Is the Deprivation of Natural Radiation a Biological Stress? Low Background Radiation Experiments (LBRE) at the U.S. Waste Isolation Pilot Plant (WIPP)

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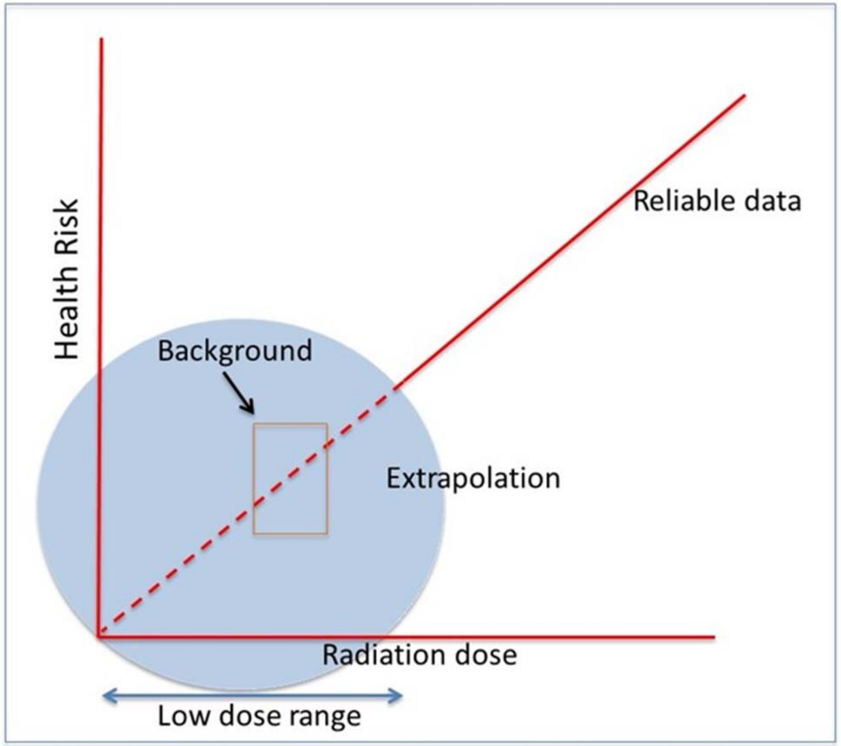
Linear No-Threshold model

"an increase in ionizing radiation exposure causes a linear increase in the accumulation of deleterious effects in organisms, with no safe level"
(ICRP 2007).



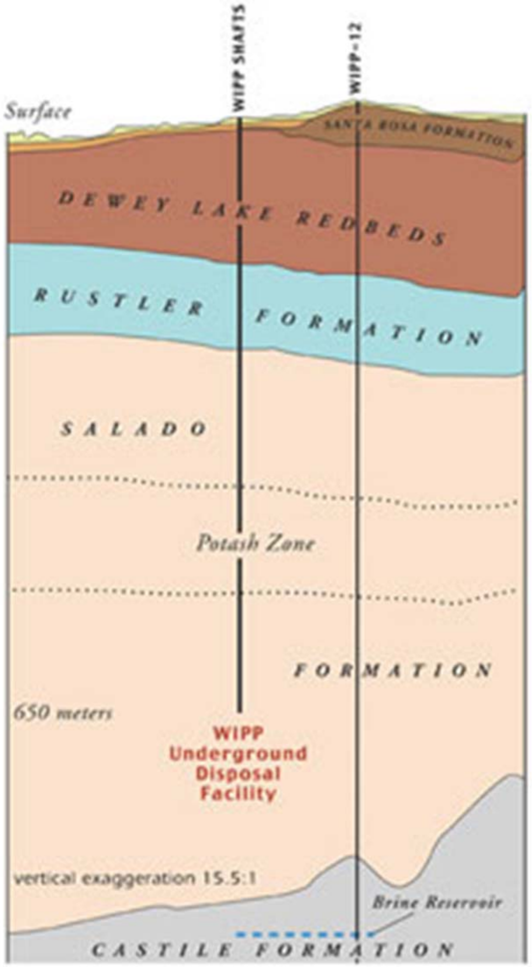
LBRE lab

- Started on 2009 after the 2006 Ultra Low Level Radiation Summit
- Initial concept by Roger Nelson, Jim Conca and Yair Grof
- Project funded by DOE
- In 2009, no lab space available underground
- BSL-1 laboratory established in 2010

LBRE research questions

- **Are organisms capable of sensing below-background levels of radiation?**
- **If they are capable, what is the response? Is it a stress response?**
- **Is this response different among prokaryotes/eukaryotes, unicellular/multicellular organisms?**

Why is WIPP special? What goes on at WIPP?



