# IN WATER-DAMAGED BUILDINGS

- SHOULD WE LOOK
FOR

"HIDDEN TREASURE"





# CONFLICT DECLARATION AUTHORED & PRESENTED BY: DAVID LARK

DIRECTOR

&

PRINCIPAL MYCOLOGIST

# NSJ ENVIROSCIENCES PTY LTD NO OTHER CONFLICTS TO DECLARE





### The Challenge ... Remains

To use Evidence Based Science to Enable Evidence Based Medicine To Answer the Burning Question:

WILL THESE BUILDINGS
MAKE YOU SICK?



### FLOODS IN OZ...

#### **Devastation at Scale Never Recorded**

90% OF AUSTRALIANS LIVE ON A COASTAL PLAIN

< 25KM FROM THE OCEAN

#### IS CLIMATE CHANGE REAL?



#### WHAT WAS THE CAUSE?

#### Severe East Coast Low: Mar/Apr 2022

### SOME AREAS IN CATCHMENTS FOR RIVERINE LOWLANDS AND DELTAS RECEIVED

> 1 Metre (~40 inches) Rainfall in <24 hours

RESULTED IN RIVER FLOOD PEAKS > 2 M ABOVE PREVIOUSLY RECORDED LEVELS

"CLUSTER FLOODING" OCCURRING IN MANY AREAS



### NOTALL ABOUT FLOODS

Massive five-metre waves batter Sydney's beaches



# HIGH SEAS ALSO COINCIDED WITH FLOODING

## ECL FROM SPACE







#### **Aerial View of "Lake" Woodburn**

WOODBURN WAS OUR HOMETOWN FOR MORE THAN 2
DECADES (1990 – 2010)
MY SON AND HIS FAMILY SOLD THEIR PROPERTY
THERE AND MOVED TO PERTH. WA
3 WEEKS AHEAD OF THESE FLOODS



### OUR HOME 1990-2010



16 CEDAR STREET, WOODBURN
10 HOMES FROM THE RICHMOND RIVER



### SON's HOME 1999-2022



AT SWAN BAY ACROSS FROM THE RICHMOND
RIVER
ON A KNOWL - NO PREVIOUS ELOOD IN

ON A KNOWL – NO PREVIOUS FLOOD IN HOUSE DURING PREVIOUS 120 YEARS

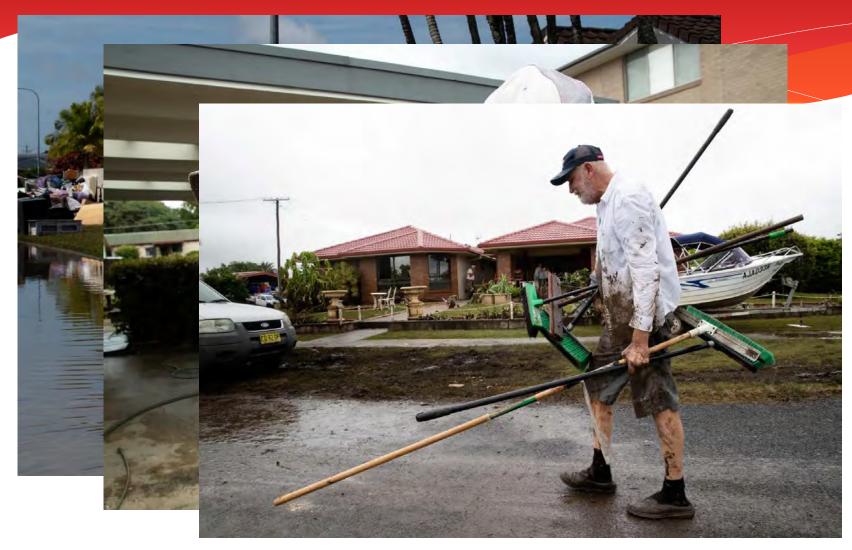


# FLOOD IMAGES





### HUMAN SUFFERING





# POLLUTION, PLUS

#### It Never Rains but it Pours!!!

At least 4 Secondary Sewage Treatment Plants
Carcasses of Dead Native Animals & Livestock
Agrichemicals, inc. Fertilizers, Pest & Herbicides
Sugar in Storage at 3 Mills
Diesel/Fuel from Storage & Submerged Vehicles

# DILUTION IS THE SOLUTION TO POLLUTION



### HIDDEN TREASURE

#### **BACK TO THE ENVIROBIOME...**

FOCUS OF NGS SO FAR – mostly on Actinobacteria

Actino Score
Dominance Index
Prevalence Index

To find the HIDDEN TREASURE you need a map -

more on this in a moment – but first lets look at the treasure revealed so far....



