

The Physical, Chemical & Biological Characterization of Dust from the Middle East: *New Terrors Hiding in the Dust of Iraq?*

U.S. Navy Bureau of Medicine and Surgery
M00– Research Program Integration & Mission
Development

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AMEN!







- *“All substances are poisons; there is none which is not a poison. The right dose differentiates a poison and a remedy”. - Paracelsus (1493-1541)*
- *“And it shall become small dust in all the land of Egypt, and shall be a boil breaking forth [with] blains upon man, and upon beast, throughout all the land of Egypt.
- 6th Plague of Egypt, Exodus 9:10*
- *“Let my armies be the rocks and the trees and the birds in the sky.” - Charlemagne*
- *“Would you tell me, please, which way I ought to go from here?” The cat replied, “That depends a good deal on where you want to get to.” – When Alice met the Cheshire Cat in Alice Through the Looking Glass*

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Military.com

New York Times - August 9, 2005

One More Affliction For Baghdad: A Day Of Blinding Dust And Grit & Breathlessness

By James Glanz

BAGHDAD, Iraq, Aug. 8 - At dawn the atmosphere glowed orange, like the embers of a fire. Objects 25 yards away disappeared, as if a curtain had been drawn in front of them. Baghdad residents began waking up with the sour taste of grit in their mouths and a film of dust on their furniture and clothing, and by 8 a.m. Nireen Abdul Khalek began to feel that she could not breathe. Five hours later, Ms. Khalek, 24,

Research Workgroup Report

Health and Environmental Consequences of the World Trade Center Disaster

Philip J. Landrigan,¹ Paul J. Lioy,² George Thurston,³ Gertrud Berkowitz,¹ L.C. Chen,³ Steven N. Chillrud,⁴

Mice exposed to WTC dust showed only moderate pulmonary inflammation but marked bronchial hyper-reactivity. Evaluation of 10,116 firefighters showed exposure-related increases in cough and bronchial hyper-reactivity..... In summary, environmental exposures after the WTC disaster were associated with significant adverse effects on health.

CENTRAL ASIA: Aral Sea crisis continues to erode health

hankooki.com

THE KOREA

Yellow Dust Storms Harmful to DNA

By Chung Ah-young
Staff Reporter

Dust storms contain materials harmful enough to destroy a cell membrane, a red blood cell and even damage DNA, according to research released Wednesday.



© David Swanson/IRIN

Scores of ships remain stranded in the Aral Sea, once the fourth largest lake in the world

Saving the Aral Sea (IFAS) to address the environmental impact.

Usman Buranov, IFAS' technical director of the Global Environment Facility (GEF) projects, said that the health problems in the region were related to the low quality of drinking water.

He said agriculture and cattle breeding around the sea were less productive, unemployment was climbing and certain diseases were more prevalent.

The polluted air around the sea contained a toxic cocktail of salt, pesticides and chemicals that contaminated drinking water and led to liver and kidney illnesses, as well as a variety of respiratory diseases.

ANKARA, 26 Jul 2006 (IRIN) - Millions of people living near the Aral Sea face a bleak future, with health experts saying diseases like tuberculosis (TB) and cancer are having a terrible impact.

The sea, located on the border of Kazakhstan and Uzbekistan, was once the fourth largest lake in the world. However, it continues to shrink despite regional commitments to halt the draining of the rivers that feed it. It is now a quarter of its original size.

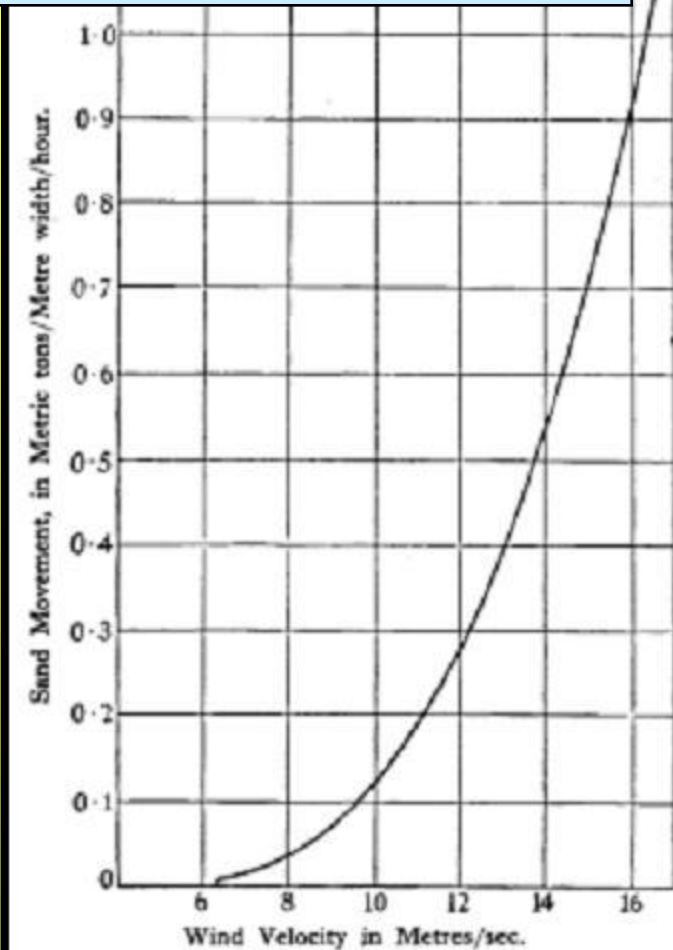
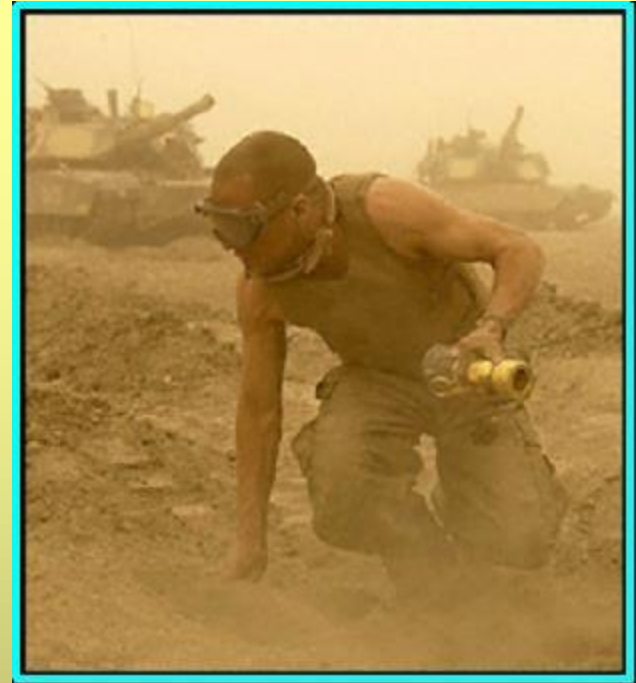
Over the last 40 years an estimated 45 million mt of salt-contaminated dust has been created due to the shrinking, resulting in massive health problems that affect millions of people, experts say.

In 1994, the governments of Kazakhstan, Kyrgyzstan, Tajikistan, Turkmenistan and Uzbekistan established the International Fund for Saving the Aral Sea (IFAS) to address the environmental impact.

Wind Velocity & Airborne Dust

10 m/s = 1968 ft/min = 22 mph

16 m/s = 3149 ft/min = 36 mph

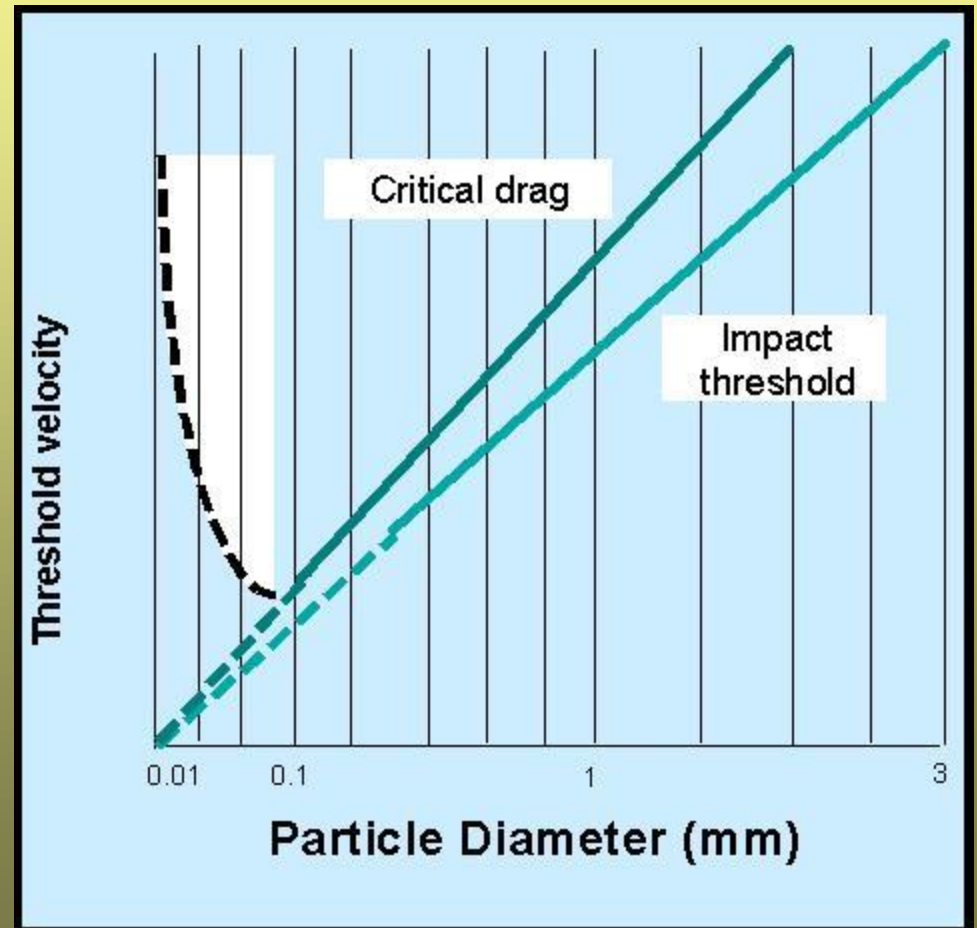


Aerodynamics of Wind Particles

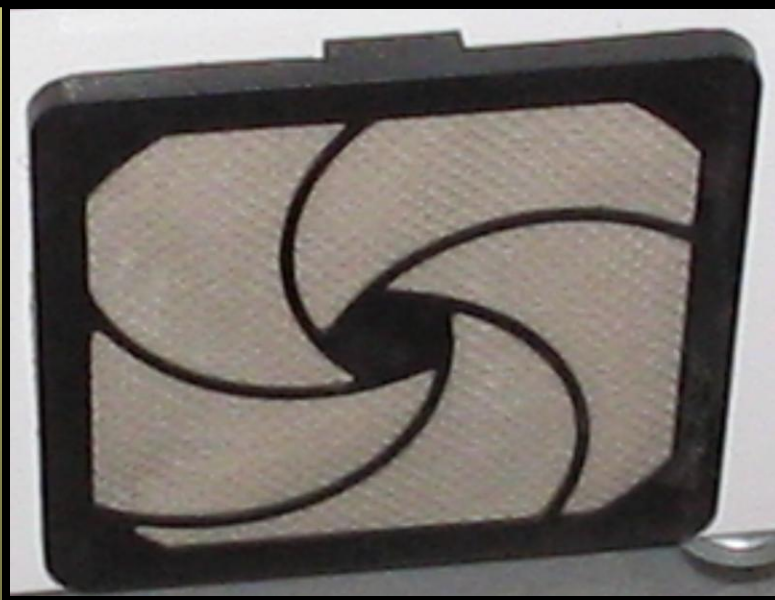
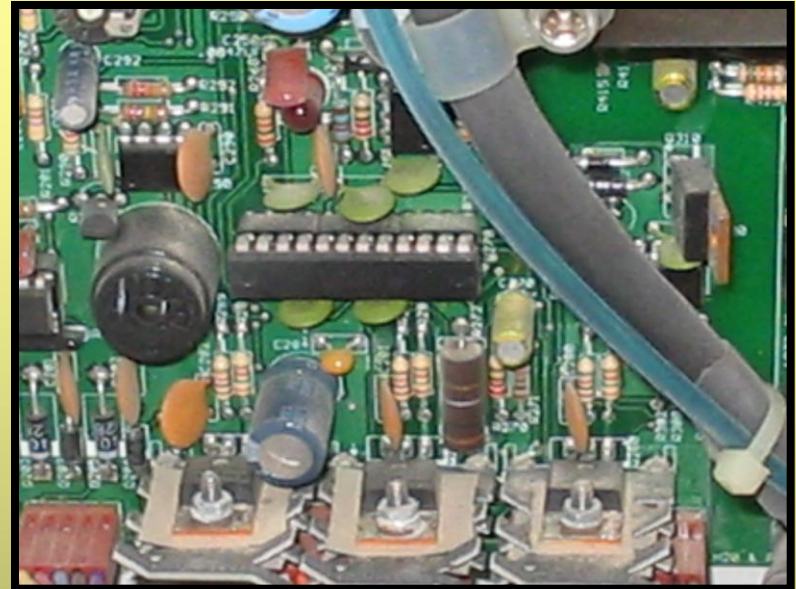
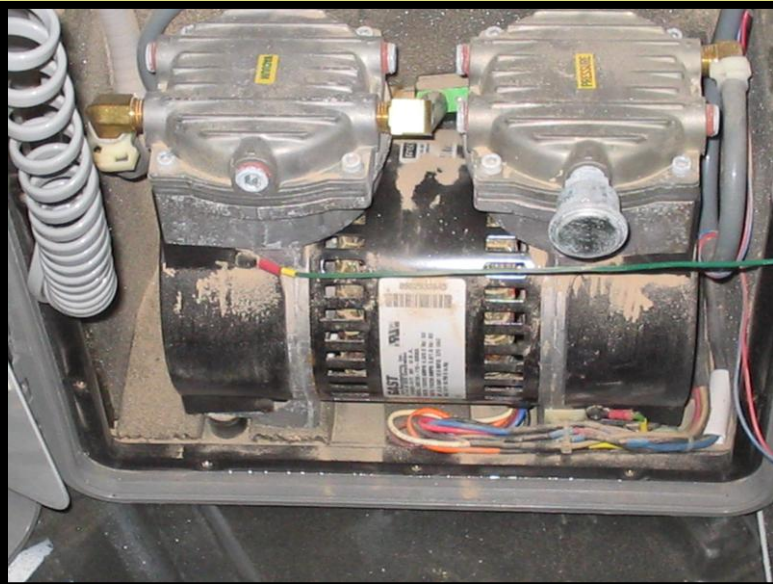
Air flow may be laminar or turbulent, but any near-surface flow is turbulent because of ground roughness.

$$\text{Velocity} = 5.75 V_d \log z/k$$

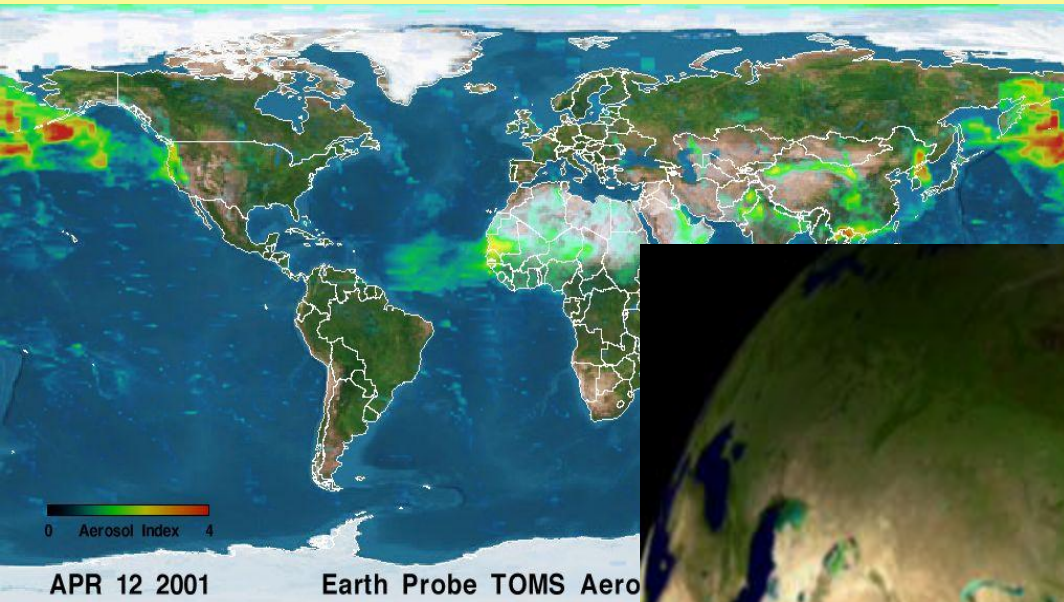
Wind erosion occurs at the instant that a grain is moved. Movement begins when the critical drag velocity is reached for a given size particle.



