

Toxicity Study for Treated Produced Water from Permian Basin

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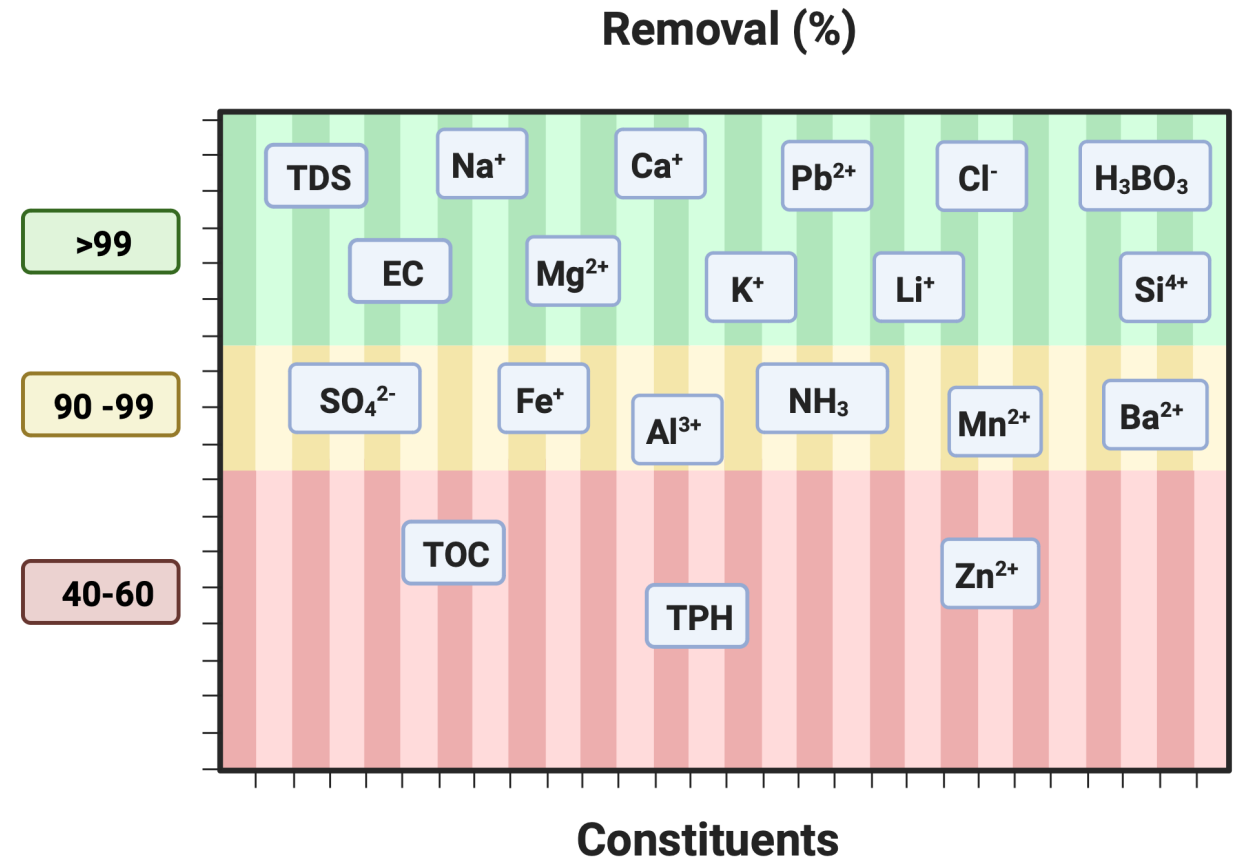
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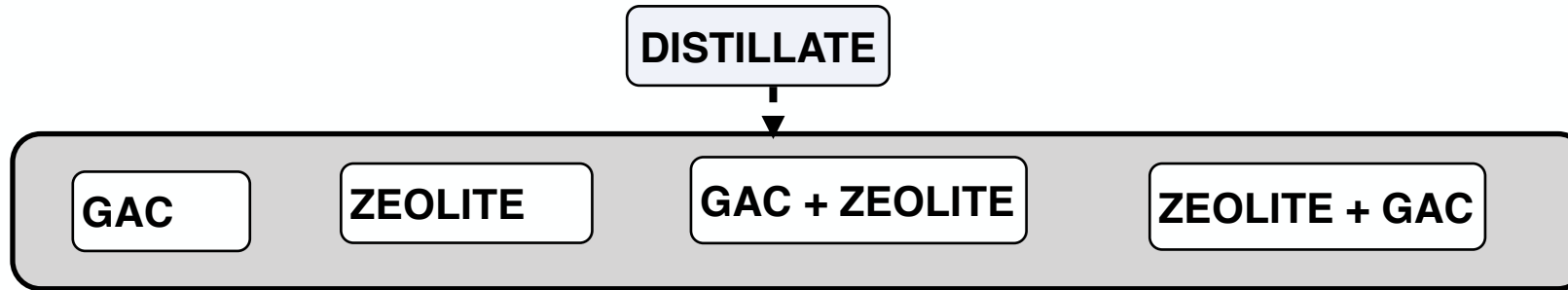
Treated PW from Pilot Scale Desalination