### NGS ADVANCES

#### **CAN NGS RELIABLY I.D. & COUNT ?:**

- \* FUNGI
- \* ACTINOBACTERIA
- \* OTHER BACTERIA



## ACTINOS & CIRS

Diagnostic Process for CIRS: Internal Medicine Review, Volume 4, Issue 5 May 2018

A Gene Primer for Health Care Providers: The Genomics of CIRS and Associated Molecular Pathways: Interpreting the Transcriptomics Results: Shoemaker R, Ryan J. eBook. 3/1/2018.

Medically sound investigation and remediation of water-damaged buildings in cases of CIRS-WDB. <u>Part 1. Oct.</u> <u>15 www.survivingmold.com.</u>

Accessed 4/20/2018.



# Actino Score

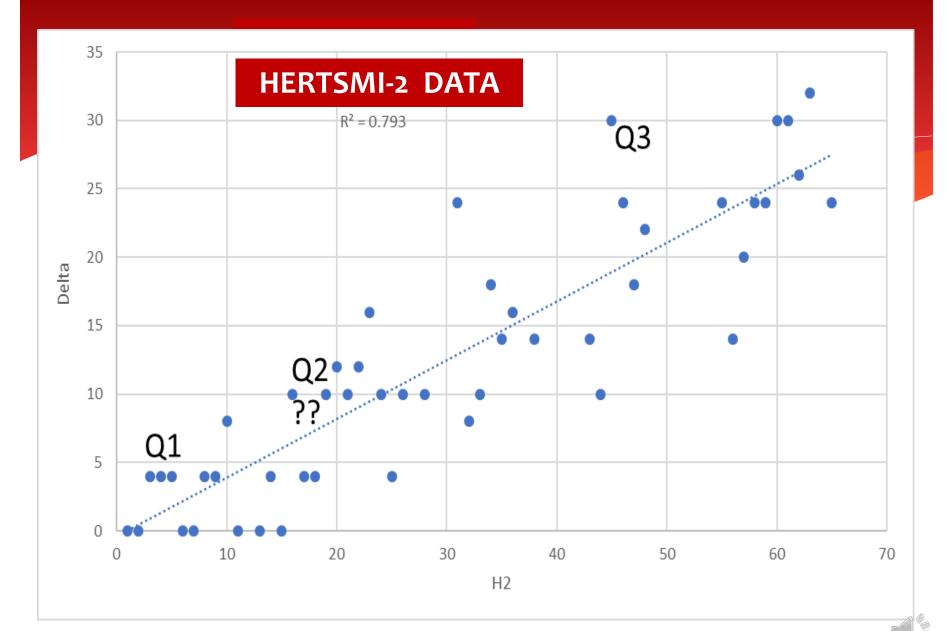
DERIVED FROM BACTERIAL ABUNDANCE/DIVERSITY MATRIX

0 -15 Normal

16 - 20 Further Investigation

> 20 Elevated





# Significant Findings

# 87% of Bacteria detected were Known Pathogens

45% were Actinobacteria

10% were Thermoactinomycetes

42% were unaccounted for -

? Worth further investigation



- Residence in Tract Development near Farmland
- Flooded Apr. 2015
- Occupant & Family Unable to Remain in Residence
- Assessed by Numerous IEPs, OHs, etc at least 10 !!!!!
- Disagreement as to Causation Flood or Shower Leaks?