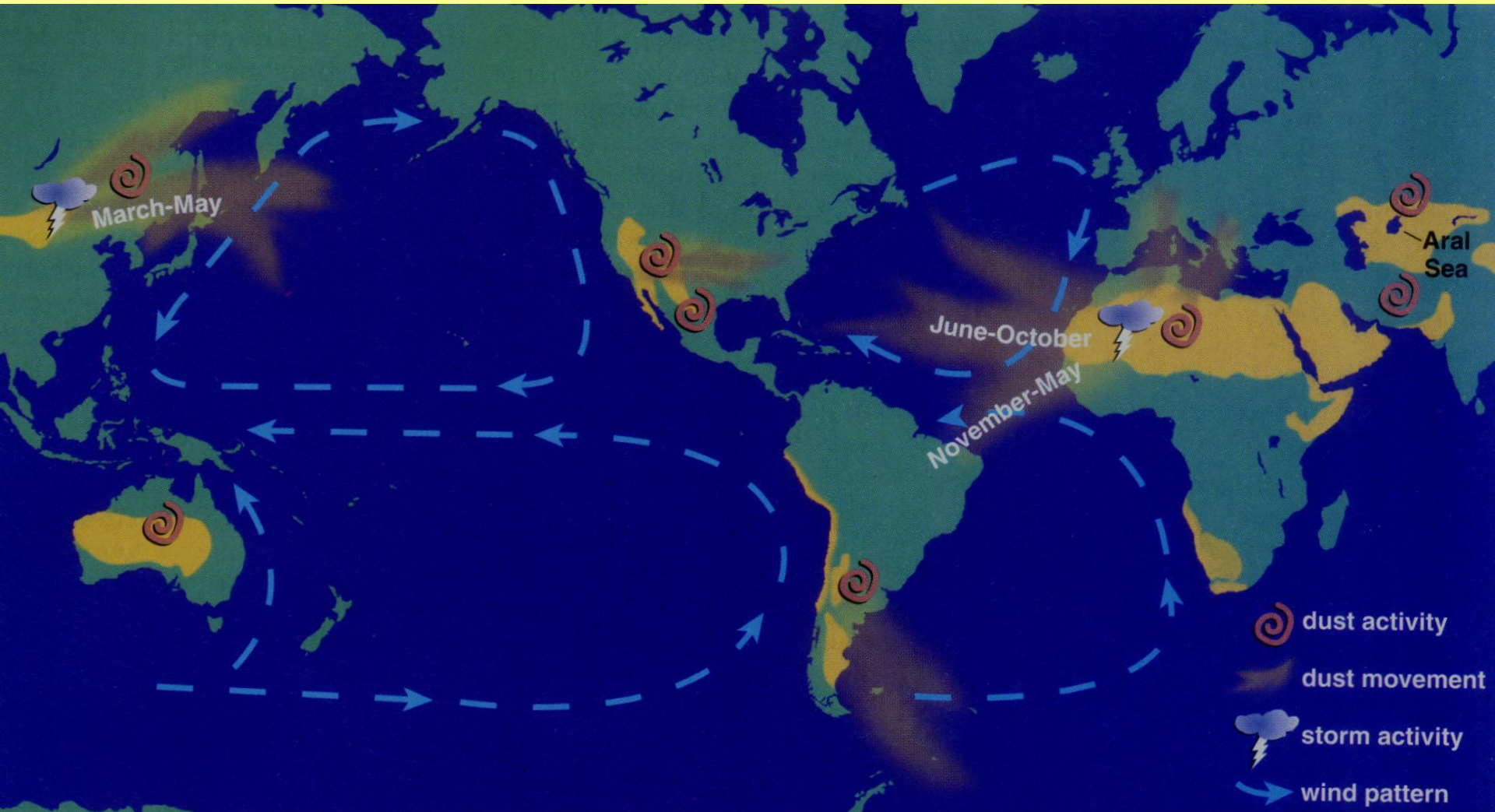
Dust Exposure



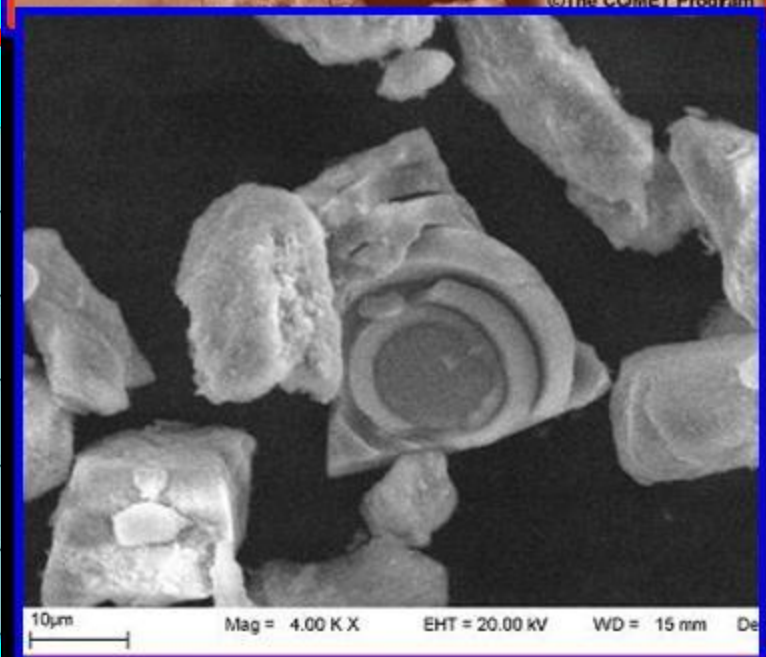
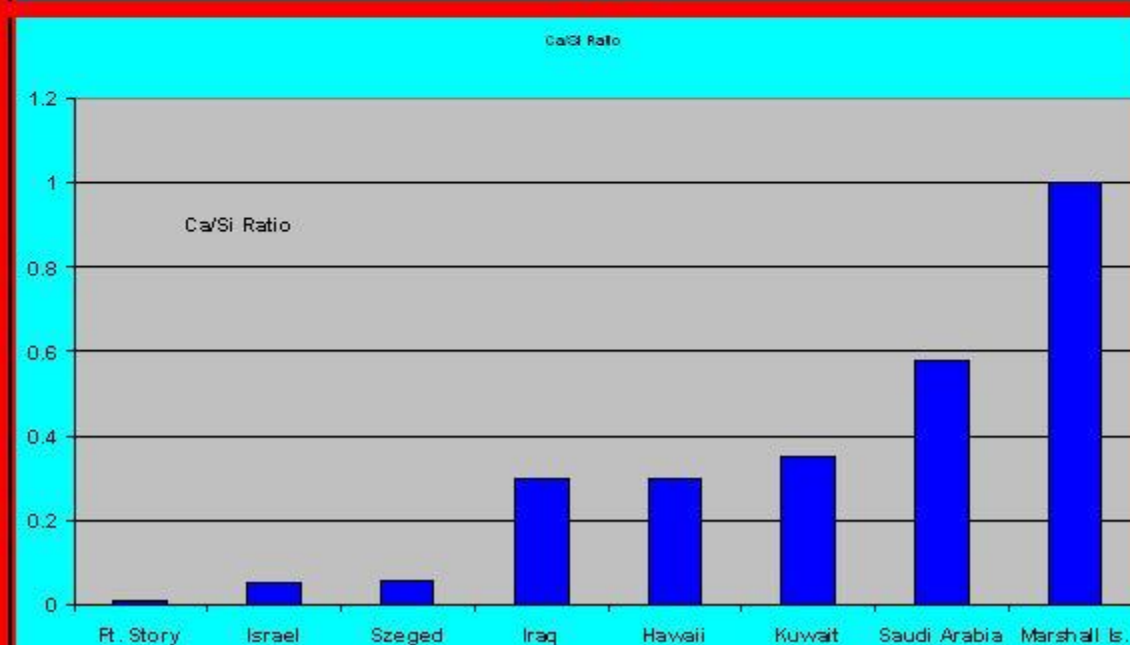
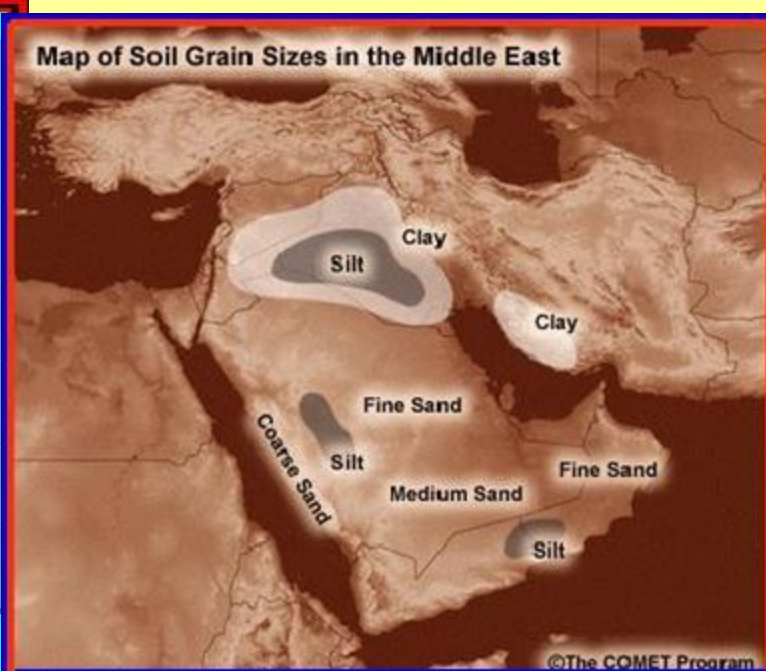
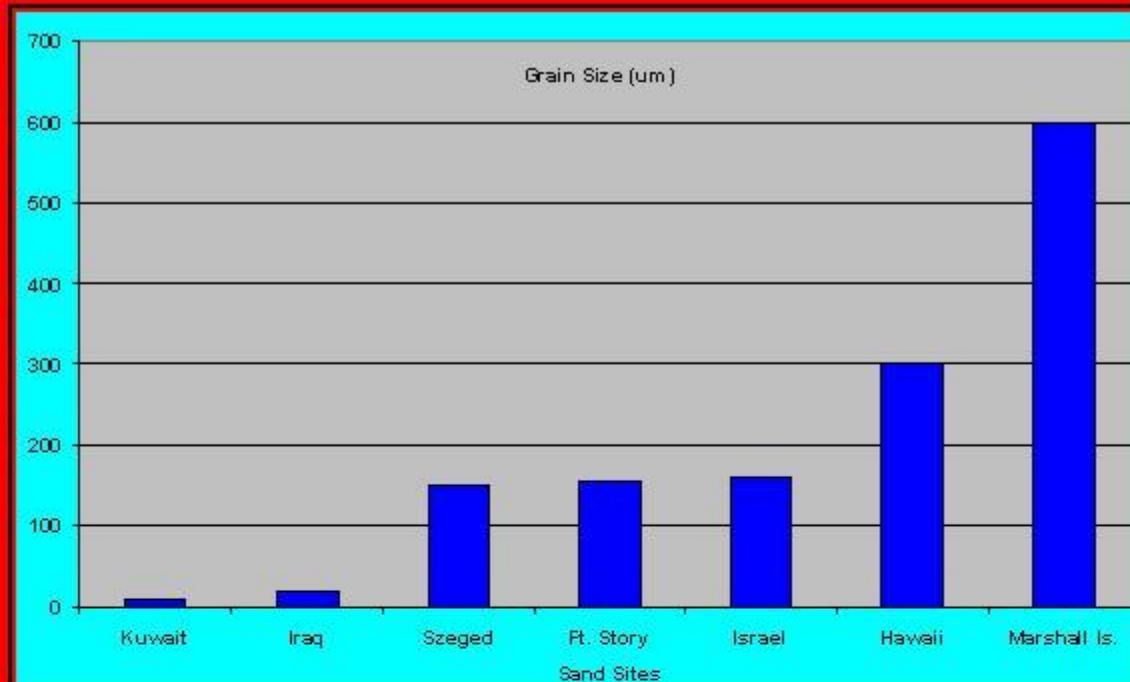
Asian Dust Storm – April 2001

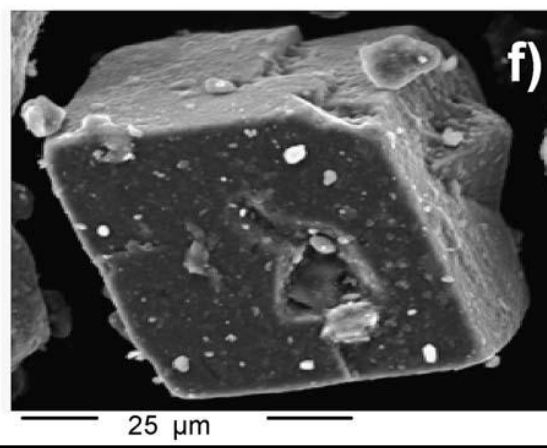
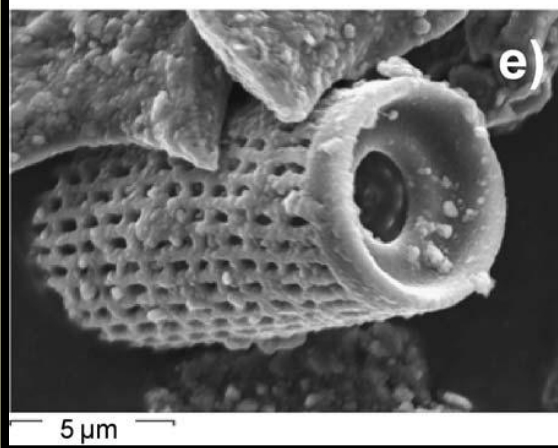
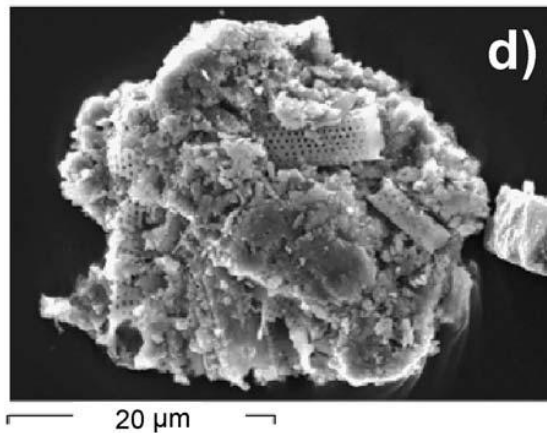
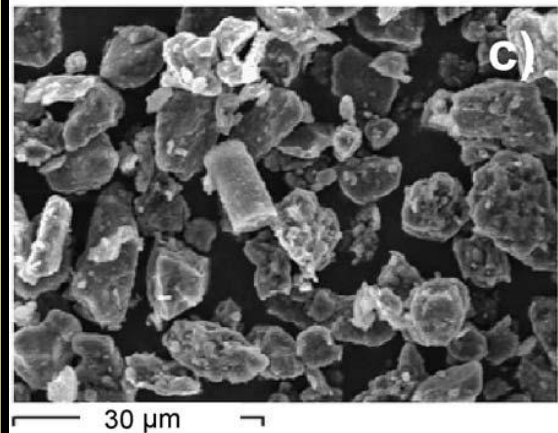
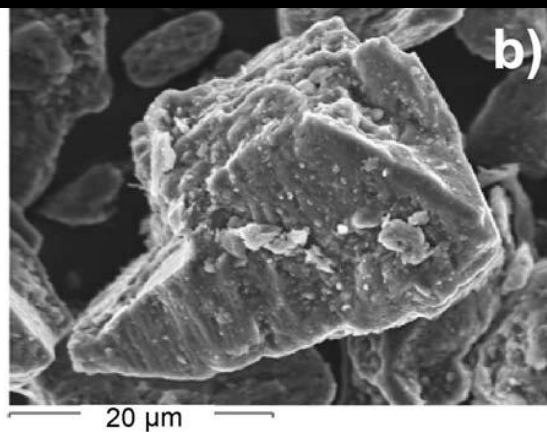
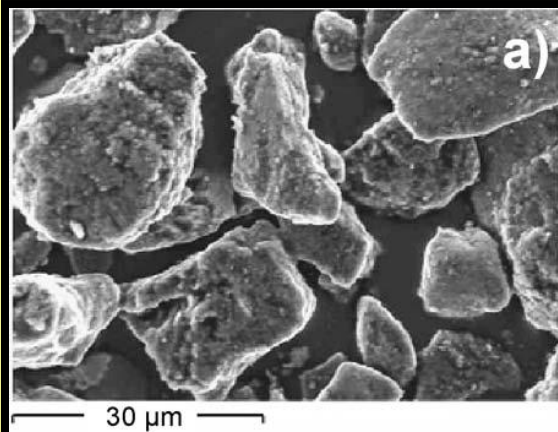


Desert Storms and their ability to move microorganisms and toxins around the globe