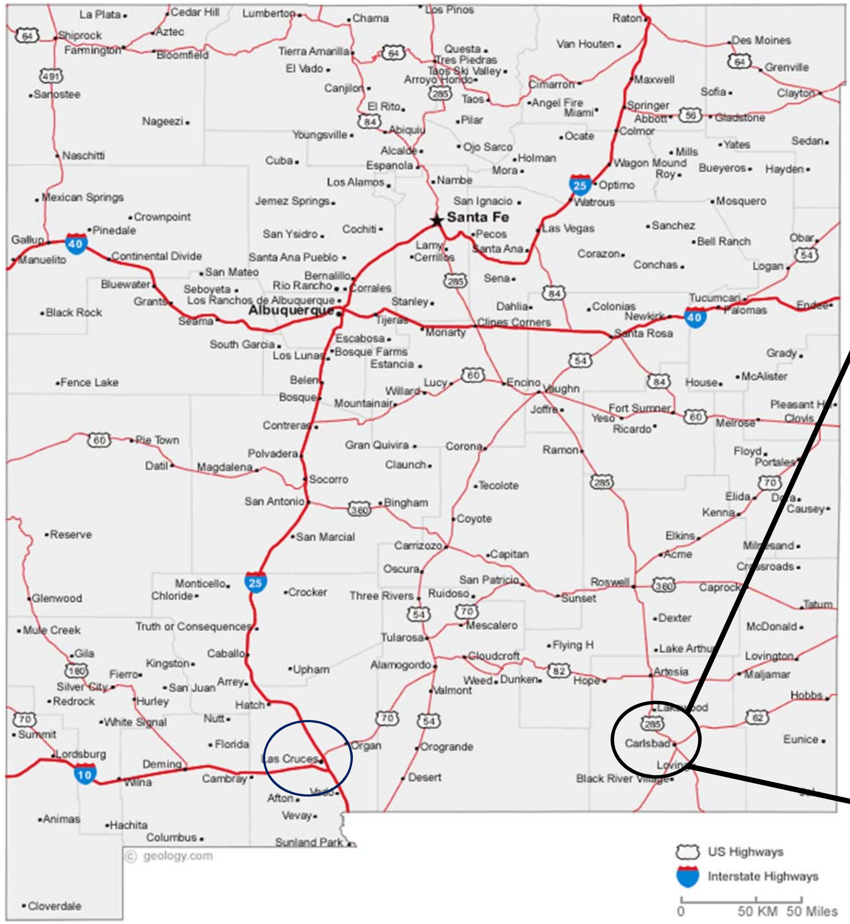
Nuclear Waste Depository

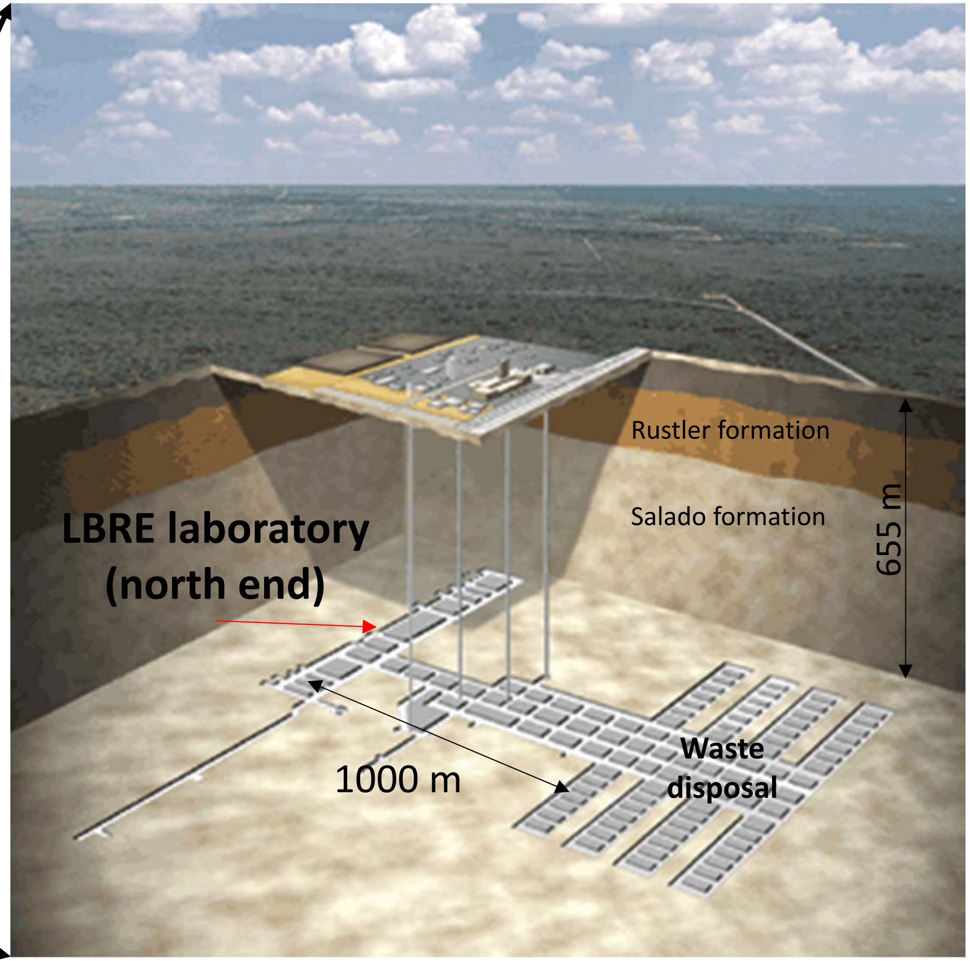
then:

- **Low Background Radiation Biology**
- **Enriched Xenon obervatory (for double beta decay)**
- **Dark matter**

Waste Isolation Pilot Plant
Carlsbad, NM

U.S. only geologic deep repository for nuclear waste





Access to the underground at WIPP

Must "brass in" and "brass out" at lamp room on the surface

Salt shaft

Waste shaft

WIPP's anatomy. They're called "back" and "ribs", not ceiling and walls!

LBRE lab @ WIPP: The challenge of doing science at a nuclear waste repository

LBRE connex-laboratory

2000 ft of shielding (with isotope "free" NaCl) reduces background dose to 5 nGy hr^{-1}. Addition of pre WWII steel vault adds shielding down to 0.2 nGy hr^{-1} (walls 15.2 cm tick)

- Limited lab space
- Access restricted to site operation hours, variable according to the site operation conditions (Summer 2015 ~12 hrs shifts, Summer 2014, no access)
- Restriction on numbers underground
- High drift temperature which challenges incubators to keep a constant 30 C.

Scenes from the LBRE WIPP lab (or how I now feel like a real biologist!)

Background controls (aboveground vs. KCl)

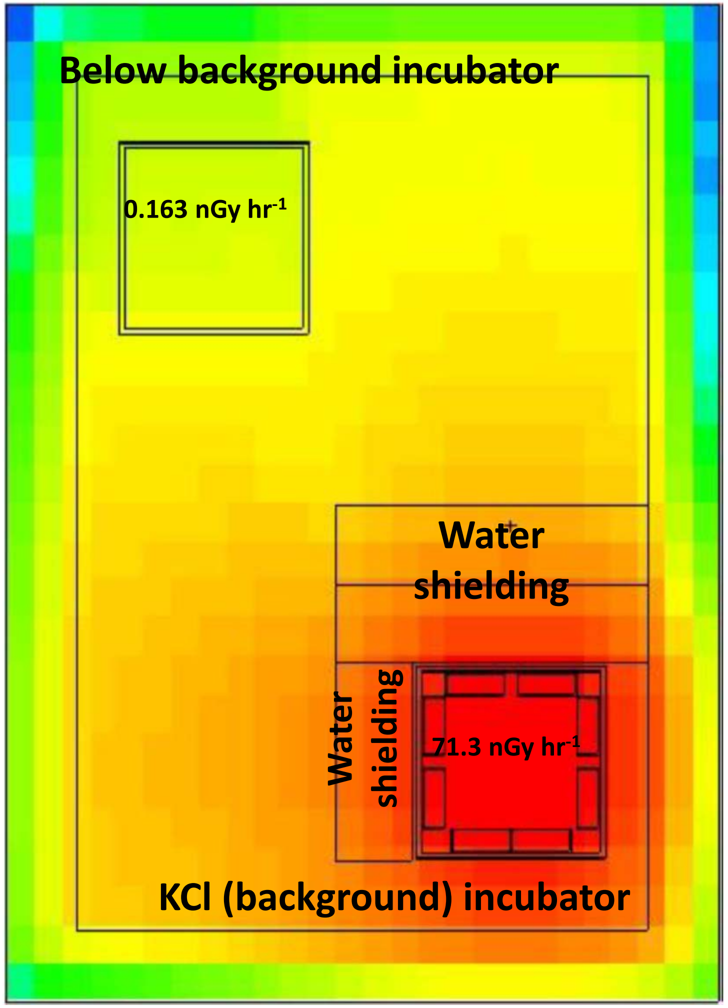
- 2009 Aboveground control
- (PI running back and forth to incubate cells and measure OD/sample (NOT very efficient or popular among WIPP personnel!).
- 2010 Belowground control at BSL-1 lab, with both incubators in the same location.