# What's Your Vote?





VIA

В	LE	CULTURE	Bacteria cfu/plate	Acremonium spp.	Atternaria spp.	Aspergillus spp.	Aspergillus niger	Ascospores	Chaetomium spp.	Cladosporium spp.	Basidiospores	Chrysonifia spp.	Dreschlera spp.	Eppicoccum spp.	Fusarium spp.	Geomyces spp.	Paecilomyces spp.	Penicillium spp.	Scopulariopsis spp.	Cunninghamella	Stachybotrys spp.	Trichoderma spp.	Zygomycetes	Yeasts	Mould cfu/plate
	//	Kitchen: Excised baseplate - outside wall	5			50												2							52
	23	Lounge: Excised base plate - outside wall	6			>100				1						1				14					>116
	24	Studio: Excised base plate - outside wall	20			>100				1					1										>102
		Bedroom#1: Excised base plate - eastern outside wall	3		1	25																			26
		Bedroom #3: Excised base plate - out side wall	6		1									1				8							10
			<5	0	50 -	100		0 - 50	>2	50						anted	when		mould						
			Abc	;												·100 c were d			more t	han 25	5 cfu/ <sub> </sub>	plate if	poter	ntially	
		Đ	Ecology							_								Hig							
		Rating	Normal Mould E			=le vated	3	= 50 E	1	, E						nould ( e moul								the s	urface.
			<u>_</u>		i	Ĭ	-		3	, e i y								ery l	-						
			Norm								was		nt on t	hese	surfac	ons ex es an									