Low Temperature Thermal Distillation

Location	Orla, TX
Water Source	Permian Basin
TDS (mg/L)	100,000-170,000
Pretreatment	ClO ₂ and H ₂ O ₂
Desalination	Low Temperature Distillation
PW Volume	500 br/day
Water Recovery	40%
Duration	6 weeks
Cost	> 1\$/bbl

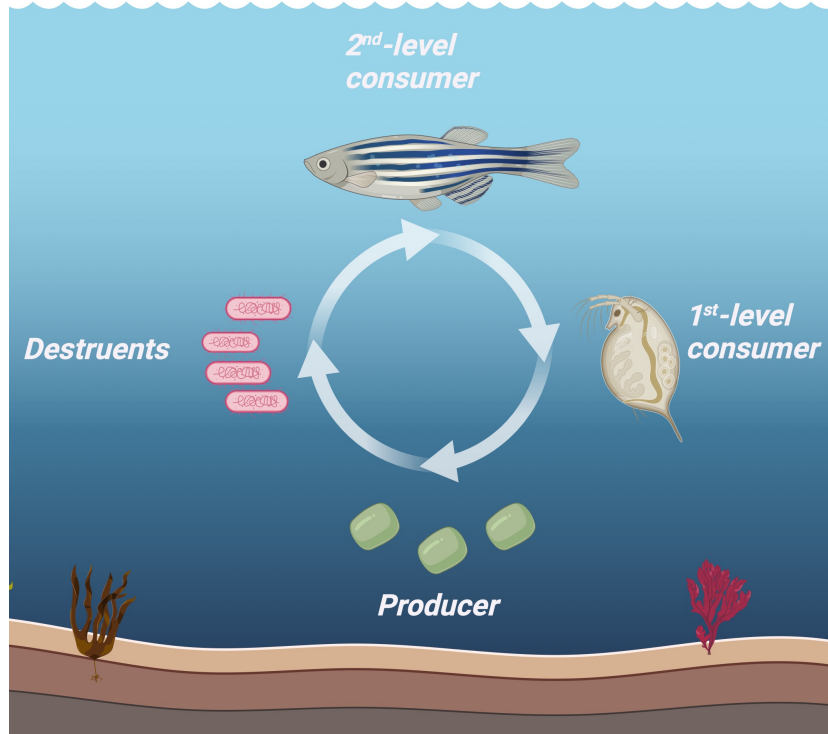


Post Treatments after Thermal Desalination in Permian Basin



POST-TREATMENTS		Target compounds					
		Ammonia		TOC		Conductivity	
1	2	Concentration	Removal	Concentration	Removal	Signal	Removal
		[mg/L]	[%]	[mg/L]	[%]	[μ S/cm]	[%]
Distillate	-	46.35 \pm 0.21	-	42.3 \pm 0.28	-	288 \pm 7.07	-
	Zeolite	0.043 \pm 0.001	99.9	45.07 \pm 3.25	0	237 \pm 2.8	17.7
	GAC	27.1 \pm 0.02	41.9	5.25 \pm 0.078	87.59	172 \pm 2.8	40.2
	GAC	0.01 \pm 0.001	99.90	5.4 \pm 0.2	87.1	171 \pm 2.8	40.60
	Zeolite	0.01 \pm 0.01	99.9	5.2 \pm 0.2	87.9	135. \pm 2.9	53.1