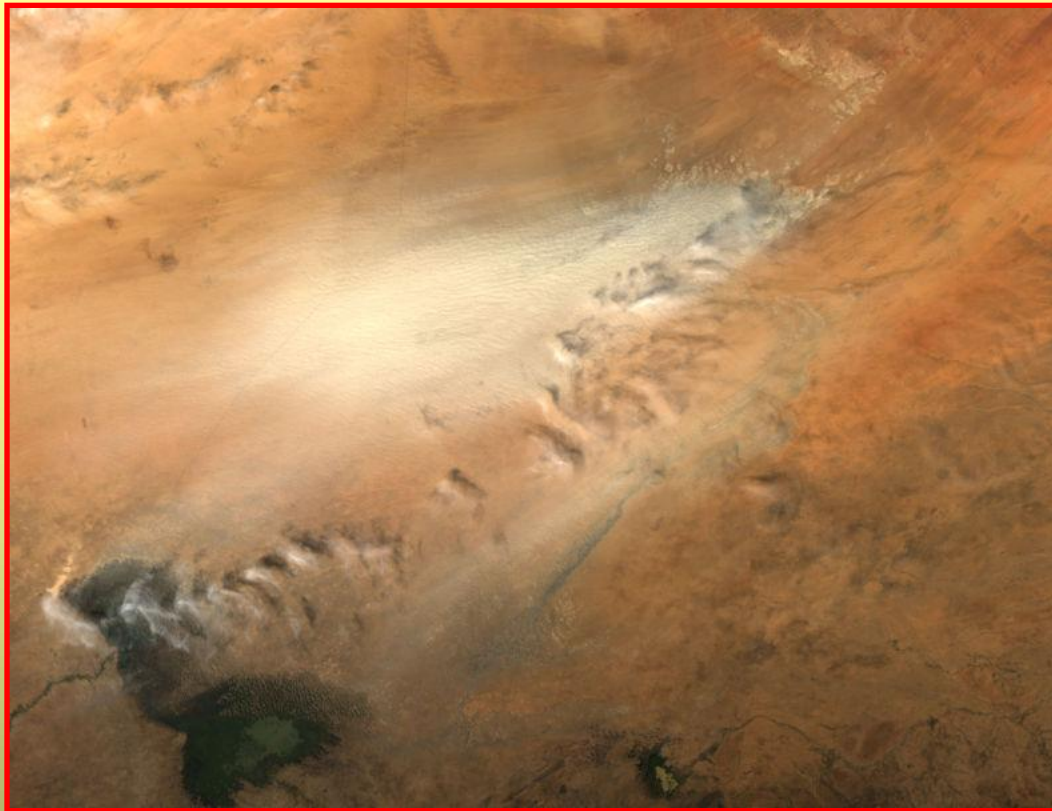


Dale W. Griffin, Ph.D., MSPN, Environmental Microbiologist
U.S. Geological Survey, St. Petersburg, Florida



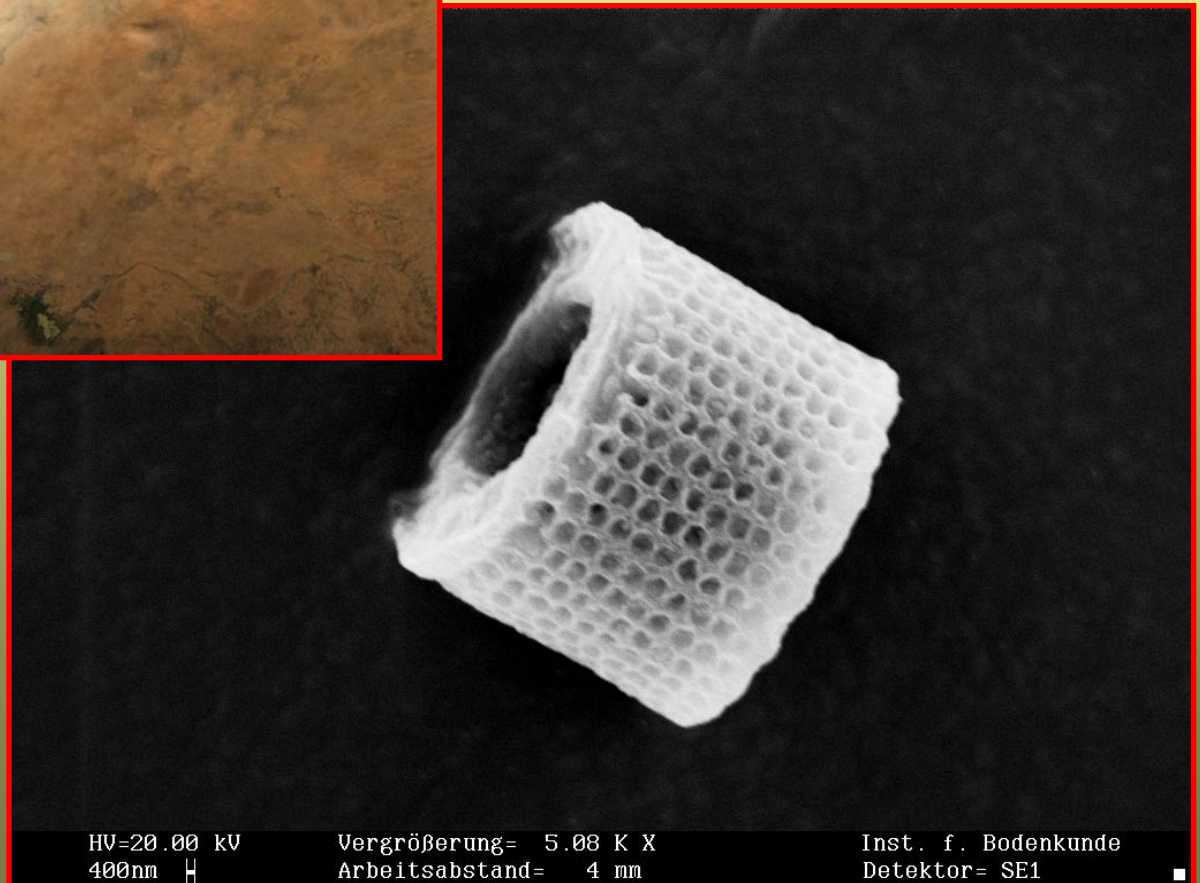


- (a) Hoggar Massif (Algeria) desert soil sample HM2,
- (b) close up of a relatively fresh alkali feldspar in HM2; \
- (c) resuspended road dust Chad Basin sample: note mix of rock forming minerals & diatomaceous materials;
- (d) typical clay-diatom agglomerated clast in CB1;
- (e) detail of an *Aulacoseira* diatom from CB2 sample,
- (f) dolomite- $\text{CaMg}(\text{CO}_3)_2$ from Western Sahara soil samples



From the Bodele Depression

Into Saharan dust



$\times 10^3$

14

12

10

8

6

4

2

0

Into Saharan dust

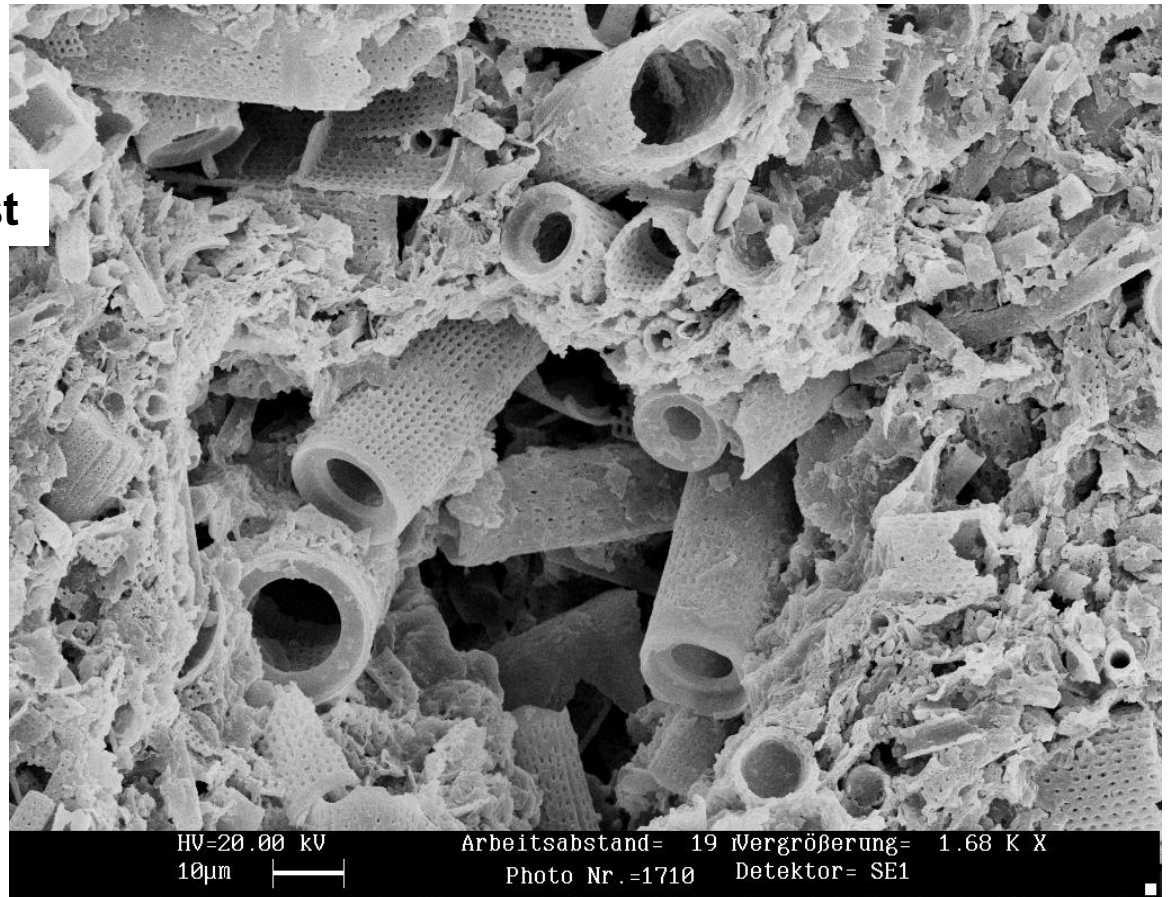
Si

Au

From the Bodele Depression

¹ c.Sa

Energy / keV



CENTCOM DUST: WHY IS IT IMPORTANT!

❖ Force Health Protection Issues

- ❖ Contains Heavy Metals
- ❖ Harbors Pathogens
- ❖ High Percentage in Respirable Range

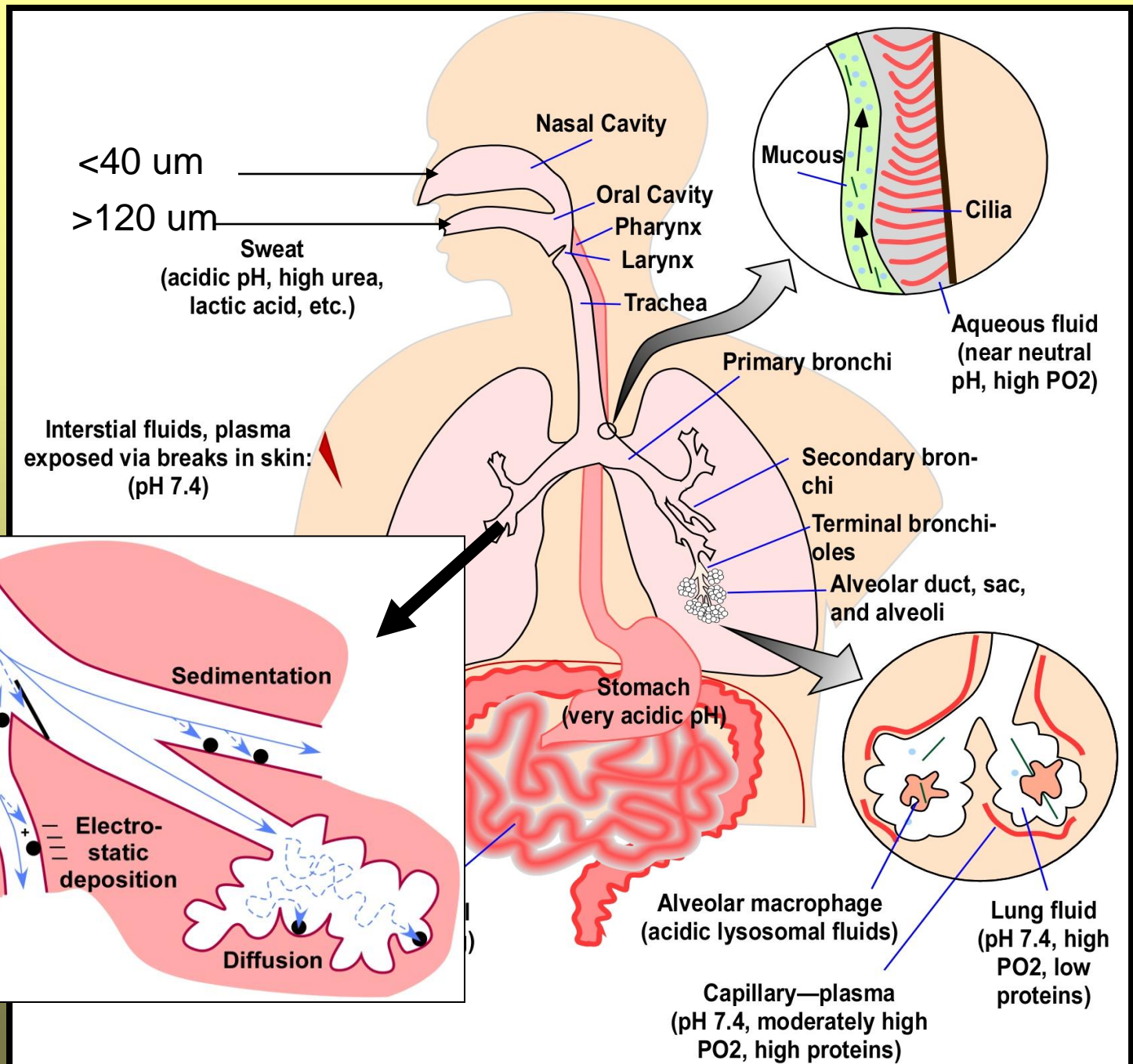
Exposure Issues

- Contact
- Ingestion
- Inhalation

CBD Issues

- Detection
- Analysis
- Contamination





Human Lung Airways

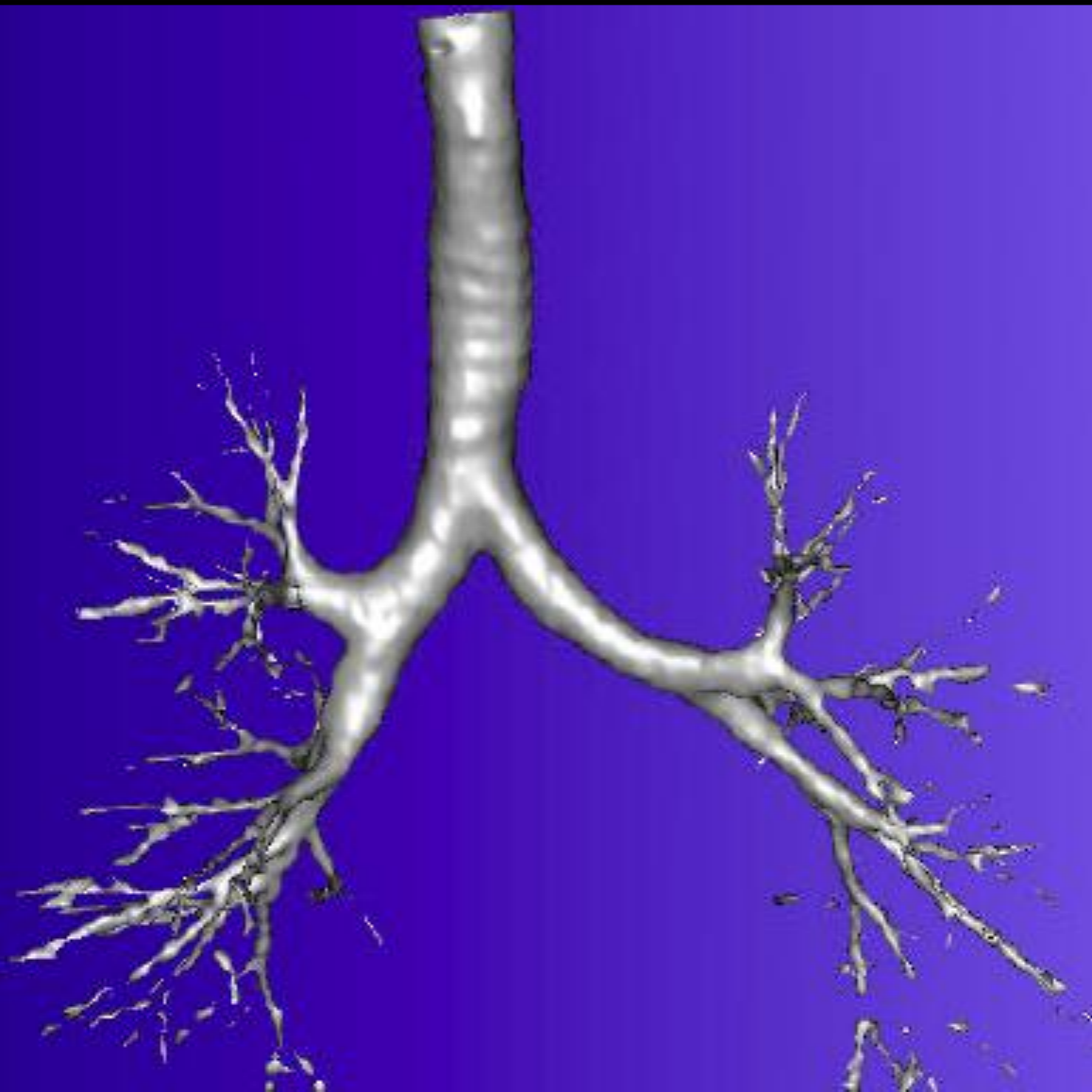


Image reconstructed from high resolution CT scan. Courtesy of Dr. Rod Clinkenbeard, University of Oklahoma