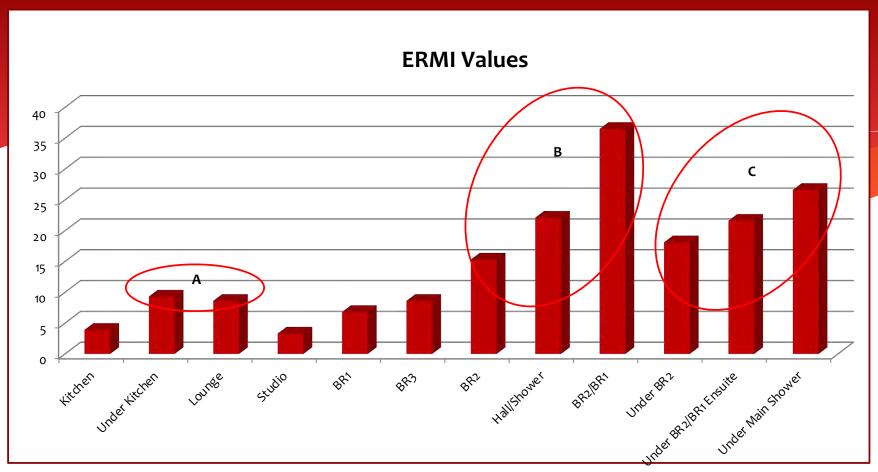


T/	_	PE LIFTS	Mould/cm^2	Area Counted %	Fungal Hyphae	Un-Id Fungal Spores		Gen Dirt & debris	Acremonium spp.	aria spp.	4 <i>scospores</i>	4s pergillus/Penicillium	Basidio spores	Bipolaris/Dreschlera	Chaetomium spp.	Cladosporium spp.	laria spp.	Еррісоссит spp.	Fusarium spp.	Scopulariopsis spp.	Pithomyces spp.	spp.	Stachybotrys spp.	Jiocladium spp.	4.ureobasidium.
	•	PE EIF13	Mould	Slide	Fungal	Un-ld F	Pollen	Gen Di	Acren	Affernaria	Ascos	Asper	Basidii	Bipola	Chaet	Clado	Curvularia	Eppico	Fusar	Scopu	Pithon	Torula spp.	Stach	Ulocia	Aureo.
6	;	Kitchen: Outside wall	1	50	1			LM								1									
7	,	Kitchen: Kitchen / lounge wall	5	50				LM			1												8		
8	;	Kitchen: Under excised base plate - outside wall	>2992	5	6	20		LM		5	3	>500	2			1		2				1			
9	1	Lounge: Lounge / kitchen wall	5	50	2			LM			1	5		1											
1	0	Lounge: Lounge / outside wall	BDL	50				LM							В	elow Lin	nit of D	)etecti	on						
1	1	Lounge: Excised base plate - outside wall	>6283	5	>100	31		MH			2	>1000						1							
1	2	Studio: Outside wall	2	50				LM		1		2				1									
1	3	Studio: Studio / hall wall	1	50	1			LM							1					ĺ					
1	4	Studio: Excised base plate - outside wall	>4898	5	>100	25		IWH			4	>750	5												
1	5	Bedroom#1: Bedroom 1 / studio wall	BDL	50				LM							В	elow Lin	nit of D	)etecti	on						
1	6	Bedroom#1: Outside wall	>6399	5	>100			M				55				>1000									
1	7	Bedroom#1: Windowsill - eastern side	>7778	5	>100			н		4		>100				>1000				ĺ					>200
1	8	Bedroom#1:Excised base plate - eastern outside wall	>4931	5	>100	20		MH		3	8	>750	2				1	4				2			
1	9	Bedroom#3: Outside wall	9	50	3			LM		1						12									
2	0	Bedroom#3: Bedroom3 / hall wall	1	50	1			LM																	
2	1	Bedroom#3: Excised base plate - outside wall	>6820	5	>200	15		н				>1000	12					4							
		Lower limit of detection = BDL 1 mould/cm2 @ 50%	<5	0	<5	00	3.3	00 -	100	0 - 5	000	>50	000			stigation				nould s	-	- hypha	ae wen	e detec	ted on
					3	έĥο								surfac	es at o	concentra	ations	greate	r than :	_	r.				
