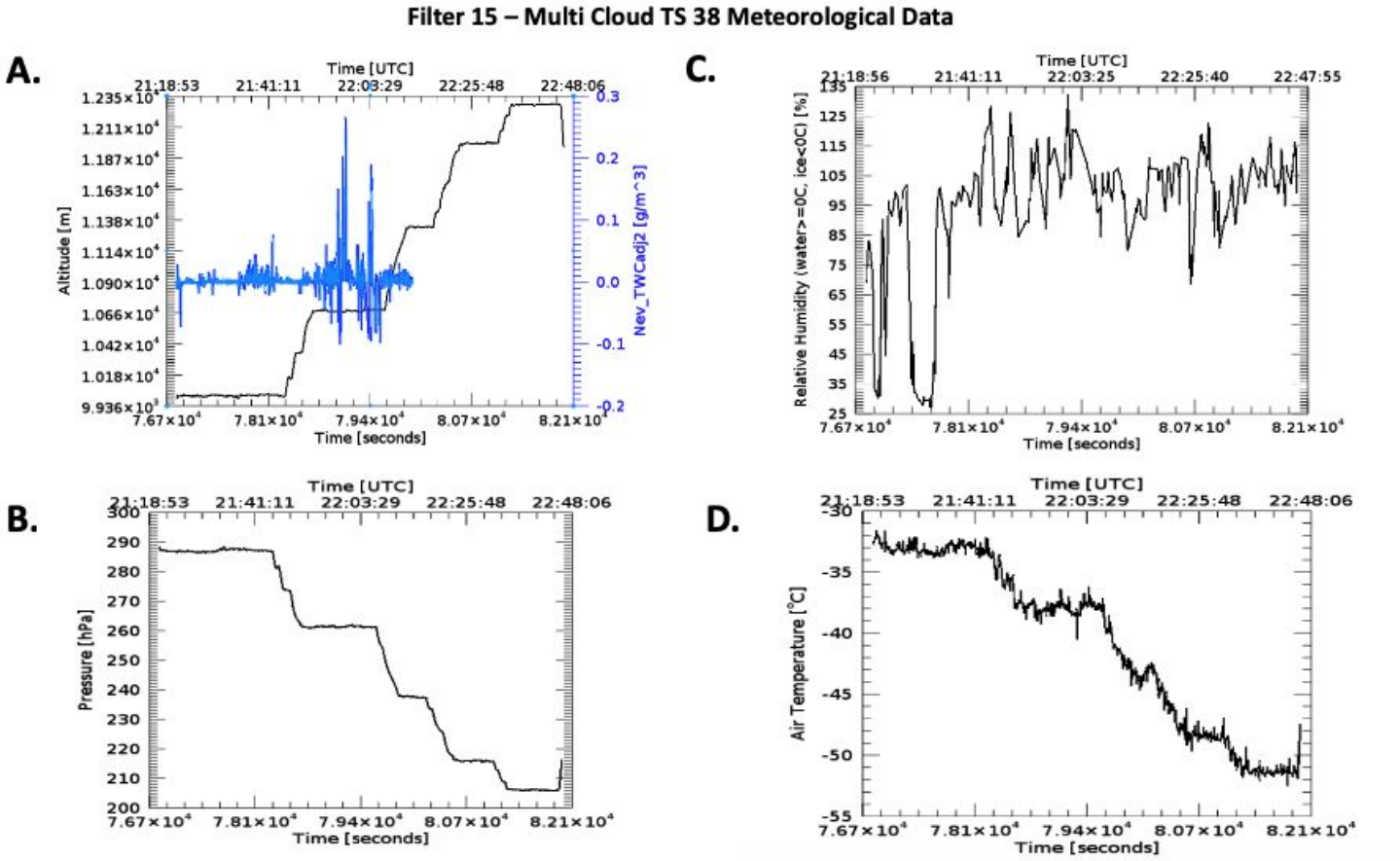


Multi-Cloud TS38 (Filter 15) Bacteria Analysis

- Streptococcus salivarius (49%) - oral and respiratory portions of human body (McDonald, 2015).
- Acinetobacter junii (11%) - linked to nosocomial infections, streptococcus, pyogenes.
- Brevibacterium jeotgali (8%) - related to producing antibiotics.
- Aeromonas (genus) (3%) - ubiquitous in fresh and brackish water.
- Total sample time: 1.5 hrs.



Multi-Cloud TS38 Aircraft Data (15:26Z to 16:26Z)



Summary of Results

- Anvil TS40 had the most variety of species and was the only filter with fungi.
- Many species pertained to natural plants found in the Florida region.
- Salt-tolerant bacterias likely came from ocean waters.
- Pseudomonas Syringae found, most well known INP bacteria.
- Higher amounts of human-related bacteria found on Multi-Cloud TS38, likely due to human contamination.

Conclusions

- Challenges collecting data still present with human-related bacteria present.
- Best ice nucleating particles come from soils, and waters.
  - Can play huge role in microphysical processes in atmospheric waters.
- Particles can be carried up into atmosphere and dispersed miles away, spreading diseases.
- Photosynthetic particles can have similar climate impacts to greenhouse gases.

Future Work

- In addition to filters, implement water sampling techniques.
- Reduce contamination exposures when working with filters - in lab and on plane.
- Sample in different types of clouds (cirrus, stratocumulus, etc.).
- Compare with different altitudes between high level and low level samples.

Acknowledgements

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- Dr. David Delene and his students for help installing filter rack and retrieving data from aircraft instruments.
- The UND MARC U-STAR program for funding and research support.

Questions?

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