**Moss, O. R. and Oldham, M. J. (2006).
Dosimetry counts: Molecular
hypersensitivity may not drive pulmonary
hyperresponsiveness. J. Aerosol Med.
19(4), 555-564.**

Summary of Exposure

TSP (Total Suspended Particle Mass) (mg/m³) PM10 (10 um) and below

- = 0.001 mg/m³ (NIDBR Lab, Great Lakes, IL)
- = 0.137 mg/m³ (Camp Virginia Clinic, Kuwait - indoors)
- = 2.469 mg/m³ (Highest hourly average - 0800)
- = 9.114 mg/m³ (Highest TSP reading)
- = 2.051 mg/m³ (Highest daily maximum - 18 June @1300)

** NOTE: >9.999 mg/m³ readings recorded during peak dust storms*

Count (Total Number of Suspended PM 10 Particles /m³)

Size Range = 0.5 um to 10 um

- = 1,314,906 (NIDBR Lab, Great Lakes, IL)
- = 12,290,917 (Camp Virginia Clinic, Kuwait - indoors)
- = 107,261,167 (Highest average hourly maximum @1300) (SD = 54,959,015)
- = 588,633,693 (Highest daily maximum – 18 June @1300)
- = 127,643,273 (Highest avg daily (0700-1900) max 13 June) (SD = 34,311,341)

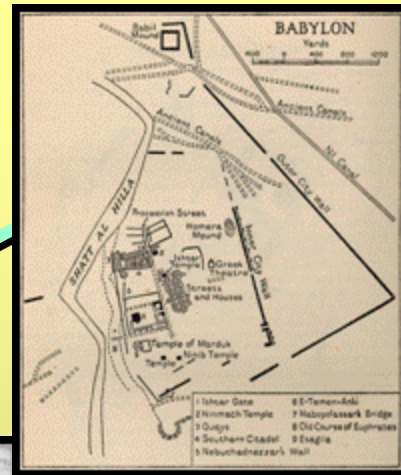
** NOTE: >20,000,000 counts /ft³ readings recorded during peak dust storms or >706,293,334 particles per cubic meter.*

Size Range = 5.0 um to 10 um

- = 36,515 (NIDBR Lab, Great Lakes, IL)
- = 507,824 (Camp Virginia Clinic, Kuwait - indoors)
- = 6,884,417 (Highest average hourly maximum @1300) (SD = 4,142,586)
- = 44,571,347 (Highest daily maximum - 18 June @1300)
- = 5,244,651 (Highest average daily maximum - 13 June) (SD = 3,632,501)



Sampling Locations



Tallil Airbase

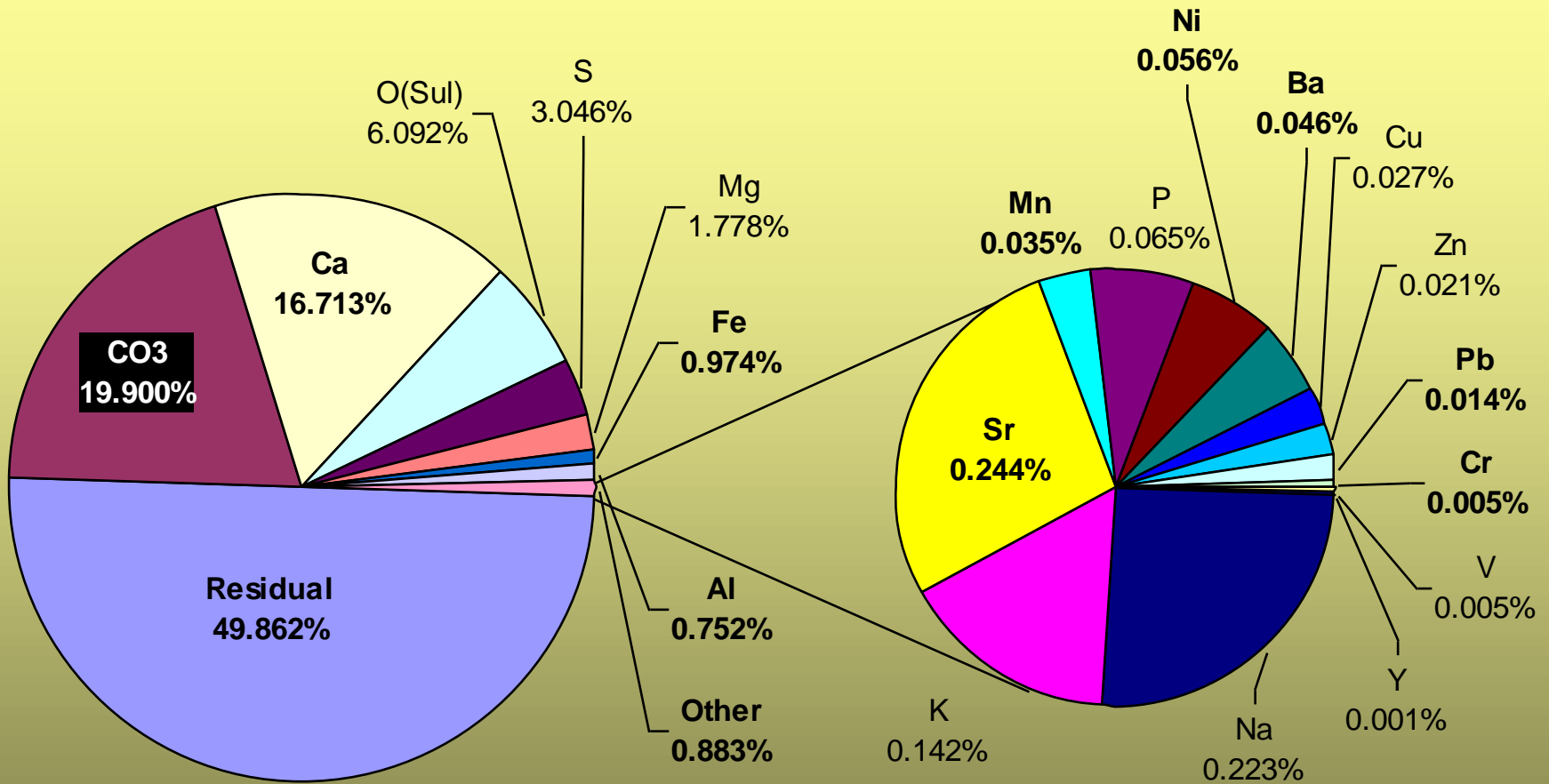


Chemical Analysis: Heavy Metals

Acid Extractables Tent 1

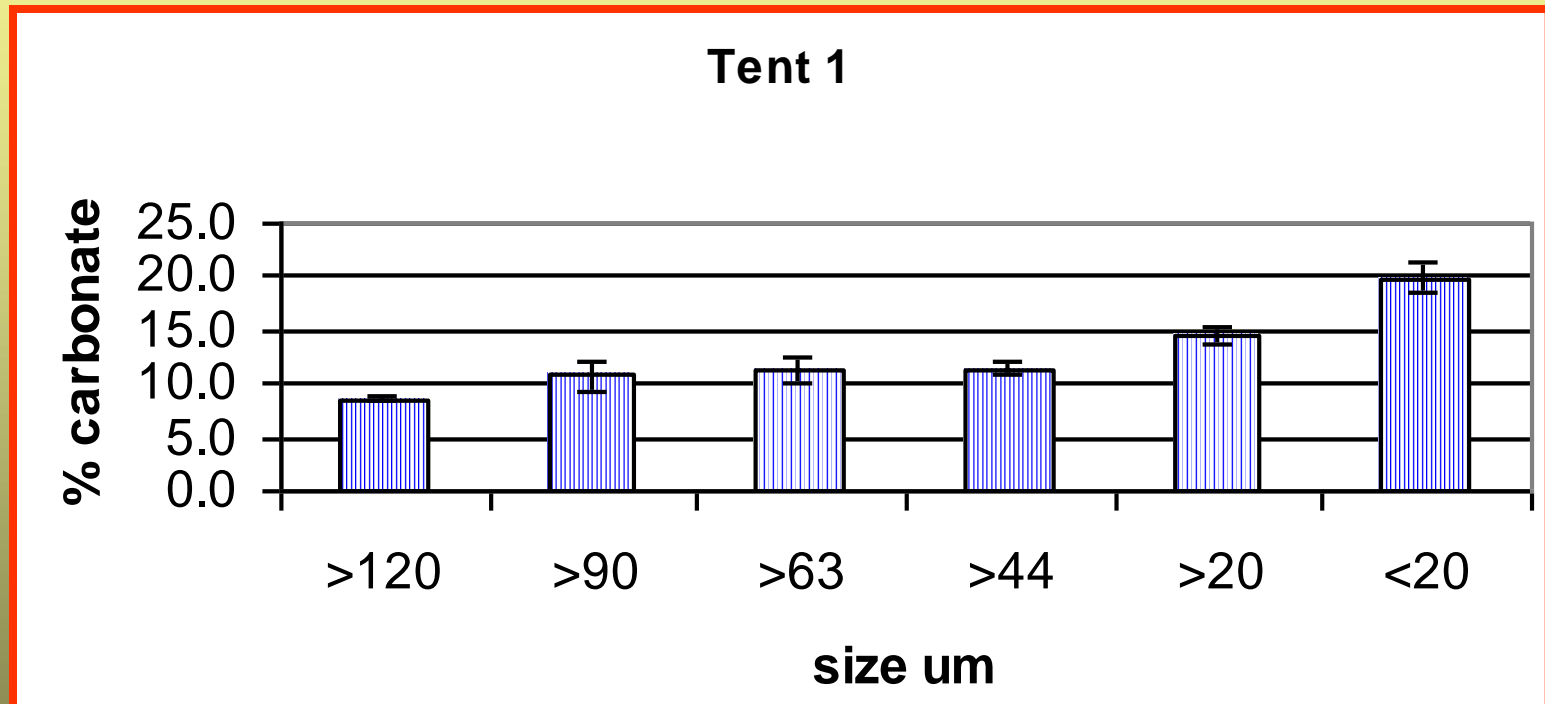
Sample	>120um	>90um	> 63um	>44um	>20um	<20um
Mass	0.2627	0.2596	0.2488	0.2626	0.2441	0.2504
Element	% dry wt	%dry wt	%dry wt	%dry wt	%dry wt	%dry wt
Sr	0.0697	0.0642	0.0995	0.1978	0.2718	0.2436
Ba	0.0068	0.0072	0.0081	0.0192	0.0308	0.0463
P	0.0160	0.0170	0.0234	0.0433	0.0549	0.0649
S	2.4413	2.4230	3.0444	4.0062	3.6646	3.0458
Mg	0.6844	0.8718	1.2672	1.5505	1.7234	1.7784
V	0.0022	0.0026	0.0032	0.0041	0.0046	0.0049
Na	0.1759	0.1963	0.1672	0.2056	0.2123	0.2225
Al	0.2969	0.3832	0.4948	0.6351	0.7164	0.7521
Ca	9.0134	10.3057	11.7495	13.9148	15.3535	16.7133
Zn	0.0053	0.0039	0.0042	0.0070	0.0112	0.0206
Cu	0.0060	0.0050	0.0036	0.0054	0.0077	0.0268
Ni	0.0089	0.0094	0.0169	0.0197	0.0305	0.0564
Y	0.0009	0.0006	0.0006	0.0007	0.0009	0.0010
K	0.0502	0.0653	0.0612	0.0942	0.1186	0.1422
Mn	0.0174	0.0222	0.0268	0.0305	0.0331	0.0352
Fe	0.3506	0.4844	0.6889	0.8419	0.9601	0.9736
Cr	0.0027	0.0032	0.0039	0.0049	0.0052	0.0052
Pb	0.0111	0.0038	0.0049	0.0056	0.0076	0.0138

<20 um Camp Buehring



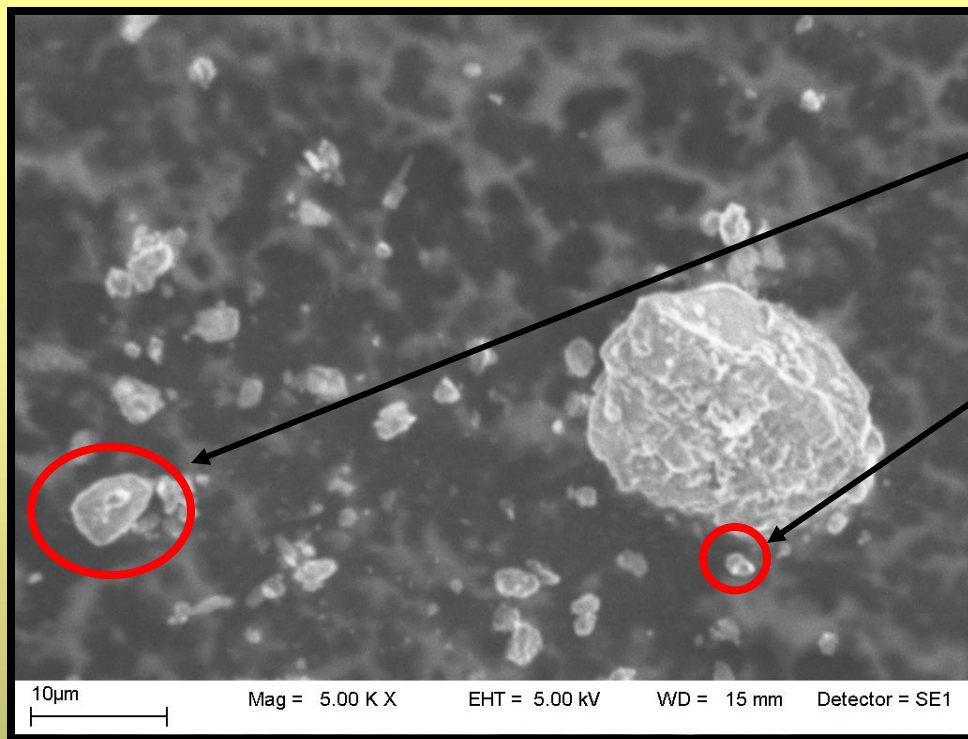
EPA method is SW-846 6010 for ICP-AES and 6020 for ICP-MS. EPA digestion method, 3050.

Chemical Analysis: Carbonates



Initial Conclusions

- Cd, Se do not differentiate by site
- As, Co, Ni, Cr, Pb contents show significant differentiation by site
- Sites S127004 and S127011 (site clusters 5,6) are distinct outliers from the general population of sampled sites
- Site clusters 4,5,6 “elevated” As, Cr
- Possible Pb-Mn association

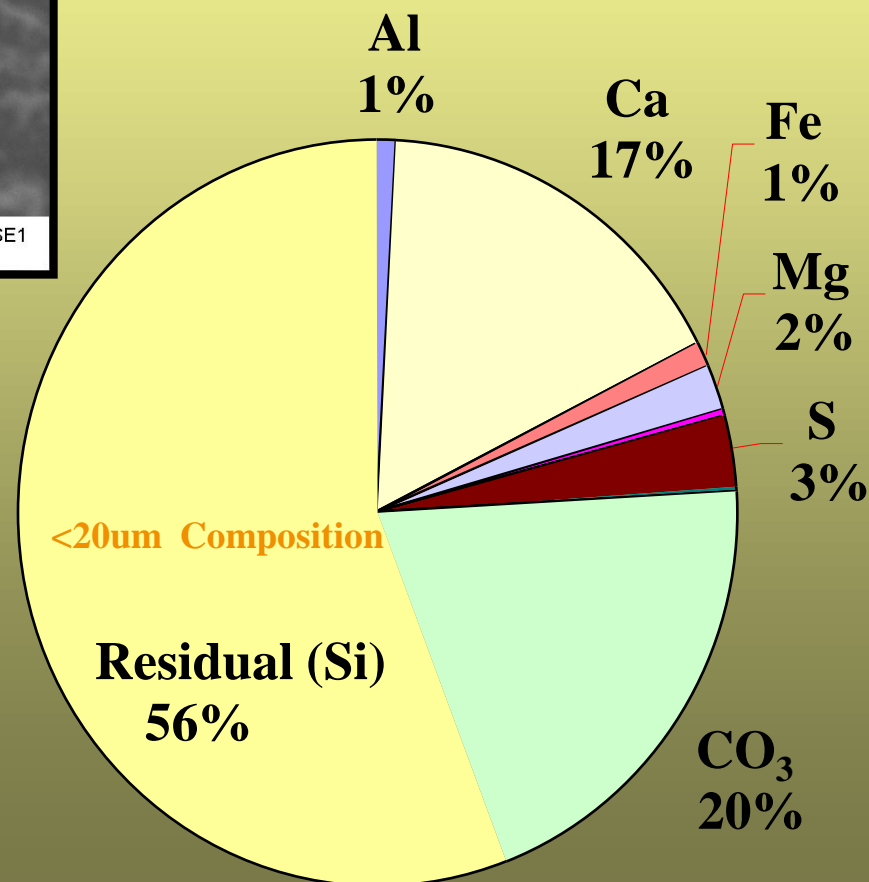


400 of these particles can fit end-to-end across the Head of a Pin.