					3	5								Where	e the tr	otal surfa	ce son	re and	hypha	l conce	entratio	n was:	above	1000/∩	m²
		5			"	3	1	ed		_		3	5	ı		may hav									
		Rating	Low		Mould Evelon			=levated		Ę		3	very mign	The ca	ause a	nd sourc	e of the	e moul	d shoul	ld be d	etermin	ed and	d redre	ssed.	
		_			-	=	'	ш				>	Š						Very	High					
					8	=										0:Y31e s									
					2	2										ctive mo				hese su	urfaces	and re	emedia	tion to	
					<u> </u>									remov	e the r	nould gro	owan is	requir	ed.						



SP	2	0	RE TRAPS	Mould/M^3	Slide Area Counted %	Flow Rate I/min	Sample Time Minutes	Spores & Hyphae Counted	Fungal Hyphae	Un-Id Fungal Spores	Pollen	Gen Dirt & debris (H,M,L)	Acremonium spp.	Alternaria spp.	Ascospores	Aspergillus/Penicillium	Basidiospores	Bipolaris/Dreschlera	Chaetomium spp.	Cladosporium spp.	Curvularia spp.	Eppicoccum spp.	Fusarium spp.	Nigrospora spp.	Stemphylium spp.	Trichoderma spp.	Stachybotrys spp.	Scopulariopsis spp.	Spegazinnia spp.
		1	Outside air - Footpath (western end)	4053	25	15	5	76	11			M		6	7		5			36	3	1		5					2
	L	2	Kitchen	4427	25	15	5	83	17	4		MH		4	2	18	1		1	26		3		7					
	L	3	Lounge	3733	25	15	5	70	12	4		MH		5	3	6	3	1		27	2	3		3			1		
	L	4	Studio	4373	25	15	5	82	13			M		2	3	28		2	2	29	2	1							
	L	5	Bedroom #1	3733	25	15	5	70	10			M		2	2	21				20		3		1				11	
	L	41	Bedroom #3	3733	25	15	5	70	10			M			5	24				31									
			Lower limit of detection = BDL <53 Mould/M^3 @25%	<10	00	<1	000	1000 - 4	225		25 - 000	>100				•		warrar		hen n	Eleva nould			yphae	were	detec	ted in	the air	r at
							λ <sub>e</sub>							conce	entrati	ons gr	eater	than 1	000/N	۲.									
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			Rating	Low		-	Normal Mould Ecology	Elevated		2	L 6	Verv Hiah			nave b			ould sp t. The		e & so	ource	of the	moul						ould
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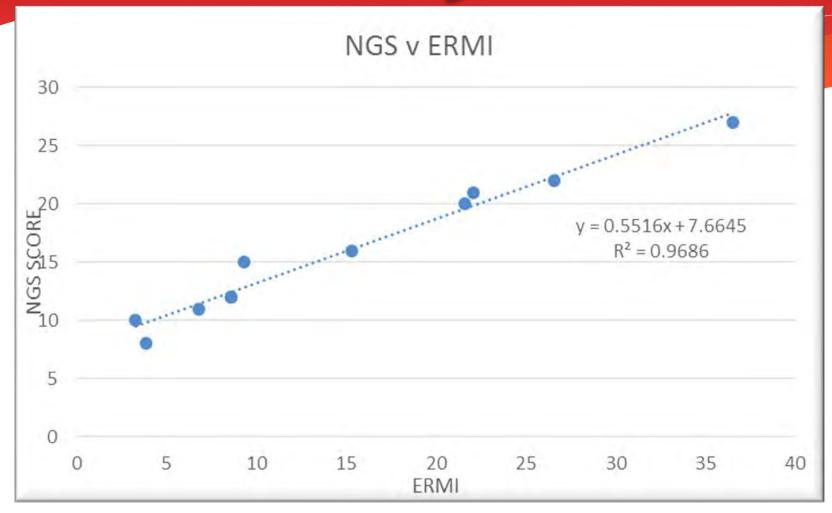
A: Kitchen end of house – moderate rise in mould indicated by ERMI - thought to be the point where flooding entered the subfloor and accumulated against the foundation wall.