HOW???

In steel chamber, one incubator is irradiated with 11.5 kg (5 uCi) of KCl, with approximately 2 kg positioned on each of the 6 sides of the cell incubator.

The 5 uCi of ^{40}K exposes the cells to approximately 100 nGy/hr dose rate, that approximates the dose rate at WIPP's surface (measured with Sodium- Iodide detector)

Dosimetry. How “real” is our background control?



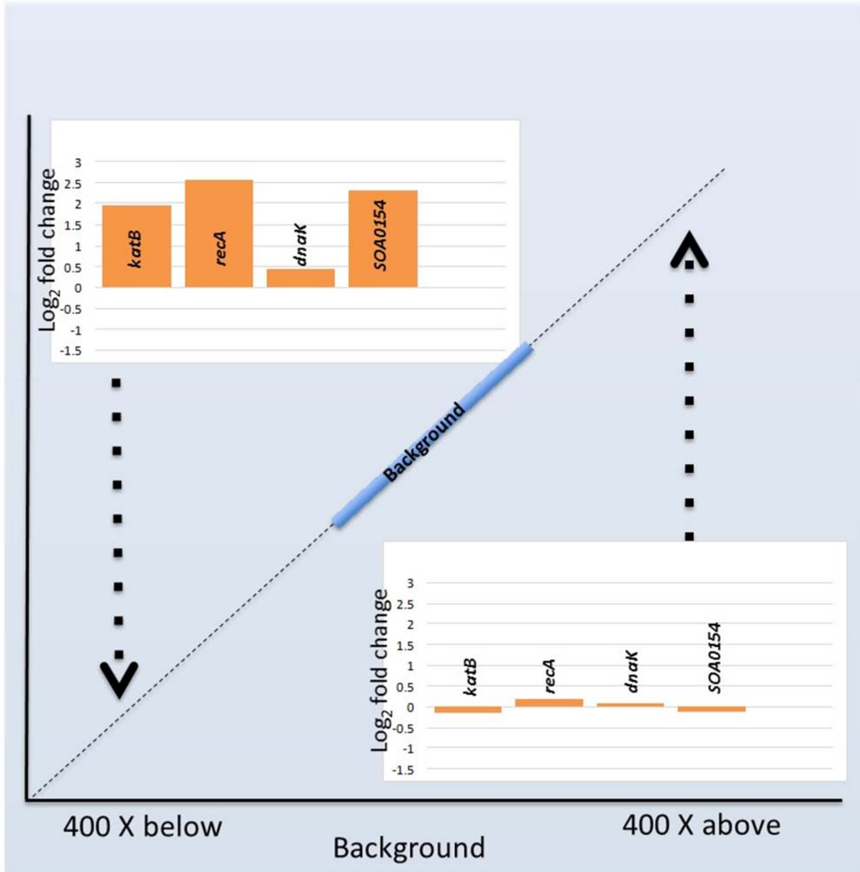
As modeled with MCNP by Jeff Wood

KCl inside the vault could irradiate the treatment incubator!

Are distance and shielding enough?

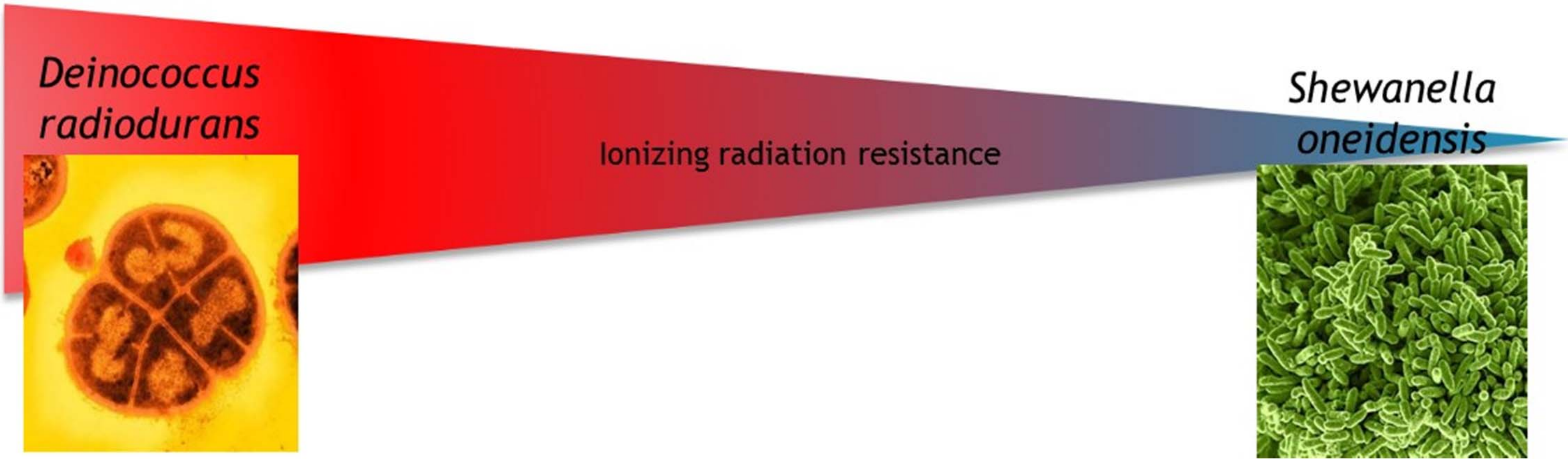
Comparing the effects at equivalent below and above background doses
(because ANY amount of radiation is bad, right?)

^{60}Co coupons irradiation "array"



Subtracting radiation more of a stress than adding some!

Prokaryotic models to study below-background radiation stress



Advantages?