Toxicity Assays



Toxicological characterization					
Organism	Method	Type	Endpoint	Exposure time	Trophic level
Fish	<i>D. rerio</i> OECD 236	Acute	Mortality	96 h	2 nd -level consumer
Invertebrate	<i>C. dubia</i> EPA 1002.0	Acute	Mortality	48 h	1 st -level consumer
Algae/plant	<i>P. subcapitata</i> EPA 1003.0	Chronic	Growth	96 h	Producer
Bacteria	<i>V. fischeri</i> MICROTOX	Acute	Inhibition	15 min	Destruents

Toxicity Summary

Toxicity TEST, TST Approach	Distillate				Distillate after GAC+ Zeolite			
	IC ₂₅ or LC ₅₀	NOEC	LOEC	Test of Significant Toxicity	IC ₂₅ or LC ₅₀	NOEC	LOEC	Test of Significant Toxicity
<i>V. Fischeri</i> (Bacteria)	40.36	3.125	6.25	Fail (42.4%)	>100	100	>100	Pass (19.3%)
<i>P. subcapitata</i> (Algae)	57.95	25	50	Fail (37.9%)	>100	100	>100	Pass (5.9%)
<i>C. Dubia</i> (Water Flea)	31.86	25	50	Fail (100%)	>100	100	>100	Pass (5%)
<i>D. Rerio</i> (Zebrafish)	31.86	25	50	Fail (100%)	>100	100	>100	Pass (13.5%)

IC₂₅ (Inhibition) or IC₅₀ (Lethality);

NOEC: No Observed Effect Concentration; LOEC: The Lowest Observed Effect Concentration

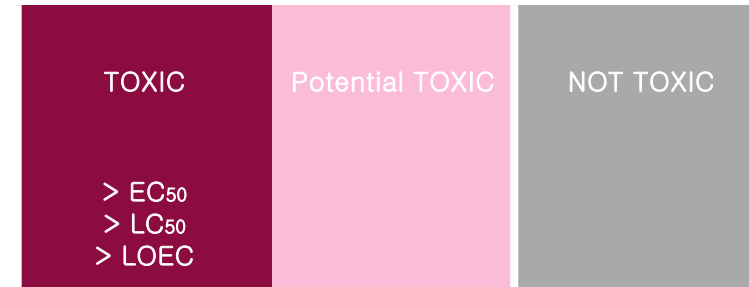
TOXICITY IDENTIFICATION MATRIX IN DISTILLATE

COMPOUNDS

TESTED: 103

IDENTIFIED: 25

Compound	Concentration (mg/L)	<i>P. subcapitata</i> (Algae)	<i>V. fischeri</i> (bacteria)	<i>C. dubia</i> (water flea)	<i>D. rerio</i> (zebrafish)
Benzene	0.501				
Toluene	0.548				
Ethylbenzene	0.0214				
Xylenes	0.377				
Acenaphthene	0.00043				
Fluorene	0.00147				
Naphthalene	0.0131				
Phenanthrene	0.00145				
Phenol	0.026				
2-Nitrophenol	0.007				
Boron	0.2102				
Barium	0.0074				
Cadmium	0.0011				
Copper	0.0095				
Chromium	0.000300				
Vanadium	0.0041				
Selenium	0.000393				
Uranium	0.0000810				
Zinc	0.0031				
NH ₃	46.35				
NO ₂ ⁻	0.0387				
NO ₃ ⁻	0.0444				



Highlights

I. **14** classified as priority pollutants under 40 CFR Appendix A to Part 423.

II. **7** determined at toxic levels.

- **4 Organics:** benzene, toluene, phenanthrene, phenol.
- **2 Metals:** cadmium and copper.
- **1 Nitrogenous:** ammonia.