B: Bedroom/bathroom end of house – where moisture from the leaking showers has allowed extensive mould amplification as indicated by the elevated ERMI's

C: Underfloor samples taken from visible mould present on the underside of the particleboard flooring adjacent to areas of water damage.



# Case Study 1-NGS



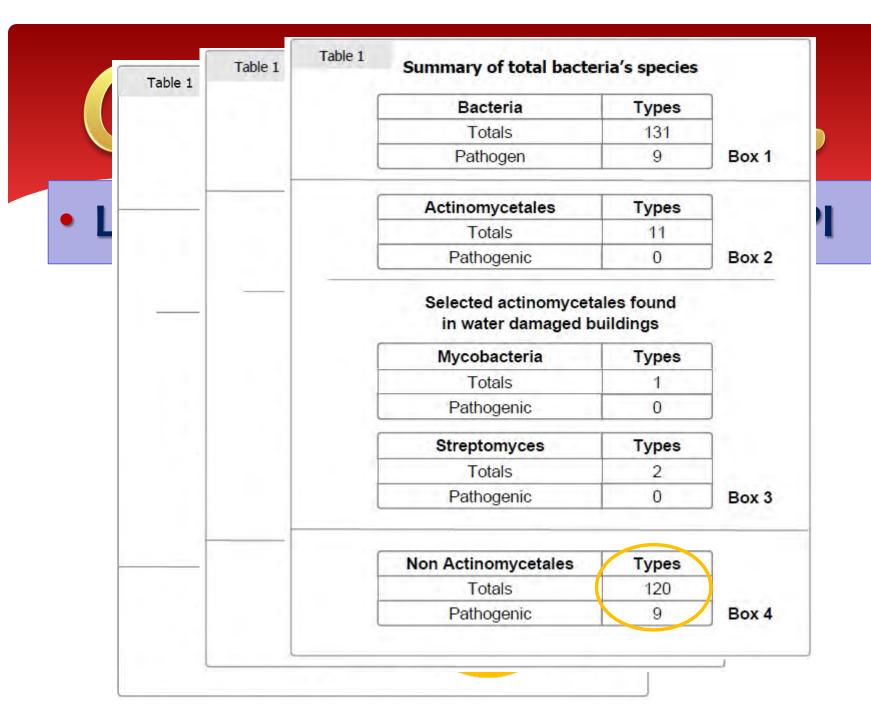


Between Baseplate &	Tape Lift		
Floor	Mold/cm2	ERMI	Predominant Molds
	>500 Asp/Pen		Asp. ochraceus
Kitchen	5 Alternaria 2 Epicoccum	3.9	Asp. restrictus Cl. sphaerospermum
	>1000 Asp/Pen		Asp. restrictus Asp. versicolor
Lounge	1 Epicoccum	8.6	E. amstelodarmi
Studio	>750 Asp/Pen	3.3	Wallemia sebi Asp. restrictus
Studio	2730 Asp/Pell	3.3	Wallemia sebi
	>750 Asp/Pen		Asp. ochraceus
BR 1	3 Alternaria	6.8	Asp. restrictus
	4 Epicoccum		Asp. versicolor
	р.сссси		Wallemia sebi
	>1000 Asp/Pen		Asp. ochraceus
BR3	=	8.6	Asp. restrictus Asp. versicolor
	4 Epicoccum		Wallemia sebi
			Asp. ochraceus
BR2	>1000 Asp/Pen	15.3	Asp. restrictus
	100071071	10.0	Asp. versicolor
			Asp. versicolor
Hall/Shower	>1000 Asp/Pen		Cheatomium globosum
Wall	4 Cheat.	22.1	Stachybotrys chatarum
			Wallemia sebi
			Asp. ochraceus
			Asp. restrictus
			Asp. sydowii
			Asp. unguis
			Asp. versicolor
BR2/BR1	>1000 Asp/Pen		Cheatomium globosum
Ensuite	4 Cheat	36.5	E. amstelodarmi
Liisuite	12 Stachy		Pen. crustosum
			Pen. purpurogenum
			Scop. bevicaulis
			Scop. chatarum
			Stachy. chatarum
			Wallemia sebi

### IMMARY

**NGS - All samples** under baseplates NGS > 20, predominantly **Actinos, including** Saccharopolyspora rectivirgula Hypersensitivity pneumonitis -"dust" Dx -Farmer's Lung







### "HIDDEN TREASURE"....

Table 4

#### **Pathogenic Non-Actinomycetales**

BE/mg= Bacteria's equivalent / milligram of sample

amilie	es Genus	Species	BE/mg**
Bacill	aceae		
1	Bacillus	cereus	180
2	Bacillus	infantis	62
3	Bacillus	megaterium	133
Burkh	elderiacea		
Burkh 4	e <del>lderi</del> acea Burkholderia	fungorum	19, 867
4		fungorum	19, 867
4	Burkholderia	<b>fungorum</b> botulinum	<b>19, 867</b>
4 Clos	Burkholderia  tridiaca  Clostridium  Clostridium	botulinum cadaveris	55
4 Clos	Burkholderia  tridiaco  Clostridium	botulinum	152



### DATABASE SEARCHES





Search PMC Full-Text Archive

Search

Journal List > Nucleic Acids Res > v.35(Database Issue); 2007 Jan > PMC1751540

OTH

 Database Automation – Examples of Significant Microbial "Isolates" were to follow, but ......

Nucleic Acids Res. 2007 Jan; 35(Database issue): D347-D353.

Published online 2006 Dec 1, doi: 10.1093/nar/qkl947

PMCID: PMC1751540

PMID: 17145713

The National Microbial Pathogen Database Resource (NMPDR): a genomics platform based on subsystem annotation

Leslie Klis McNeil, 1,\* Claudia Reich, 1,2 Ramy K. Aziz, 3 Daniela Bartels, 4 Matthew Cohoon, 4 Terry Disz, 4
Robert A. Edwards, 5,6,7 Svetlana Gerdes, 5 Kaitlyn Hwang, 4,8 Michael Kubal, 4 Gohar Rem Margaryan, 4 Folker Meyer, 4,8

William Mihalo, 4 Gary J. Olsen, 2 Robert Olson, 4 Andrei Osterman, 5,7 Daniel Paarmann, 4 Tobias Paczian, 4

Bruce Parrello,<sup>5</sup> Gordon D. Pusch,<sup>4,5</sup> Dmitry A. Rodionov,<sup>7</sup> Xinghua Shi,<sup>4</sup> Olga Vassieva,<sup>5,9</sup> Veronika Vonstein,<sup>5</sup>

Olga Zagnitko,<sup>5</sup> Fangfang Xia,<sup>4</sup> Jenifer Zinner,<sup>4</sup> Ross Overbeek,<sup>5</sup> and Rick Stevens<sup>4,8</sup>

Author information
 Article notes
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 Disclaimer





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Biodegradation of Phenanthrene and Heavy Metal .... (2022). Retrieved on April 16, 2022, from https://www.frontiersin.org/articles/10.3389/fmicb.2019.00408/full.