- **Short generation time**
- **Amenable to relatively easy genetic analysis**
- **Taxonomically different**

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Stress induction in the bacteria *Shewanella oneidensis* and *Deinococcus radiodurans* in response to below-background ionizing radiation

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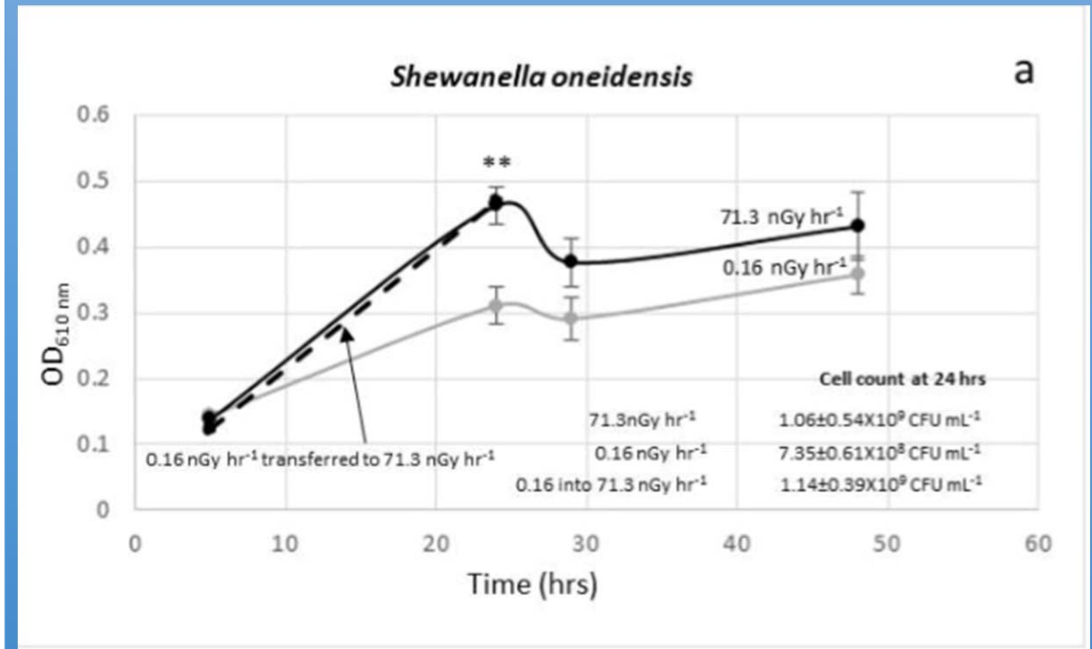
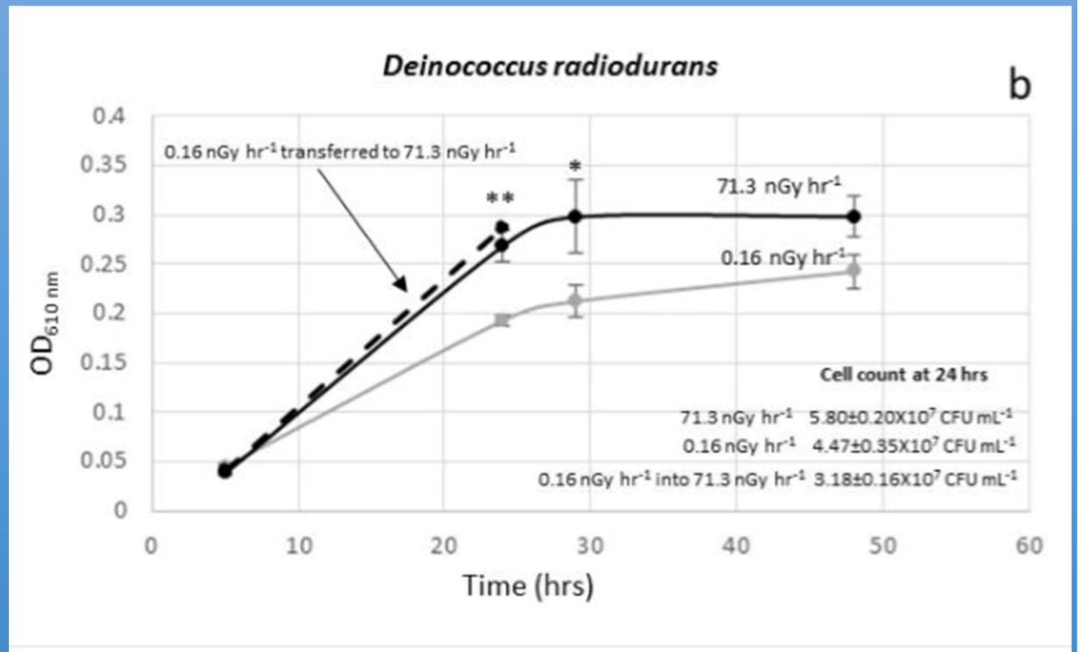
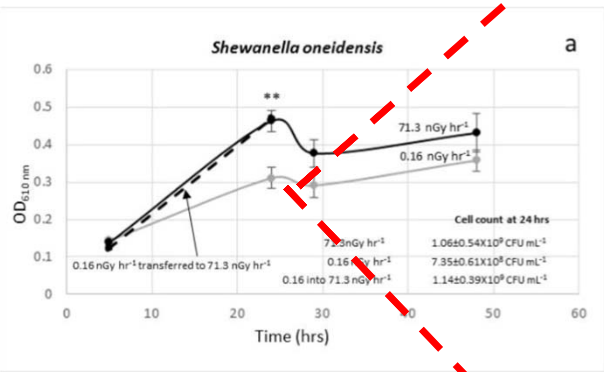
Were *S. oneidensis* and *D.radiodurans* to tell the difference?