800 - 1000 of these particles can fit on the Head of a Pin.

Summary

- As particle size decreases, % heavy metals increases.
- Over or near maximum exposure levels for many metals.
- Significant daily loading of trace metals possible.



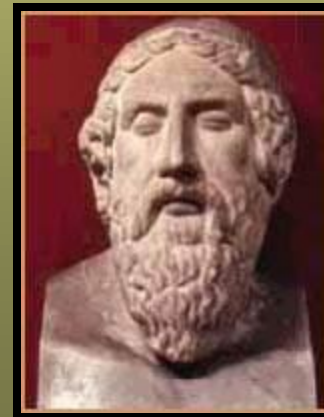
Microbiological Study of Micro-particulates

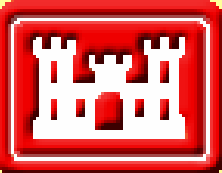
Fifth Plague of Egypt – *“Murrain of beasts” Exodus 9:6*

Sixth Plague of Egypt – “And it shall become small dust in all the land of Egypt, and shall be a boil breaking forth [with] blains upon man, and upon beast, throughout all the land of Egypt”. Exodus 9:10

25 BC: Poet Virgil

- *The Iliad (Homer) “the burning wing of plague...”*
- *Middle Ages: European pandemic “Black Bane” killed 60,000 cattle.*





Culturettes

~32 samples X 6 types of Culturettes = ~192 Culturettes



Black Aerobic & Anaerobic with Charcoal

Blue Aerobic & Anaerobic without Charcoal

Green Industrial quality control

Mico Fungi

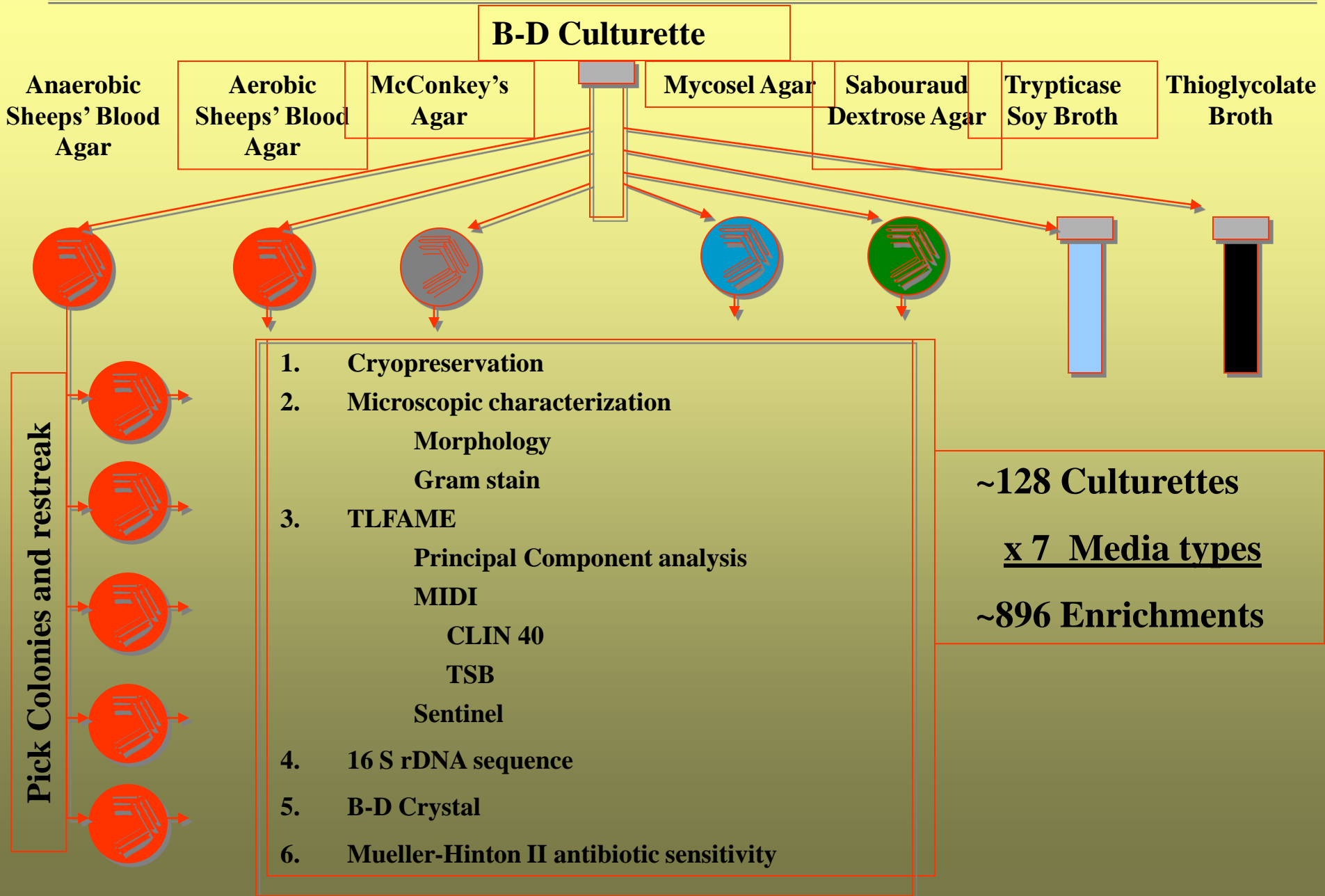
Yellow Chlamydia

Green Virus

Stored at 3⁰C

~32 samples X 4 types of Culturettes = ~128 Culturettes

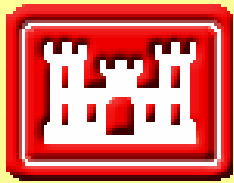
Microbial Isolation and Characterizations



Summary of Soil Isolates

Best ID thus Far	Comment
<i>Neisseria meningitidis</i>	meningitis
<i>Staphylococcus aureus</i>	cystic fibrosis
<i>Bacillus circulans</i>	gastro-enteritis
<i>Pantoea agglomerans</i>	septic arthritis
<i>Pseudomonas agrici</i>	
<i>Ralstonia paucula</i>	opportunistic-septicemia, peritonitis, abscesses
<i>Staphylococcus pasteurii</i>	various infections
<i>Arthrobacter crystallopoietes</i>	
<i>Pseudomonas balearica</i>	cystic fibrosis
<i>Paenibacillus thiaminolyticus</i>	bacteremia
<i>Bacillus vedderi</i>	obligate alkaliphile
<i>Bacillus subtilis</i>	
<i>Pantoea agglomerans</i>	epiphyte
<i>Pseudomonas pseudoalcaligenes</i>	
<i>Cryptococcus albidus</i>	septicemia and meningitis
<i>Bacillus clausii</i>	Oral bacteriotherapy
<i>Kurthia gibsonii</i>	Diarrhea
<i>Bacillus firmus</i>	alkaliphile; bread spoilage
<i>Staphylococcus kloosii</i>	various infections
<i>Bacillus mojavensis</i>	biosurfactant
<i>Bacillus licheniformis</i>	food poisoning
<i>Pseudomonas oryzae</i>	Hickman catheter biofilm





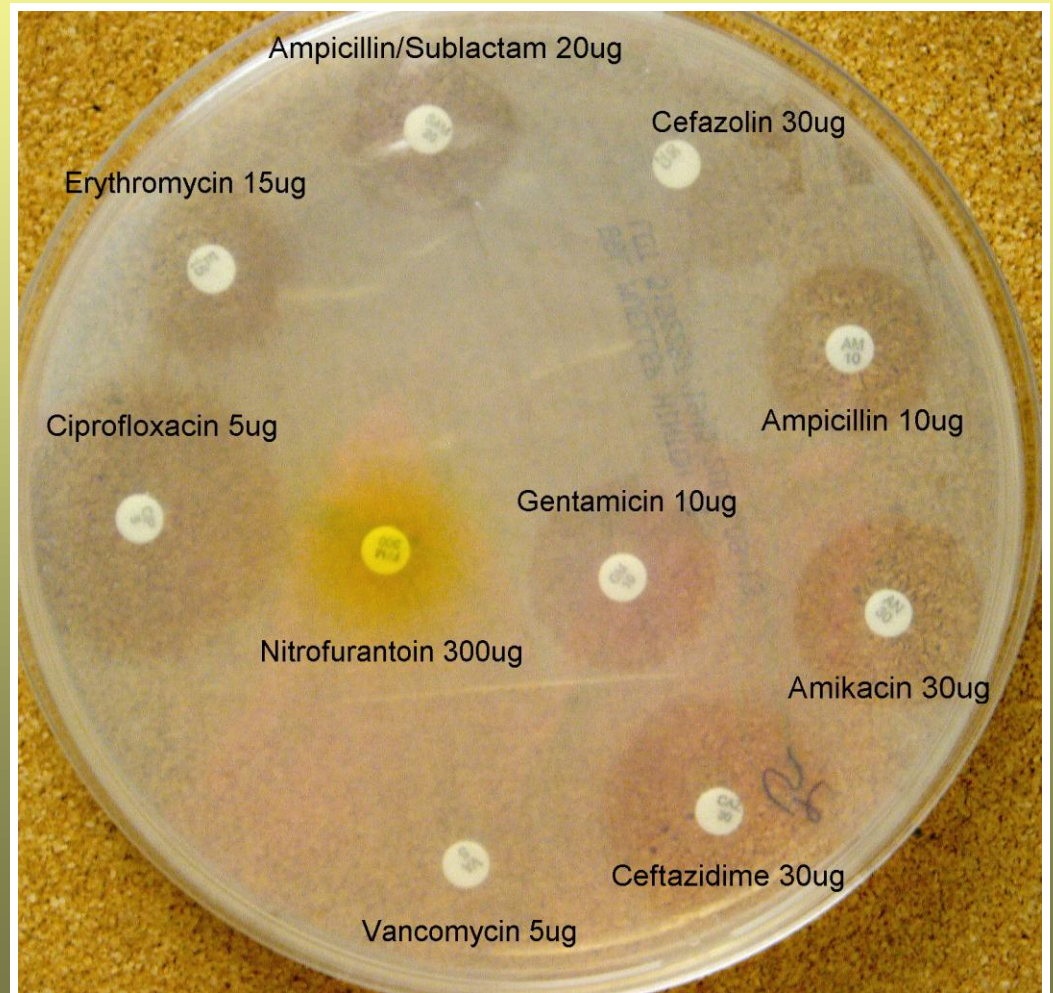
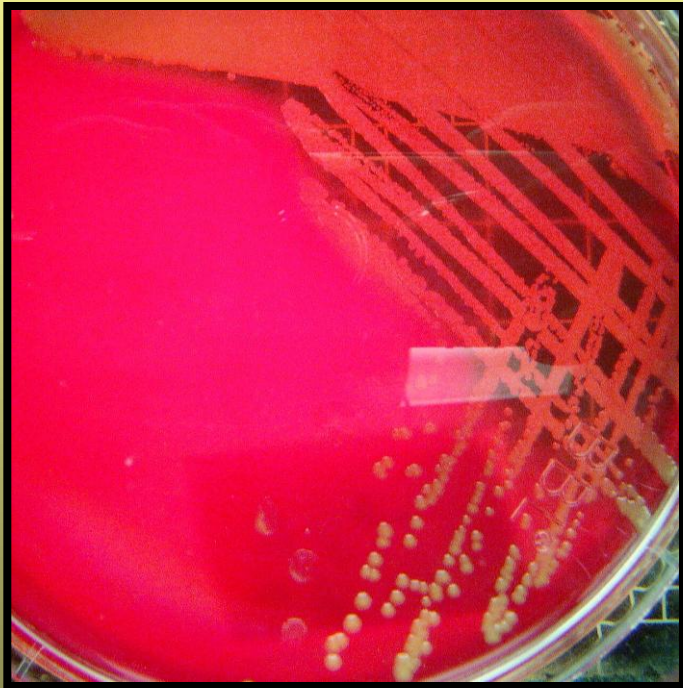
Culturettes

Isolates of Concern - Hemolytic

Isolate Number	Location	Culturette	Hemolytic	Genus and Species Identification							
				16S	Match	TLFAME					
						CLIN 40	Match	Sentinel	Match	TSB	Match
56	Kuwait 2	Blue	Beta	Submitted		Legionella vjordanis	0.16	Microbacterium luteolum	0.25	Microbacterium-liquefaciens*	0.42
69	Kuwait 2	Black	Beta	Submitted		Micrococcus luteus C	0.80	Arthrobacter luteolus	0.49	Arthrobacter-atrocyaneus	0.69
70	Kuwait 2	Blue	Beta	Submitted		Micrococcus luteus C	0.81	Arthrobacter luteolus	0.54	Arthrobacter-atrocyaneus	0.68
72	Kuwait 2	Blue	Beta	Submitted		Tatlockia micdadei*	0.26	Arthrobacter oxydans	0.04	NO MATCH	
I-10	Udari	Orange	Alpha	Pantoea agglomerans	0.95	Neisseria cinera	0.20	Providencia rettgeri	0.02	Ewingella americana	0.78
I-11	Udari	Blue	Alpha	Pseudomonas agrici	0.01	No data		No data		Pseudomonas stutzeri	0.90
I-17	Udari	Black	Alpha/Beta	Paenibacillus thiaminolyticus	0.03	No data		No data		Paenibacillus thiaminolyticus	0.53
I-18	Udari	Orange	Beta	Submitted		Bacillus subtilis	0.52			Bacillus subtilis	0.90
I19	Udari	Blue	Beta/Alpha	Bacillus subtilis subtilis	0.00	No data		No data		Bacillus subtilis	0.92
I-20	Udari	Green	Alpha	Pantoea agglomerans	0.01	Pantoea agglomerans	0.62	Ralstonia paucula	0.27	Pantoea agglomerans	0.82
I-30	Tallil AB	Orange	Beta	Bacillus mojavensis	0.00	Bacillus subtilis	0.42	No Match		Bacillus atrophaeus	0.87
I-31	Tallil AB	Blue	Beta	Bacillus licheniformis	0.02	No data		No data		Bacillus licheniformis	0.61
I-32	Tallil AB	Green	Beta	Flavimonas oryzihabitans	0.00	No data		No data		No data	

Antibiotic Sensitivity

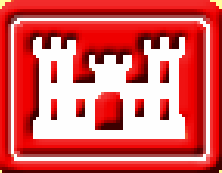
Beta Hemolytic Isolate Number 69



Bacteria Isolated from Kuwait and Iraq that have Shown Antibiotic Resistance.