Burkholderia fungorum Coenye et al. | ATCC. (2022). Retrieved on April 16, 2022, from https://www.atcc.org/products/baa-463.

Burkholderia fungorum DBT1: a promising bacterial strain for .... (2022). Retrieved on April 16, 2022, from https://academic.oup.com/femsle/article/319/1/11/552555.

Burkholderia fungorum Septicemia. (2022). Retrieved on April 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3371813/.

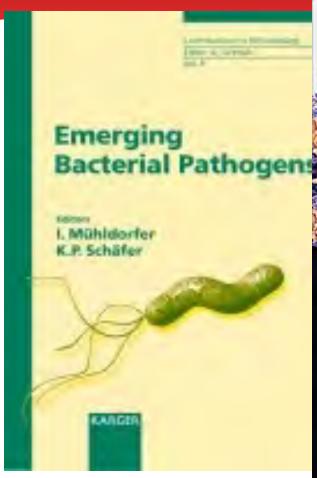
Burkholderia fungorum promotes common bean growth in a .... (2022). Retrieved on April 16, 2022, from https://annalsmicrobiology.biomedcentral.com/articles/10.1007/s13213-014-1020-y.

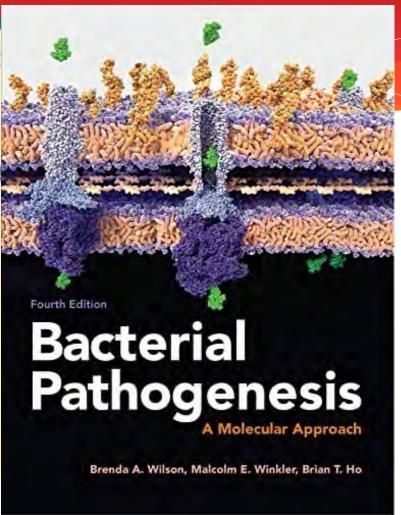
Burkholderia fungorum sp. nov. and Burkholderia caledonica sp .... (2022). Retrieved on April 16, 2022, from https://pubmed.ncbi.nlm.nih.gov/11411678/.

Burkholderia. (2022). Retrieved on April 16, 2022, from https://microbewiki.kenyon.edu/index.php/Burkholderia.



### RECENT PUBLICATIONS







# Is this REALITY?

NGS IS A VALUABLE SERVICE



- IMO THOSE WHO LIVE IN WDB (FLOODED OR OTHERWISE)
- DESEARVE TO KNOW WHAT THEY ARE EXPOSED TO SO THAT THEIR EXPOSURE
- IS NO LONGER "HIDDEN" BUT THE REALITY OF
- DETECTION BECOMES THE "TREASURE"



### Thomas' Postulates

1972

- IT'S NOT THE DOSE
- IT'S NOT THE PAMPs or DAMPs
- IT'S NOT THE INFLAMMAGENS
- IT'S NOT THE BIOTOXINS
- IT 15 "THE RESPONSE" THAT "MAKES THE DISEASE"

Lewis Thomas, MD (NEJM 1972; 287: 553-555)



### NOW and the FUTURE..

- EXPAND THE DATA DEPTH & CORRELATIONS GENIE
- EXPAND THE RANGE & SCOPE OF DATABASES ACCESSED
- BETTER BIOINFOMATICS & DATA REDUCTION
- CORRELATE WITH CLINICAL & ENVIRO. DATA (HIPAA)
- COMPARE NEXTGEN WITH TRANSCRIPTOMICS
- EXPAND NGS DIAGNOSTIC CAPABILITY TO THE MANAGEMENT OF MICROBIALLY RELA





### THANK YOU FOR YOUR

QUESTIONS
WILL BE
ADDRESSED IN
THE PANIEL
DISCUSSION

ATTENTION



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