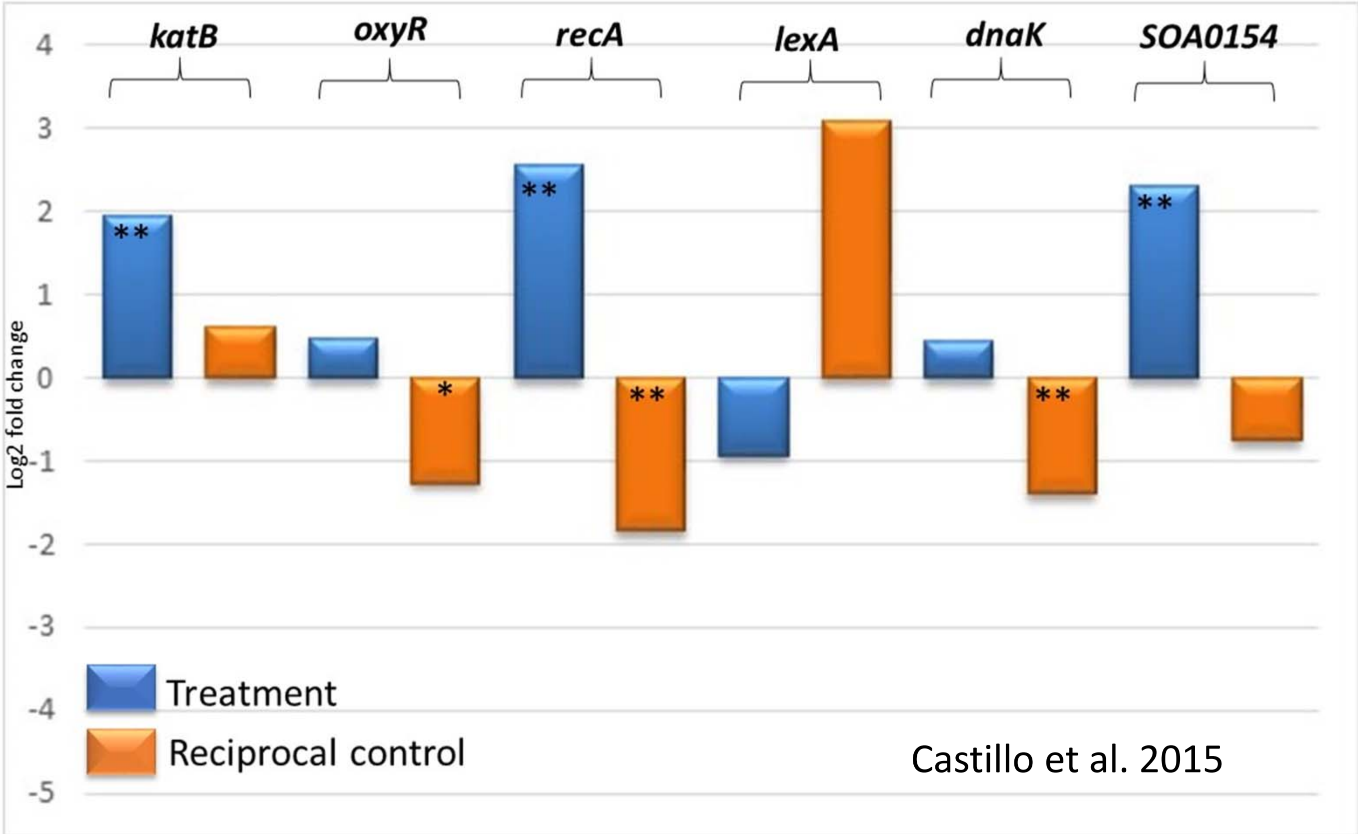




Castillo et al. 2015

What about differences at a gene expression level?

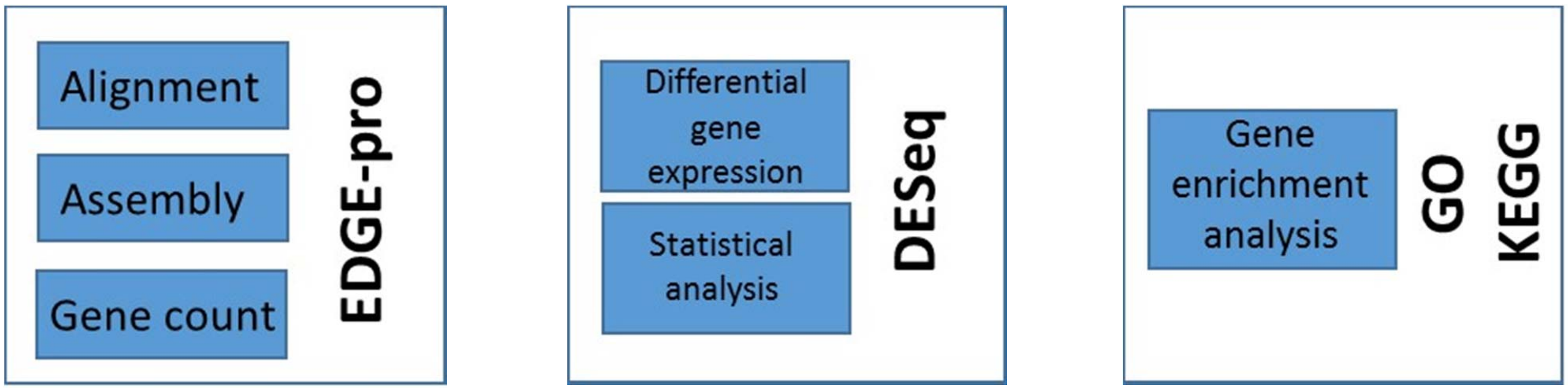




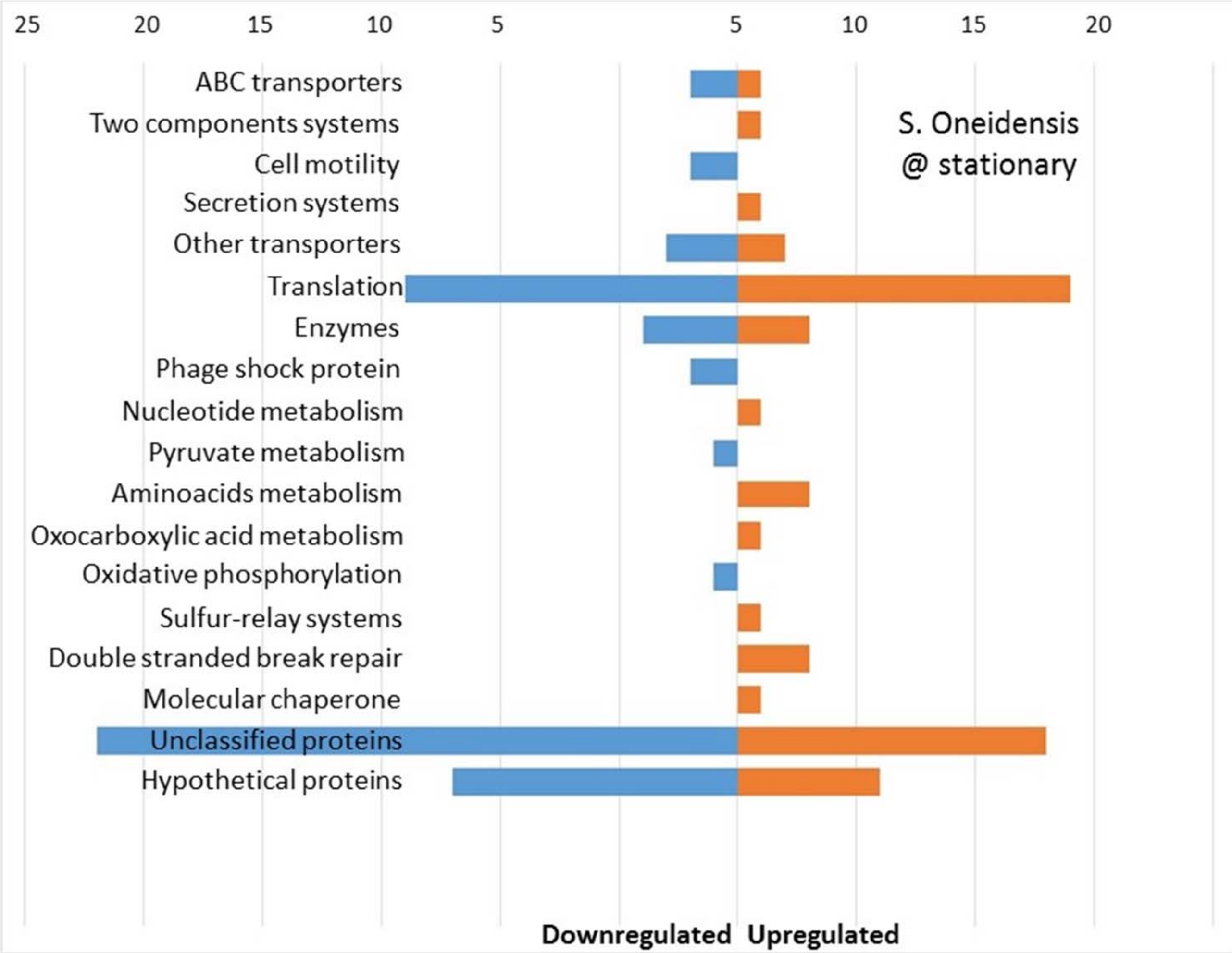
What if we look at the expression of the whole-genome?