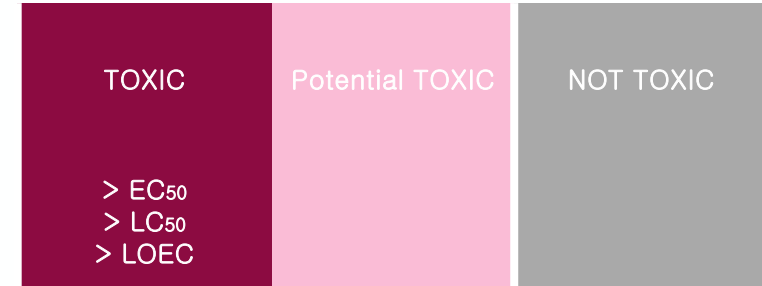
TOXICITY IDENTIFICATION MATRIX AFTER GAC POST-TREATMENT

COMPOUNDS

TESTED: 103

IDENTIFIED: 14

Compound	Concentration (mg/L)	<i>P. subcapitata</i> (Algae)	<i>V. fischeri</i> (bacteria)	<i>C. dubia</i> (water flea)	<i>D. rerio</i> (zebrafish)
Benzene	<0.000214				
Toluene	<0.000500				
Ethylbenzene	<0.000515				
Xylenes	<0.000330				
Acenaphthene	<0.00139				
Fluorene	<0.000103				
Naphthalene	<0.0131				
Phenanthrene	<0.0000866				
Phenol	0.000693	Potential TOXIC	Potential TOXIC	Potential TOXIC	Potential TOXIC
2-Nitrophenol	<0.00167				
Boron	0.1945	Potential TOXIC		Potential TOXIC	
Barium	0.0041			Potential TOXIC	
Cadmium	0.0000				
Copper	0.0055	TOXIC	TOXIC	TOXIC	Potential TOXIC
Chromium	0.003	Potential TOXIC	Potential TOXIC	Potential TOXIC	Potential TOXIC
Vanadium	0.0049	Potential TOXIC	Potential TOXIC	Potential TOXIC	Potential TOXIC
Selenium	<0.000368				
Uranium	<0.0000764				
Zinc	0.000				
NH ₃	27.1		TOXIC	TOXIC	TOXIC
NO ₂ ⁻	0.0432	Potential TOXIC			Potential TOXIC
NO ₃ ⁻	0.0782			Potential TOXIC	Potential TOXIC



Highlights

- I. **3** classified as priority pollutants under 40 CFR Appendix A to Part 423.
- II. **2** determined at toxic levels.
 - **1 Metals:** copper.
 - **1 Nitrogenous:** ammonia.

Conclusions

- Volatile/semivolatile organic compounds, ammonia and copper are the main constituents contributing to aquatic toxicity in distillate
- After GAC, ammonia and copper can still cause toxicity.
- *Danio rerio* (zebrafish) is the most sensitive organism for contaminants in treated PW

Human Cell Line Toxicity Study

- Human cell lines used

Name	Description
MCF-7	a human breast cancer cell line with estrogen, progesterone, and glucocorticoid receptors
HEK 293	a specific immortalized cell line derived from an aborted fetus or human embryonic kidney cells
Caco-2	an immortalized cell line of human colorectal adenocarcinoma cells. It is primarily used as a model of the intestinal epithelial barrier.