Culture#	Description	Location	Culturette	Hemolysis	Colony Morphology	MIDI @ DE Environmental	Similarity Index	Comment
8	BSSI	Babylon	Green	No	Dry Fungal type colonies; White spreader on TSA & Blood	Bacillus circulans	0.61	N/A
12	>20<44 um	Udairi	Green	No	Cream colored mucoid colonies on Blood and TSA;	Not growing when others sent off	N/A	N/A
16	>44<63 um	Udairi	Green	No	Small mucoid colonies on Blood; Spreading mucoid on TSA;	Staphylococcus warneri	0.881	N/A
20	>44<90 um	Udairi	Green	alpha	Small dry cream colored colonies	Pantoea agglomerans Pantoea agglomerans	0.82 0.711	GC subgroupB GC subgroupC
24	<20 um	Udairi	Green	No	Shiny yellowish/cream spreading colonies on TSA; Purple spreader on Blood;			
28	TAB II Sand A	Tallil	Green	No	Large shiny mucoid colonies	Not sent to MIDI	N/A	N/A
32	TAB II Sand B	Tallil	Green	beta	Clear white cauliflower colony on TSA; Shiny clear runny colony on Blood;	Not sent to MIDI	N/A	N/A

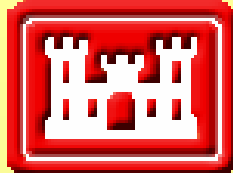
Culture#	MIDI @ MS CLIN 40	Similarity Index	Comment	MIDI @ MS Sentenial	Similarity Index	MIDI 500 bp rDNA sequence analysis % Diff	Comments
8	No match/Too dilute	N/A	N/A	No match			
12	Vibrio alginolyticus Aeromonas hydrophilia	0.366 0.366	N/A N/A	Ralstonia paucula Erwinia mallotivora	0.127 0.103		
16	Pseudomonas stutzeri	0.44	N/A	Pseudomonas balearica	0.097		
20	Pantoea agglomerans Aeromonas hydrophilia	0.623 0.386	GC subgroup N/A	Ralstonia paucula Buttiauxella gaviniae	0.274 0.175	Pantoea agglomerans	0.85% Plant/Human Pathogen
24							
28	Not Extracted	N/A	N/A				
32	Not Extracted	N/A	N/A			Flavimonas oryzae	0.10% Hickman Cath Pathogen



Fungal Isolates

~300 bp of D2 region of LSU rDNA

Microseq Library database							
Midi D2(300 bp)LSU rRNA	% diff	LSU D2 Genbank Database	% ID	Associated Disease			
Allewia eureka	0.31	Ulocladium sp.	99				
Allewia eureka	4.64	Cryptococcus uzbekistanensis	100				
Allewia eureka	0.31	Ulocladium sp.	99	None			
Altemaria altemata	0	Alternaria sp	100	Plant pathogen			
Rhodotorula minuata	5.73	Rhodotorula minuata	99	Eye infections			
Cryptococcus albidus	2.44	Cryptococcus sp.	100	Cryptococcus neoformans - meningoencephalitis			
Ulocladium chartarum	0.31	Stemphylium sp	99	Fungal biocontrol agent			
Filobasidium uniguttulatum	4.64	Cryptococcus uzbekistanensis	100	Teleomorph of Cryptococcus, non pathogenic yeast			
Ulocladium consortiale	0	Stemphylium sp	100	cutaneous mycoses			
Ulocladium chartarum	0.31	Stemphylium sp	99				
Mortierella polycephala	7.1	Mortierella polycephala	92	Pulmonary mycosis in cattle			
Embellisia chlamydospora	0	Ulocladium sp.	99	none			
Filobasidium uniguttulatum	4.64	Cryptococcus uzbekistanensis	100				
Penicillium camembertii	0	Penicillium sp.	100				
Cryptococcus albidus	0	Cryptococcus albidus	100				
Allewia eureka	0.31	Ulocladium sp.	99				
Embellisia chlamydospora	0	Ulocladium sp.	99				
Filobasidium uniguttulatum	4.64	Cryptococcus uzbekistanensis	100				
Embellisia chlamydospora	0	Ulocladium sp.	99				
Filobasidium uniguttulatum	4.64	Cryptococcus uzbekistanensis	100				
Penicillium camembertii	0	Penicillium sp.	100				
Allewia eureka	0.31	Ulocladium sp.	99	Plant pathogen			
Phoma glomerata	0	Phoma herbarum	99				



Dust – Pad 15 PM₁₀₋₂₀



Agar Enrichments – Comparison of media/methods

Chocolate

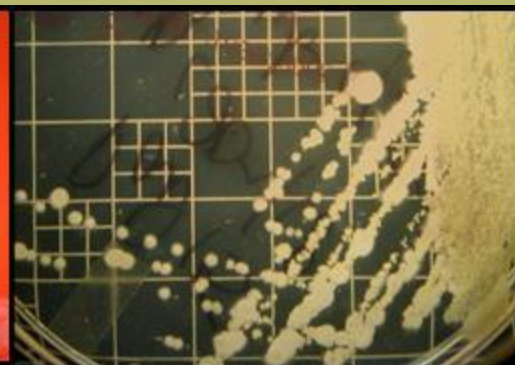
Sheeps' Red Cell

Trypticase Soy

Swab Dust



**TSB Enrich &
Swab**



Dust

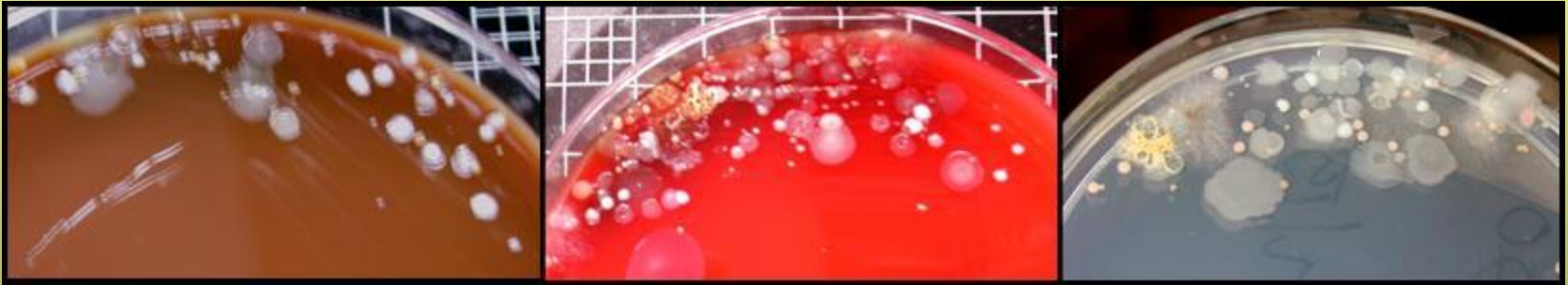
Particle size – media comparisons

Chocolate

Sheeps' Red Cell

Trypticase Soy

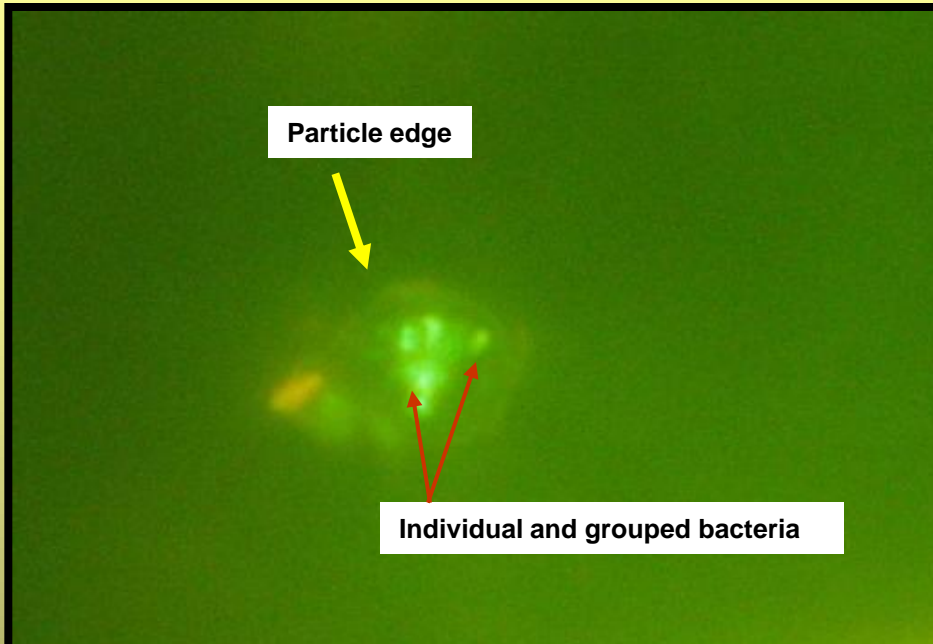
Pad 15 PM₁₀₋₂₀



Pad 15 PM₂₀₋₄₀

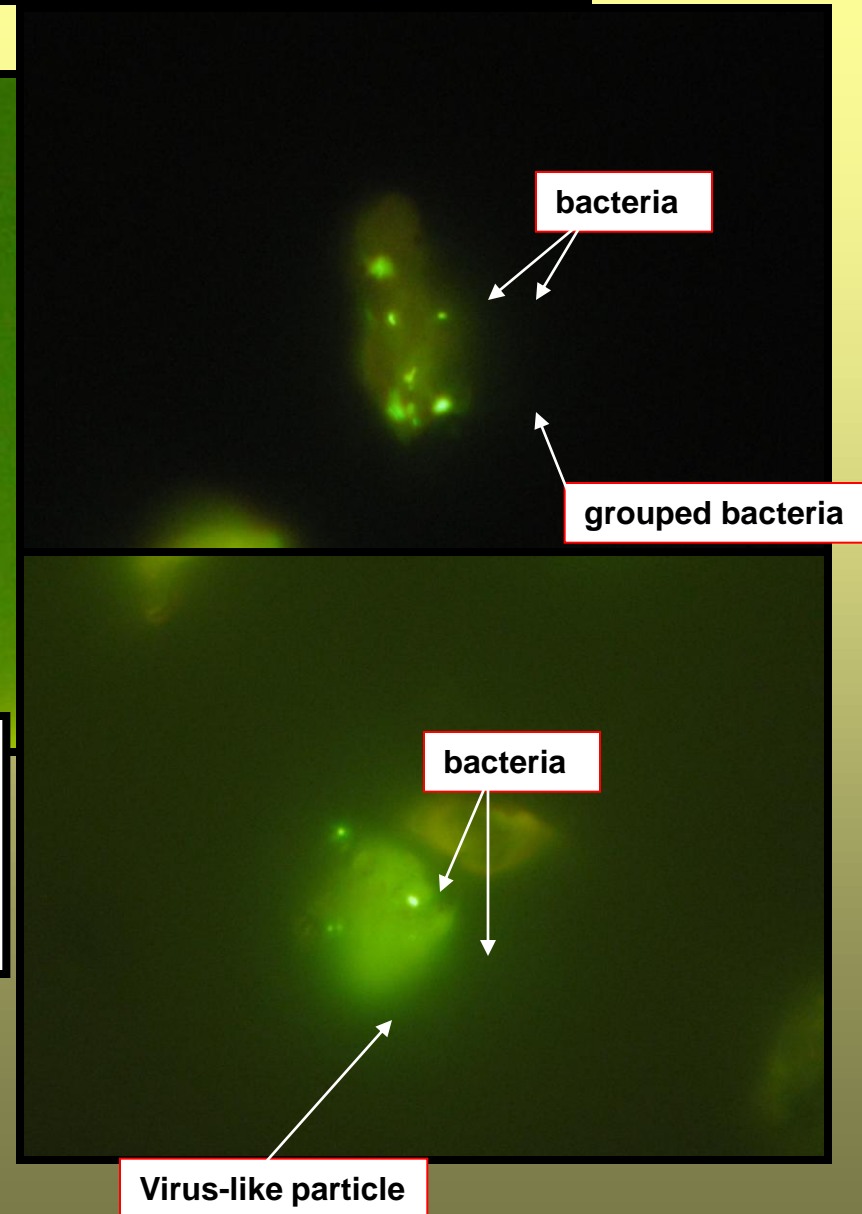


Kuwait dust, Camp Buehring, size fraction 10 to 20um



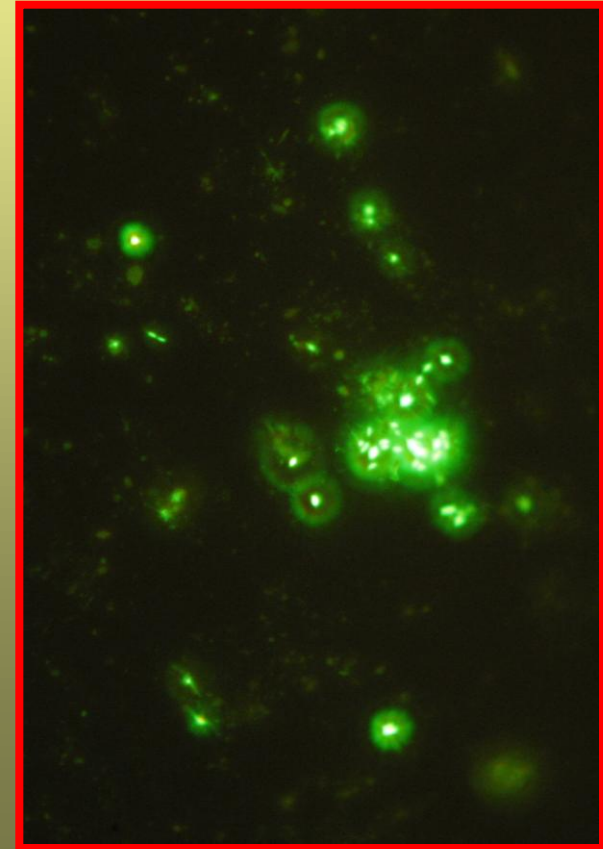
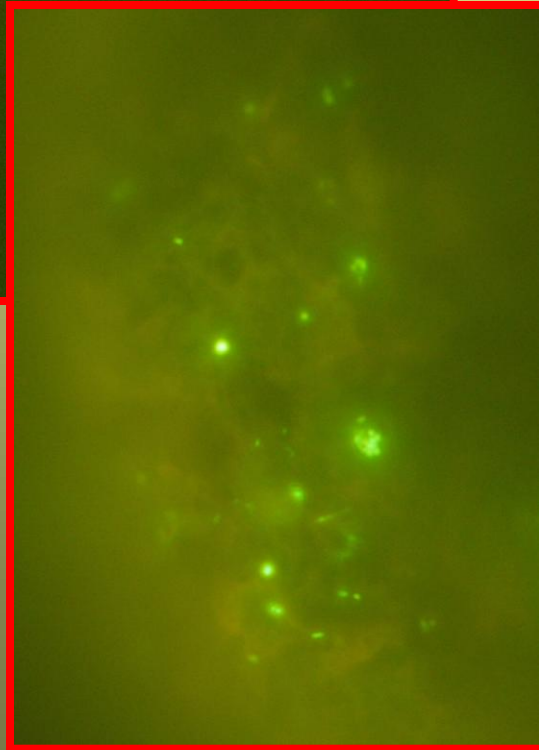
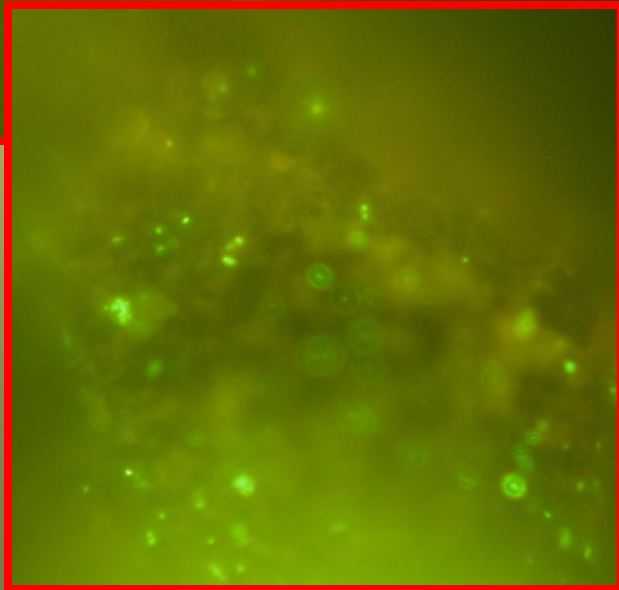
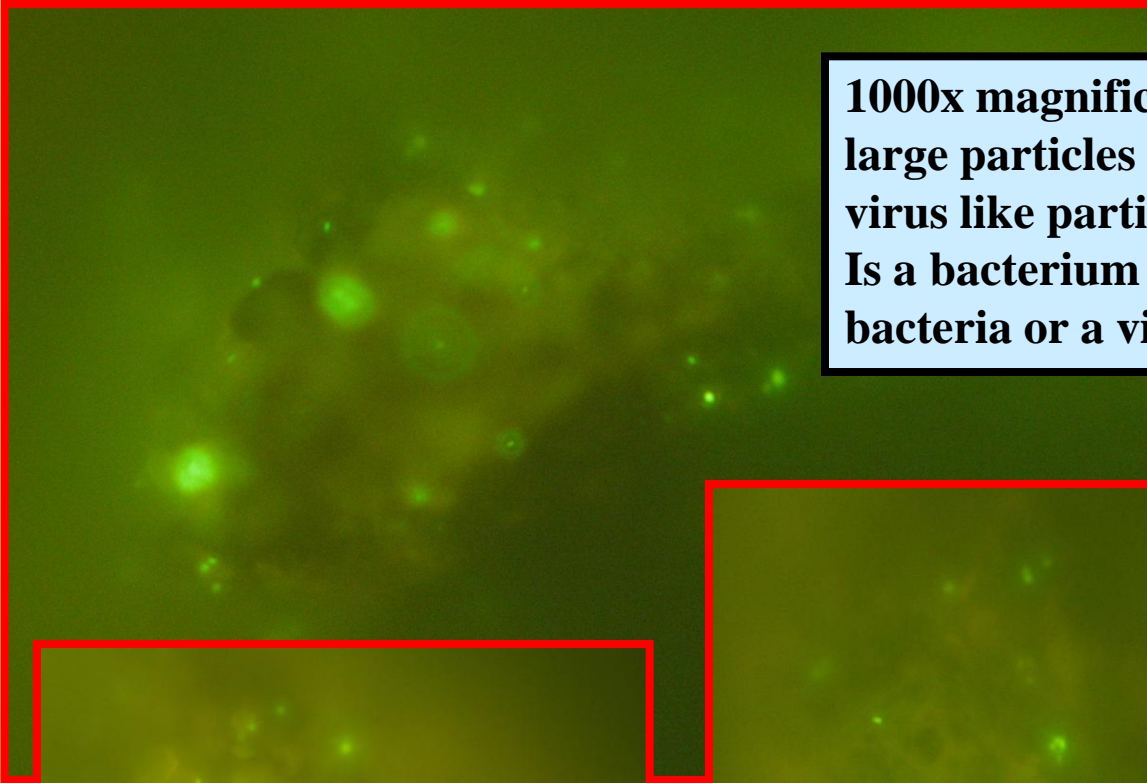
This photo shows a particle that appears to contain numerous bacteria in its core rather than on its surface. Bacteria size ~1um. 1000X with digital zoom.