(Initial insight to transcriptome analysis)

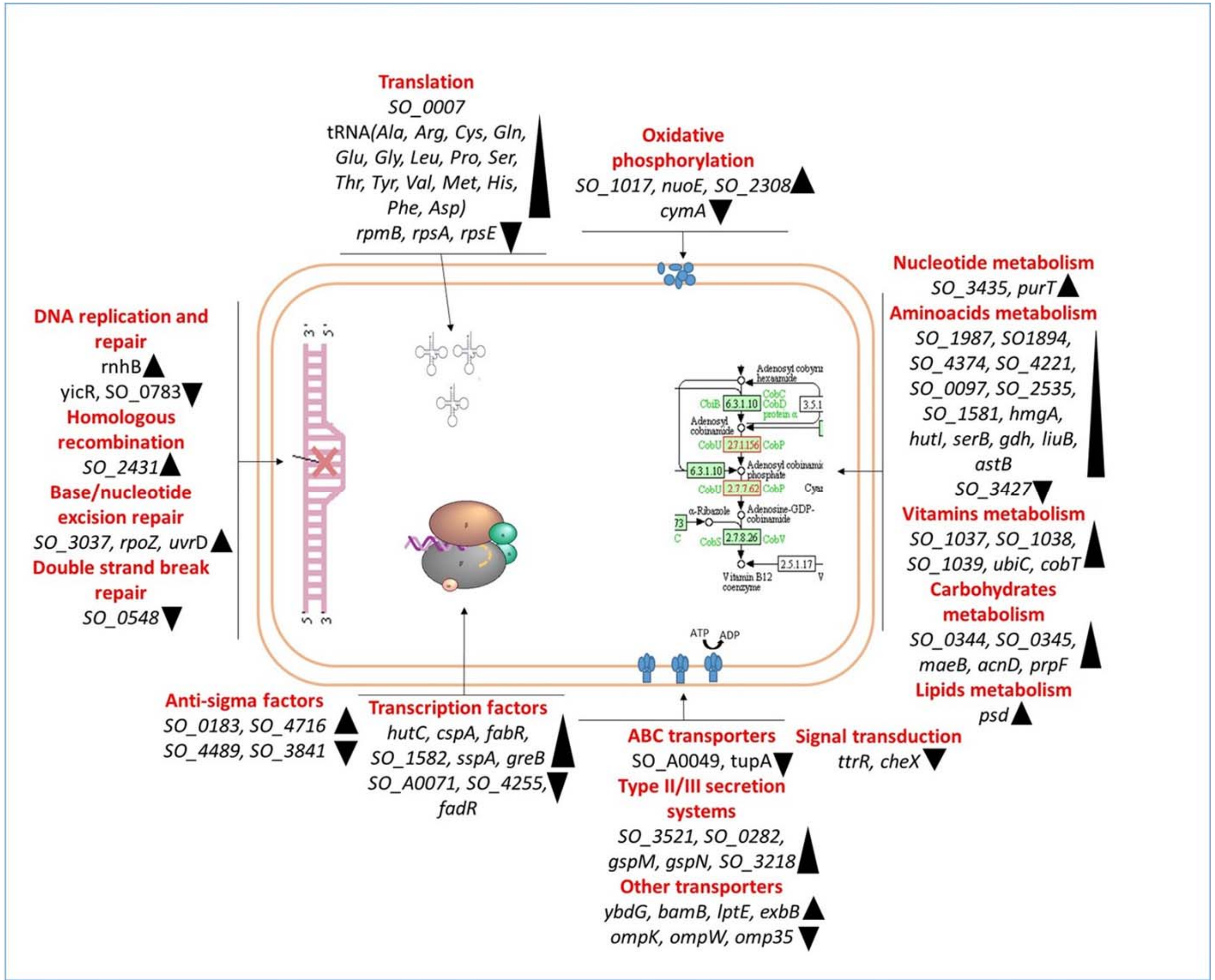
12 HiSeq Illumina libraries equivalent to one lane



Initial results from RNA-Seq transcriptome analysis….



How close are we to explain the molecular basis for what it seems to be a stress response?



