



# A STUDY OF FREEZER AND ROOM CONTAMINANTS

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## INTRODUCTION and ABSTRACT

Previous studies have been conducted over the presence or absence of microbes in accretion ice from the sub-glacial Lake Vostok in Antarctica. This study was to add more evidence in the argument over whether there is or is not life in Lake Vostok. The nature of the research changed once the tube containing the accretion ice cracked. At that time, the focus changed to finding what contaminants are present in the freezer and in the lab.

## METHODS and MATERIALS

1. Prepare media plates
2. Pipette 200 µl of melted sample onto each plate
3. Allow to incubate at 15°C
4. Make note of colonies as they appear
5. Re-plate colonies and perform colony PCR (primers used were ITS-4/ITS-5 for fungus and Eub16s\_383F/Eub16s\_1546R for bacteria)
6. Purify DNA and send it for sequencing
7. BLAST results using NCBI's nucleotide BLAST
8. Do phylogenetics (future)

## RESULTS

Colonies were found on the following days on the following media:

The melt-water was spread on the media on June 11-12, 2009.

6.15.2009	Blood Agar	(Colonies 2, 3)
6.24.2009	Nutrient Agar	(Colonies 5, 6, 7)
	R2A Agar	(Colonies 10, 11, 12)
	Sabouraud Agar	(Colony 13)
6.29.2009	R2A Agar	(Colonies 14, 15, 16, 17, 18, 20)
	Sabouraud Agar	(Colony 22)
7.06.2009	Nutrient Agar	(Colony 25)
	Lauria-Bertani Agar	(Colonies 27, 28)
7.09.2009	Blood Agar	(Colony 29)
	Sabouraud Agar	(Colony 30)
	R2A Agar	(Colonies 31, 32)

## CONCLUSIONS

Although the sample was contaminated, we believe that there are some Lake Vostok microorganisms in the mix, such as Colony 29. It is probable that most of the fast-growing microorganisms are contamination, while the slower-growing microorganisms have a higher chance to actually be from the Lake Vostok accretion ice.

## FUTURE PROSPECTS

The sequences found from this study will be used to make a list of known contaminants from the lab.

Phylogenics will also be done with these sequences.

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### Best BLAST hits:

	<u>% COVERAGE</u>	<u>% IDENTITY</u>	
Colony 2	<i>Cryptococcus diffluens</i>	89%	99%
Colony 3	<i>Brevundimonas sp.</i>	96%	99%
Colony 5	<i>Cryptococcus diffluens</i>	74%	98%
Colony 6	<i>Rhodotorula mucilaginosa</i>	77%	99%
Colony 7	<i>Dermaococcus nishinomiyaensis</i>	96%	98%
Colony 10	<i>Actinomucatales bacterium</i>	97%	99%
Colony 11	<i>Moraxella osloensis</i>	97%	99%
Colony 12	<i>Brevundimonas sp.</i>	95%	98%
Colony 13	<i>Kocuria sp.</i>	95%	99%
Colony 14	<i>Actinomycetales bacterium</i>	97%	99%
Colony 15	<i>Micrococcus luteus</i>	93%	99%
Colony 16	<i>Actinomycetales bacterium</i>	97%	99%
Colony 17	<i>Rhodotorula mucilaginosa</i>	68%	100%
Coloby 18	<i>Xanthomonas sp.</i>	96%	99%
Colony 20	<i>Xanthomonas sp.</i>	97%	99%
Colony 22	<i>Rhodotorula mucilaginosa</i>	96%	99%
Colony 25	<i>Actinomycetales bacterium</i>	90%	90%
Colony 27	<i>Sphingomonas sp.</i>	92%	99%
Colony 28	<i>Sphingomonas sp.</i>	96%	99%
Colony 29	<i>Kocuria sp.</i>	91%	97%
Colony 30	<i>Sphingomonas sp.</i>	96%	99%
Colony 31	<i>Brevundimonas sp.</i>	95%	99%
Colony 32	<i>Dermaococcus nishinomiyaensis</i>	97%	94%