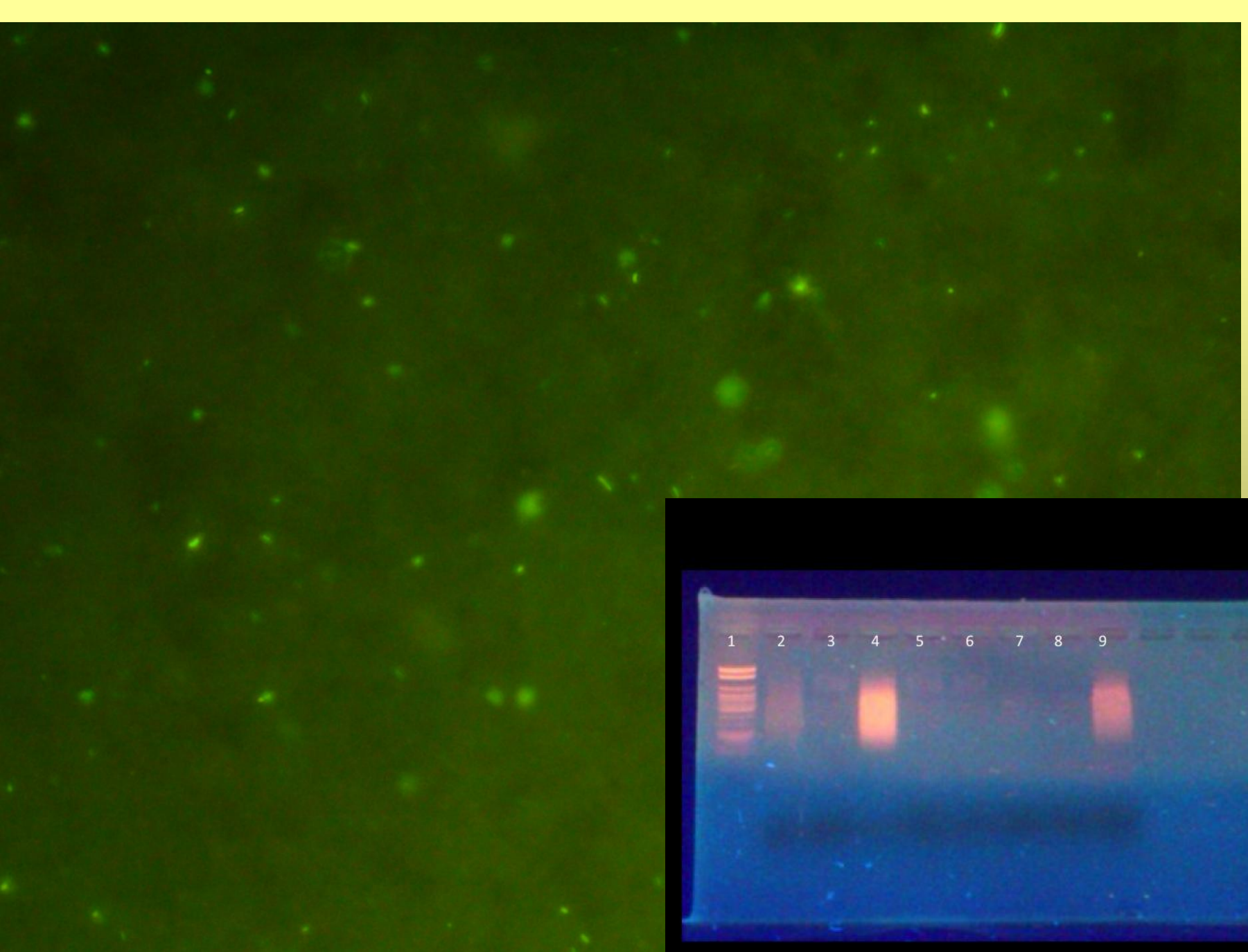
Sterilization Problems!

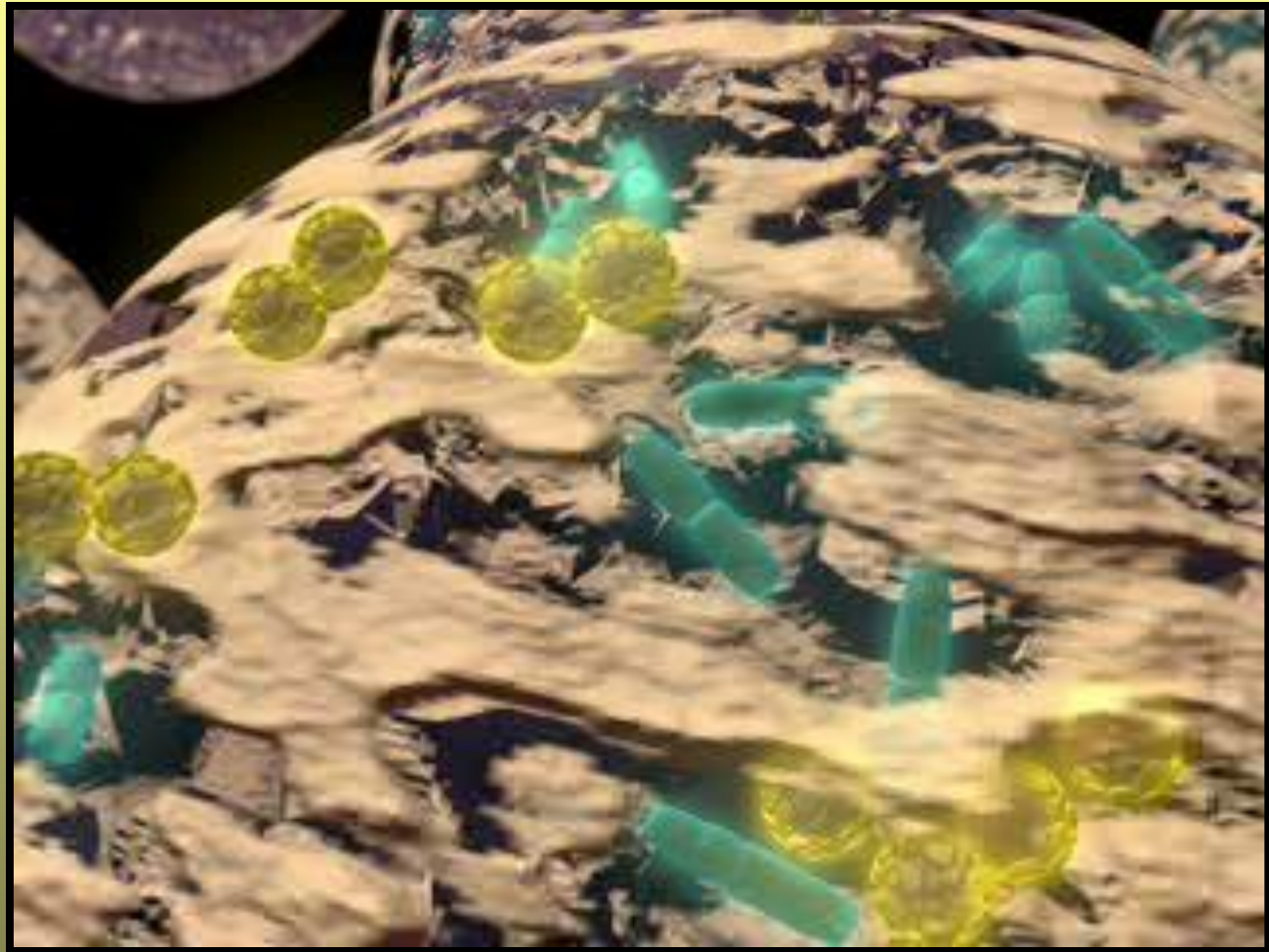


Kuwait dust, sample #2 raw (bulk)

1000x magnification. These images show large particles coated with bacteria and virus like particles. Each fluorescing particle is a bacterium (~1µm in size), group of bacteria or a virus-like particle





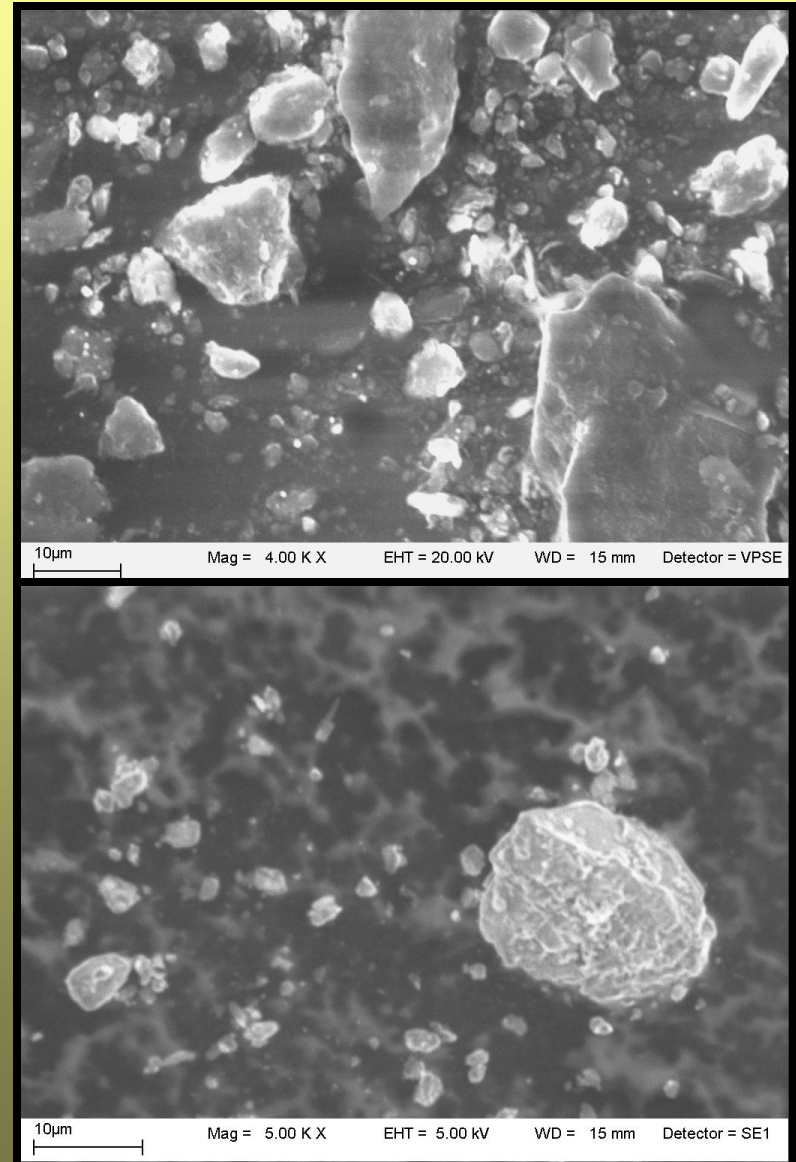


Summary of Scientific Results

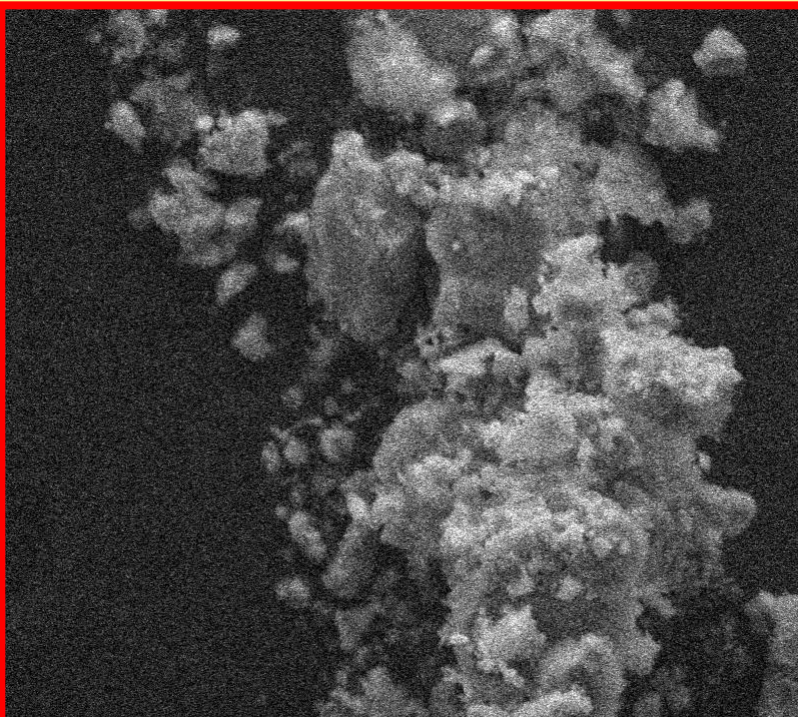
- Approximately 147 different isolates identified to date (6 Genera by 16s DNA analysis). 13 alpha/beta hemolytic species have been identified and 27 fungal isolates have been identified (7 different Genera). Several species have shown antibiotic resistance. More are expected from the low nutrient studies.
- *Acinetobacter spp.* has been identified by FAME analysis, but species yet to be determined.
- We have 6 isolates of *Neisseria* with FAME analysis.
- A total of 54 elements were screened for with 37 different elements identified of which 15 are bioactive metals including Uranium. Of these the ones of greatest concern are: Arsenic (10 ppm), Chromium (52 ppm), Lead (138 ppm), Nickel (564 ppm), Cobalt (10 ppm), Strontium (2700 ppm), Tin (8 ppm), Vanadium (55 ppm), Zinc (194 ppm), Manganese (369 ppm), Barium (327 ppm), Aluminum (9400 ppm).
- Elemental data suggests that minerals and elements tend to cluster geochemically within sites, and there are significant geochemical differences between some of the sites that seem to impact presence of specific toxic trace metals and their concentrations in the dust/dirt.
- Sterilization Experiments suggest an exceptional ability for microbes to survive.
- Early animal studies have suggested long term inflammation with mild to moderate eosinophilia.

This is **NOT** sand!

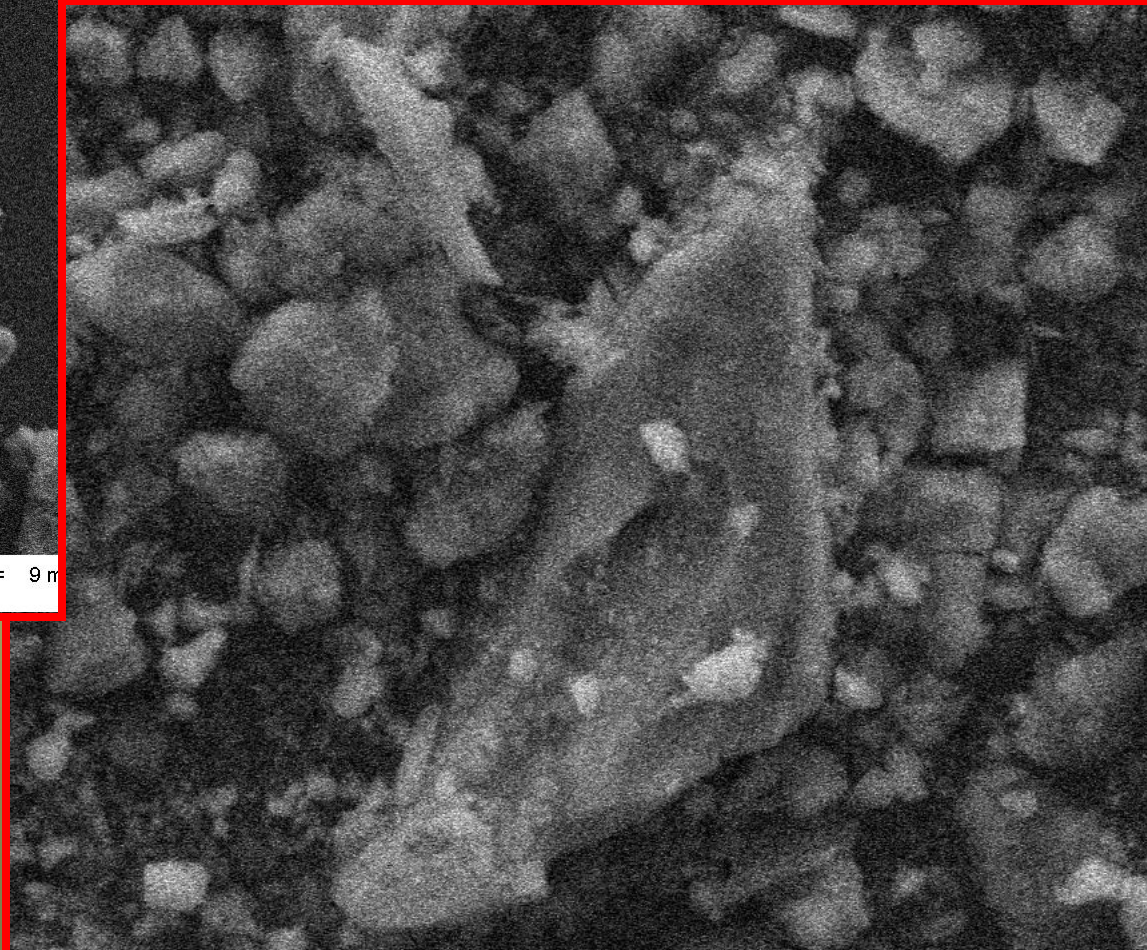
- These micro-particulates are composed of a porous silica core (crystalline metallic silica and amorphous silica dioxide) surrounded by a type of clay consisting of, primarily, Calcium Carbonate (CaCO_3) and Magnesium Sulfate (MgSO_4).
- The laminar nature of the clay coating greatly magnifies the surface area and contributes to the hygroscopic nature of the particle.
- The micro-particulates act as a protective coating and carrier for the micro-organisms.