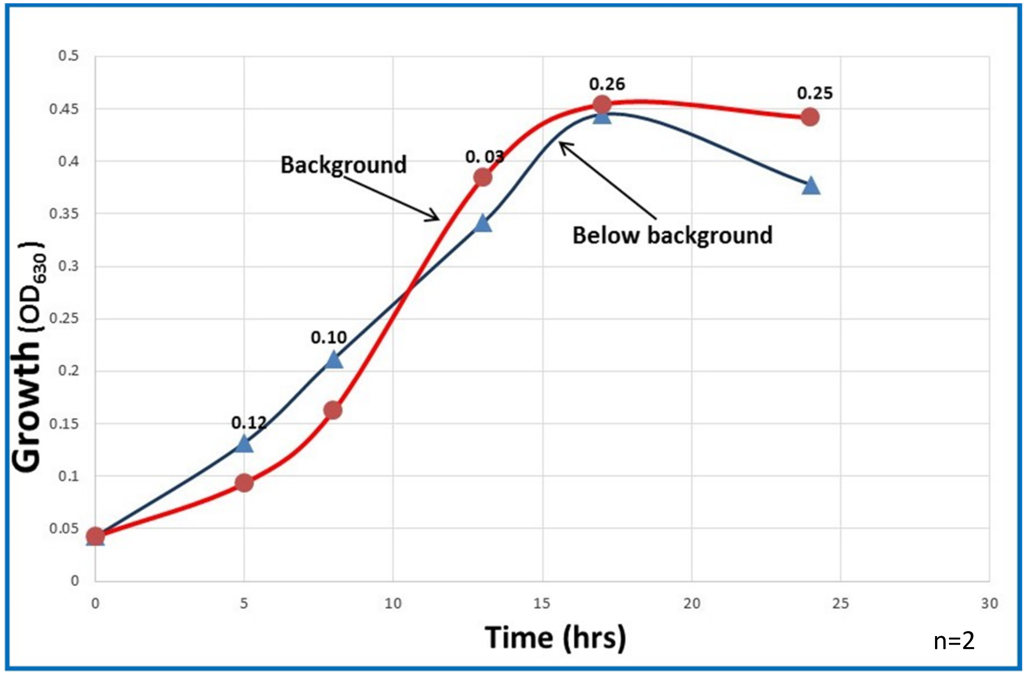
Let's take some liberties to speculate:

- Upregulation of translation and aminoacids metabolism genes suggest the need for significant protein repair.
- Substantial indication of regulation of transport processes across the membrane. What are the cells trying to dispose of?
- Upregulation of carbohydrates and vitamins metabolism genes. Need to keep up with higher energy demands?
- Regulation of DNA-repair related genes, enough to suggest DNA damage?

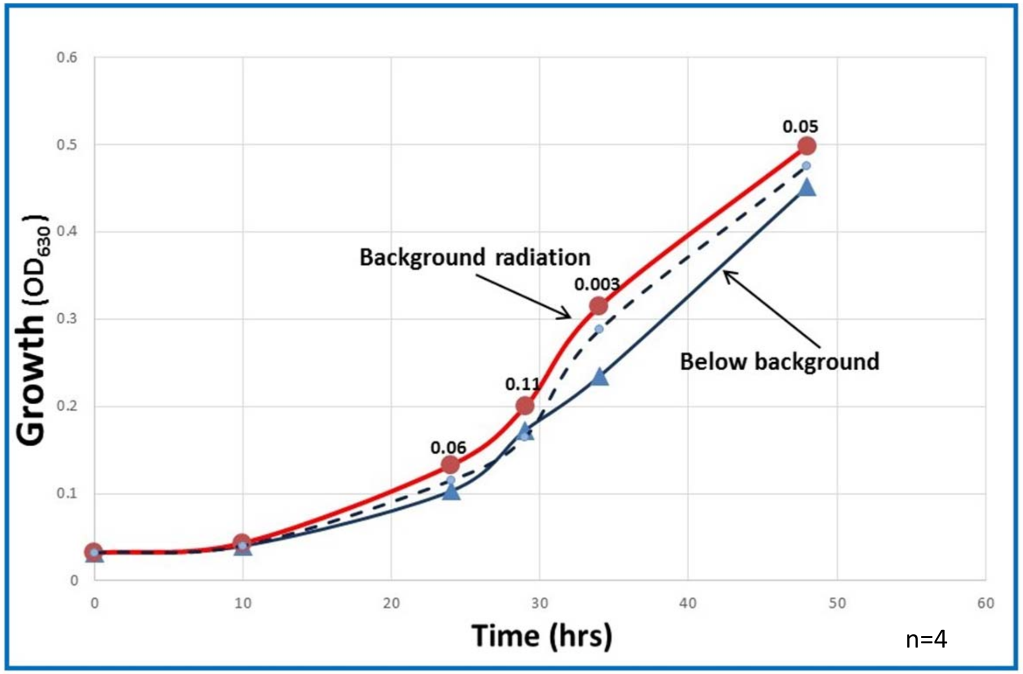
Perfecting the art of doing science under less than ideal conditions.

Initial results from Summer 2015

S. oneidensis



D. radiodurans

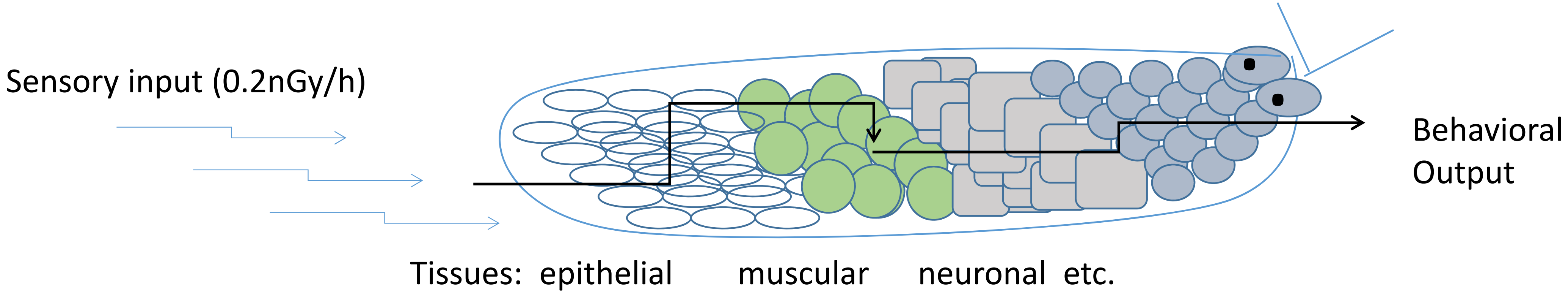


Waiting back at NMSU, final analysis of metabolites and fitness assays…….

Future research/plans

1. New research-driven hypotheses from transcriptome analysis: Expansion on challenge studies

2. **In a multicellular organism, is low radiation signal transmitted across tissues to give a behavioral output ?**



Short-short term plans: *C.elegans* (Full life cycle in only 5 days)

Medium-term plans*: Aedes aegypti* (Relatively long term exposure of adult males)

Start looking for a permanent position!

Acknowledgments

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- Initial concept: Roger Nelson, Jim Conca, Yair Grof
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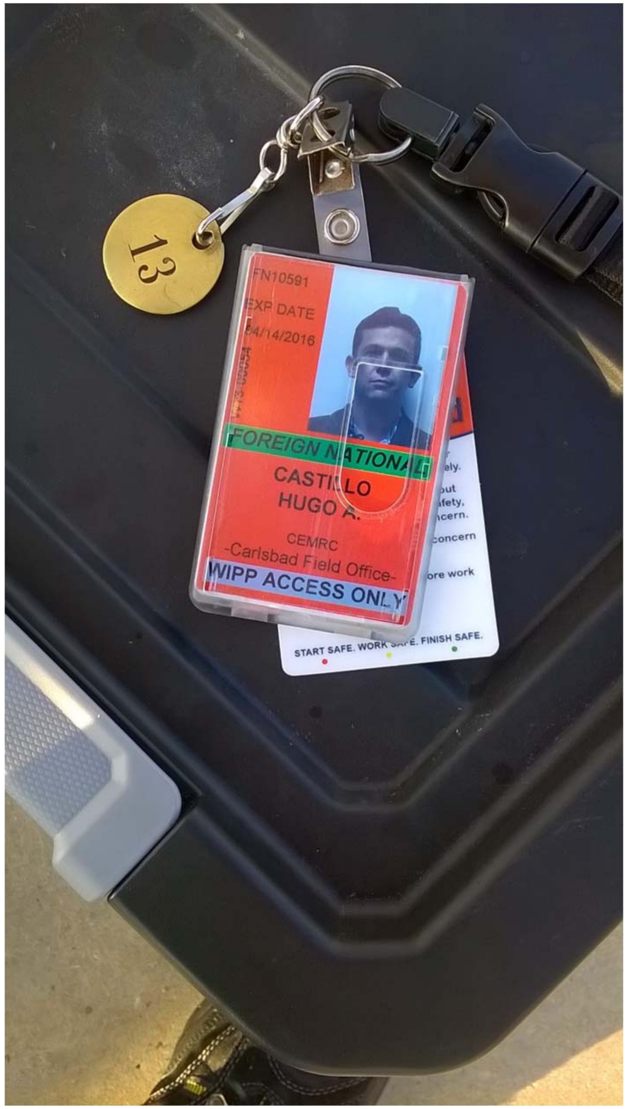
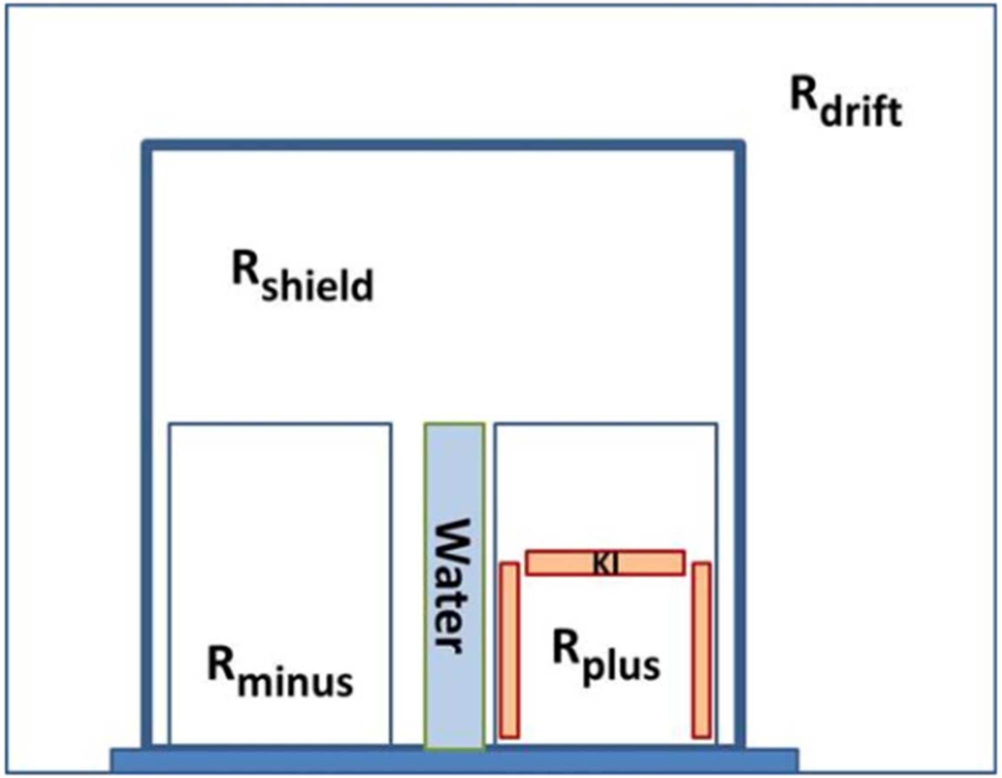
- WIPP: Robert Rempe, Ray Carrasco, Kevin McIlwee, Jeff Netherlin
- DOE: Jeffrey Wood
- LANL: Doug Weaver, Brian Dozier, Shawn Otto



Brass No. 13 out of 98? We must be doing something right!

!Gracias¡



Total dose rates		
Description	Dose rate (nR/h)	Relative Error (%)
Total dose rate in +KCl incubator (R_{plus}):	7.13e3	0.5
Total dose rate in -KCl incubator (R_{minus}):	16.3	1.0
Total dose rate in drift (R_{drift}):	234.3	0.19
Total dose rate in geometric center of LBRE vault (R_{shield}):	630	3.41

Table 9: A list of total dose rates of interest for the LBRE experiment is shown here. A diagram showing the approximate regions of interest is shown in Figure 2.

Activity of radon in drift	
Description	Value
Average Rn concentration in drift:	0.2 pCi/L
Average Rn concentration (SI):	7.4e-6 Bq/cc
Volume of modeled drift air:	1.408e9 cc
Gamma activity of radon inside modeled drift:	1.042e4 Bq
Activity of radon inside vault and incubators	
Description	Value
Average Rn concentration in vault:	7.4e-6 Bq/cc
Volume of air in vault:	5e6 cc
Gamma activity of radon in vault:	37 Bq

Abundance of U, Th, and K in a typical WIPP salt sample						
Isotope	Abundance (ppb)	Specific Activity (Bq/g)	Mass Fraction (amu/total)	Normalized Mass Fraction	Effective Total Activity Fraction	Branching Ratios for Gamma Radiation
U-238	29	1.26e4	6902	2.36e-7	2.97e-3	Complex Scheme
Th-232	78	4.20e3	18096	6.19e-7	2.60e-3	Complex Scheme
K-40	169	2.74e5	6760	2.31e-7	6.32e-2	10.75%