Afghanistan Dust Sample

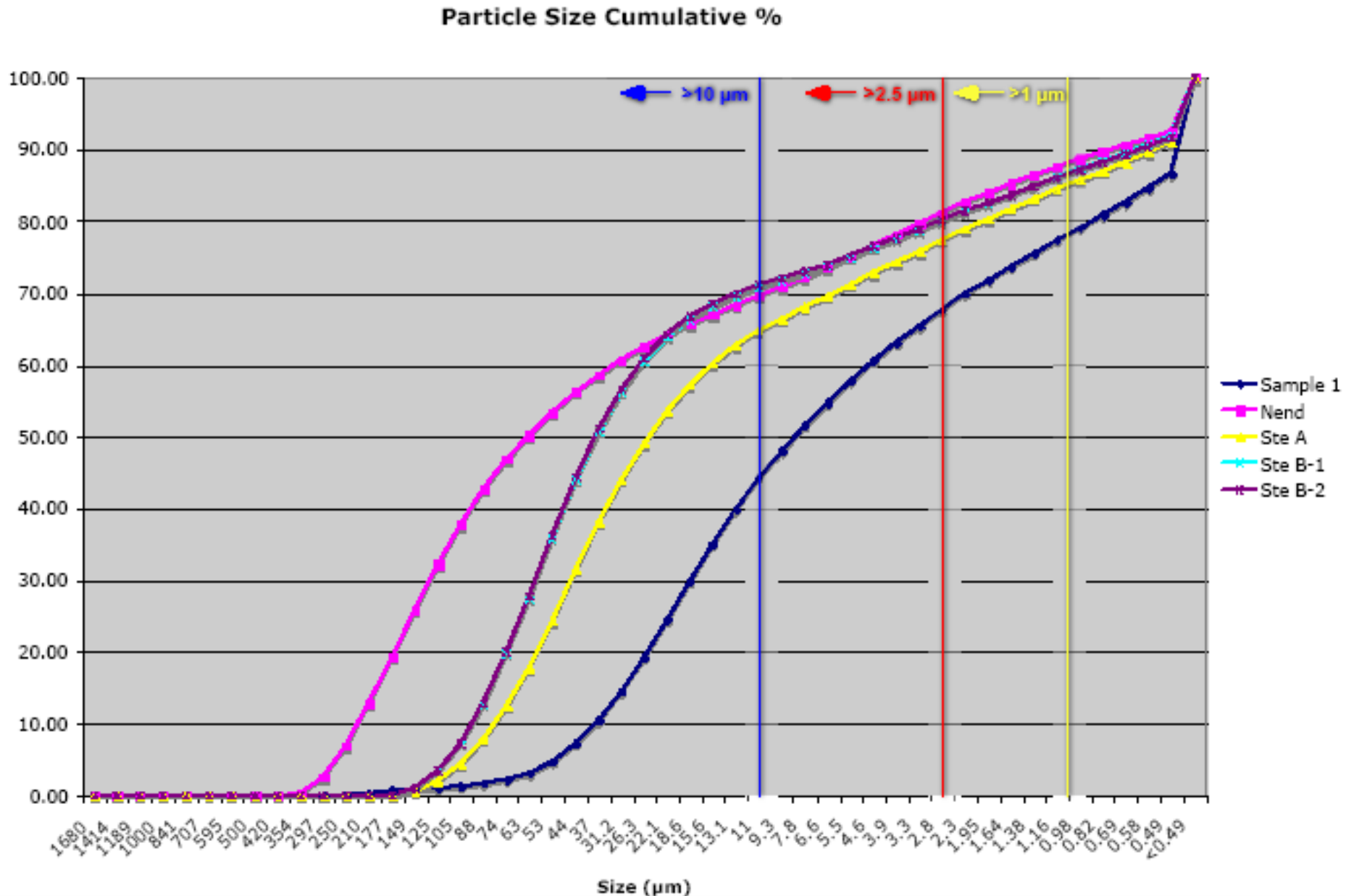


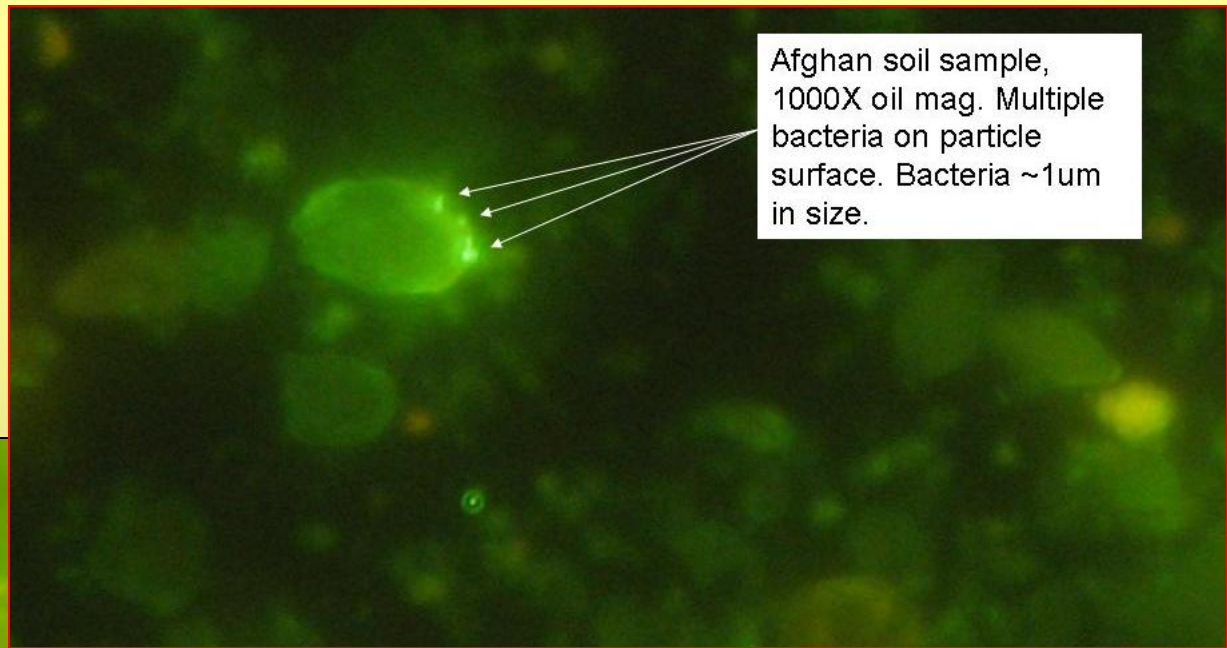
10 μ m Mag = 5.00 K X EHT = 12.12 kV WD = 9 mm



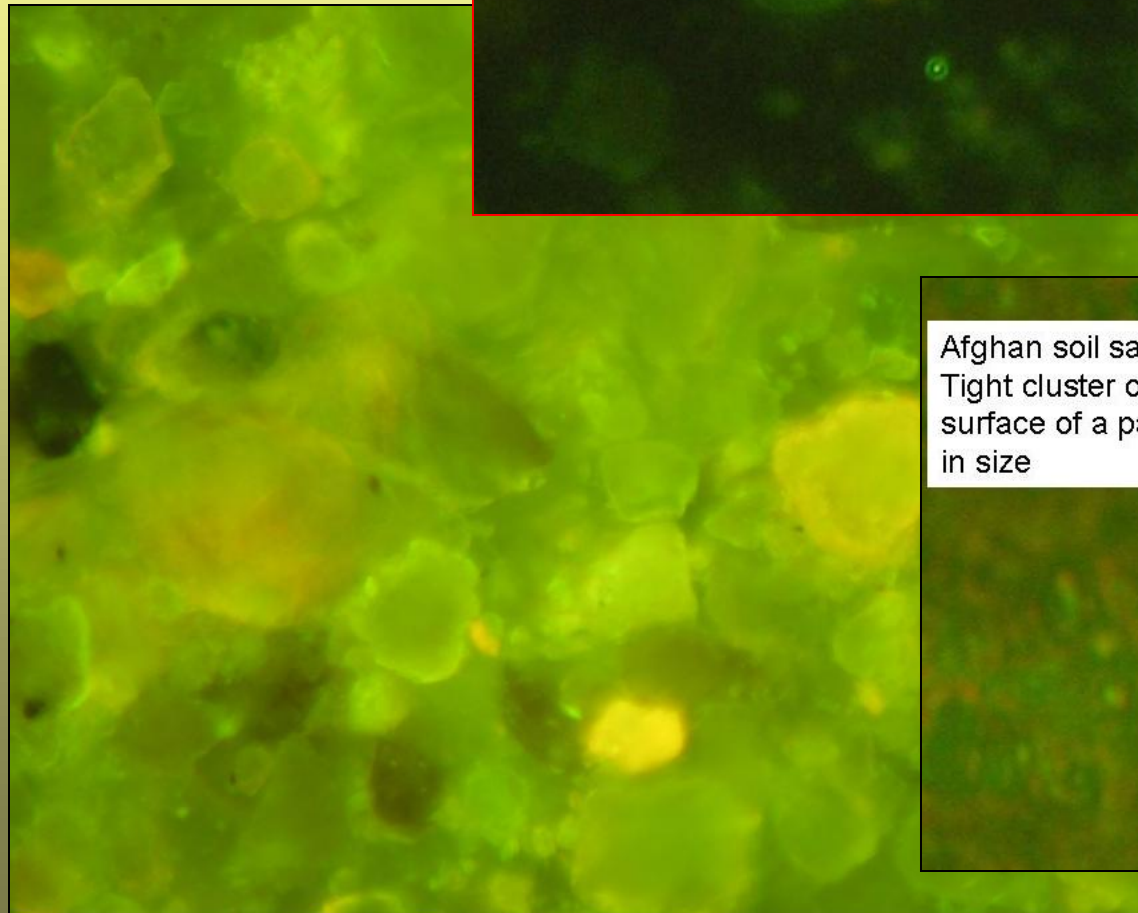
2 μ m Mag = 10.00 K X EHT = 12.12 kV WD = 9 mm

Afghanistan Dust Sample

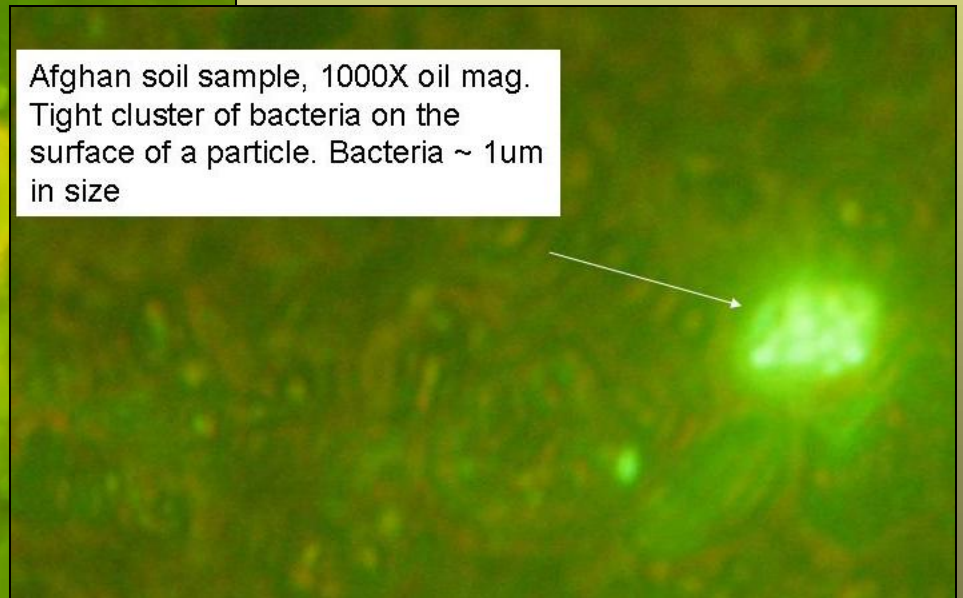




Afghan soil sample, 1000X oil mag. Multiple bacteria on particle surface. Bacteria ~1 μ m in size.



Afghan soil sample, 1000X oil mag. Tight cluster of bacteria on the surface of a particle. Bacteria ~ 1 μ m in size



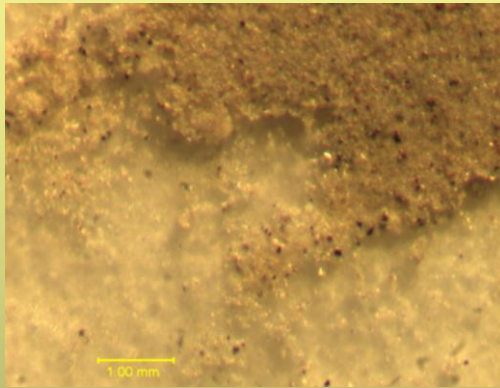
A combination of epifluorescence and white light = illuminated particles...neat!

COMPARISONS

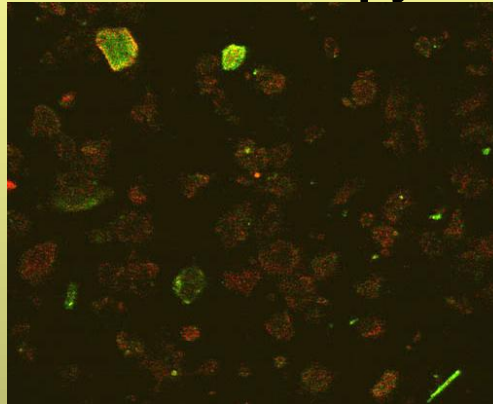
Laboratory Comparisons of Iraqi and Ft. Irwin Dust initiated

Iraq

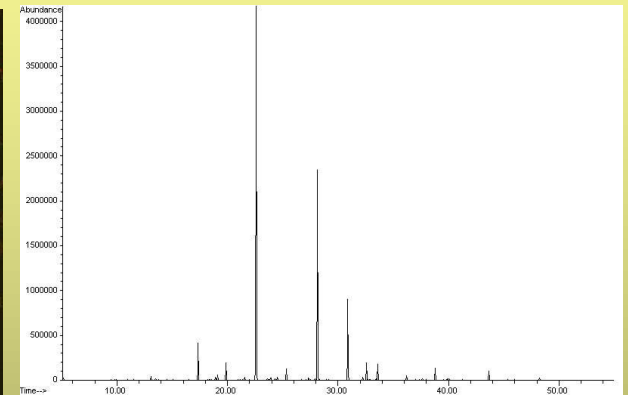
Light Microscopy



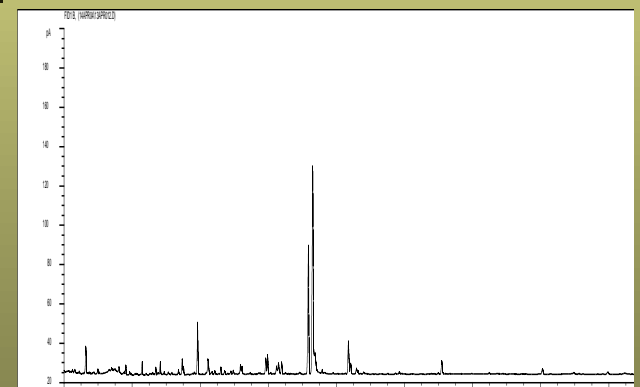
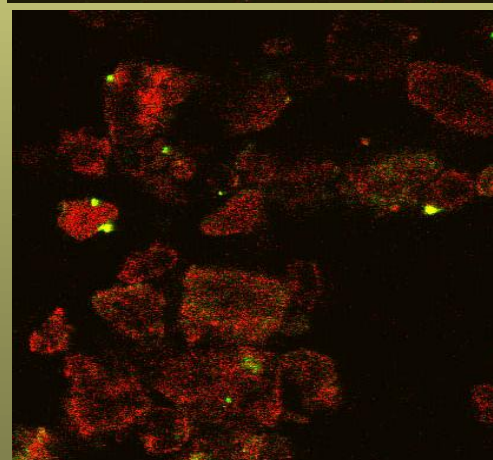
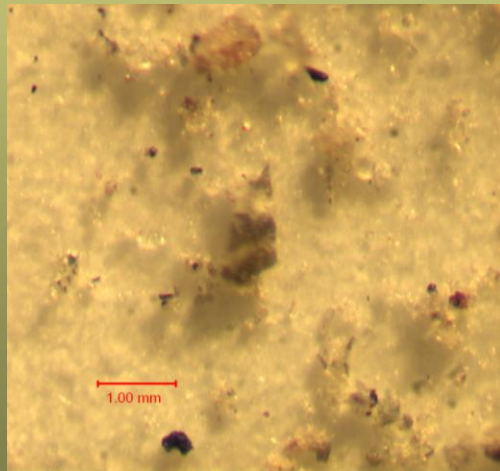
Fluorescent
Microscopy



PLFAME Analysis



Ft. Irwin



Ultrastructure

Biological Structure

Microbial Community
Composition

Discussing the Problem





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