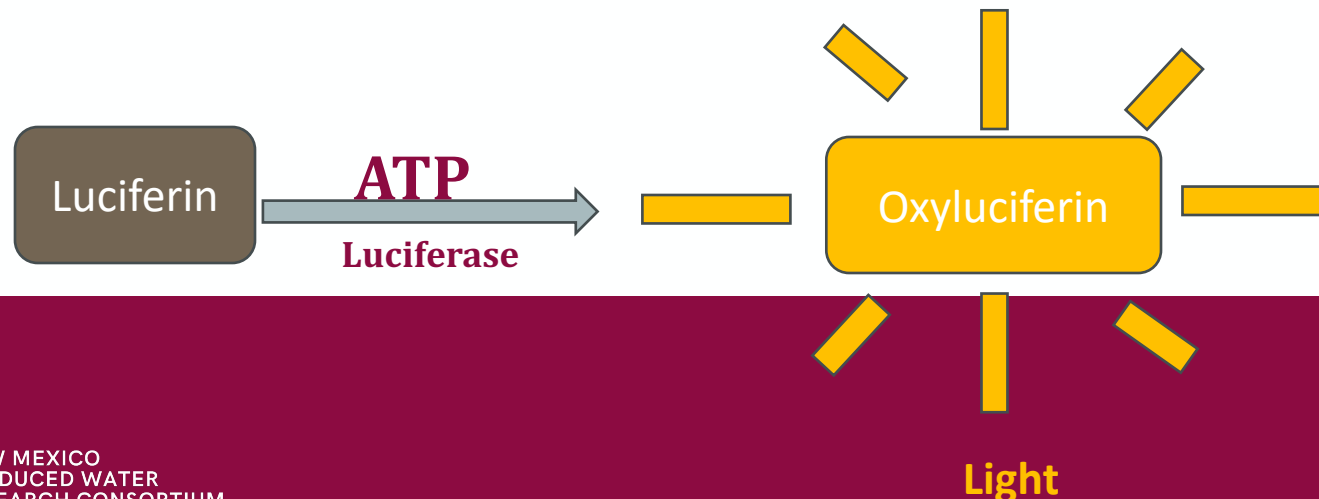
Human Cell Line Toxicity Study

Cell Viability

- **MTT Assay:** Enzyme activity assays for assessing cell metabolic activity.



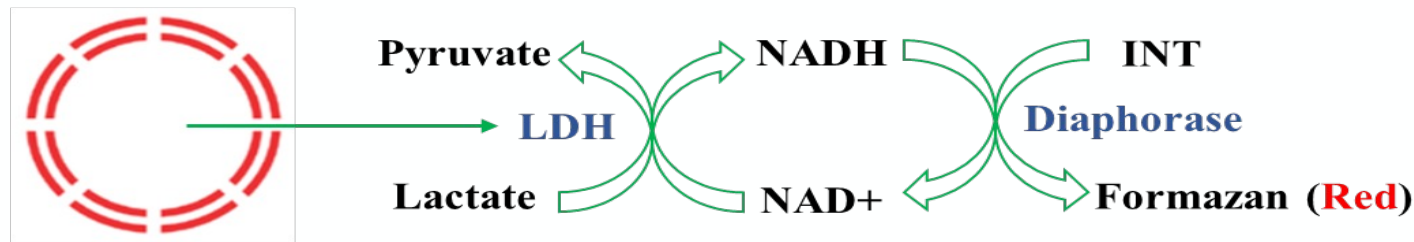
- **CellTiter-Glo[®] 2.0 Assay:** ATP assay for detection of viable cells



Human Cell Line Toxicity Study

Cell Membrane Damage

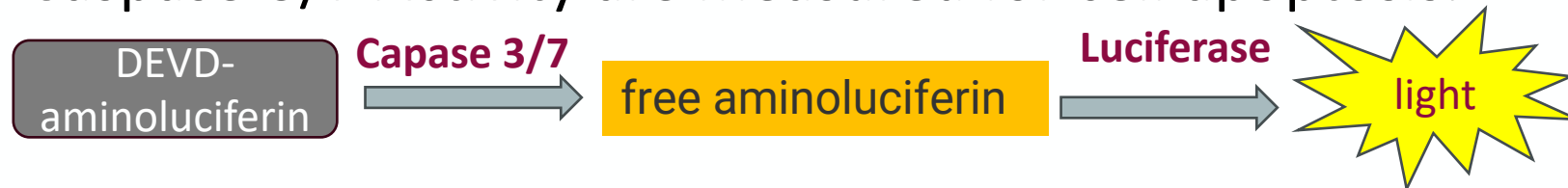
- lactate dehydrogenase (LDH) levels are related to cell membrane disruption and are a reliable indicator of cytotoxicity.



Damaged Cells

Apoptosis Assays (Programmed Cell Death)

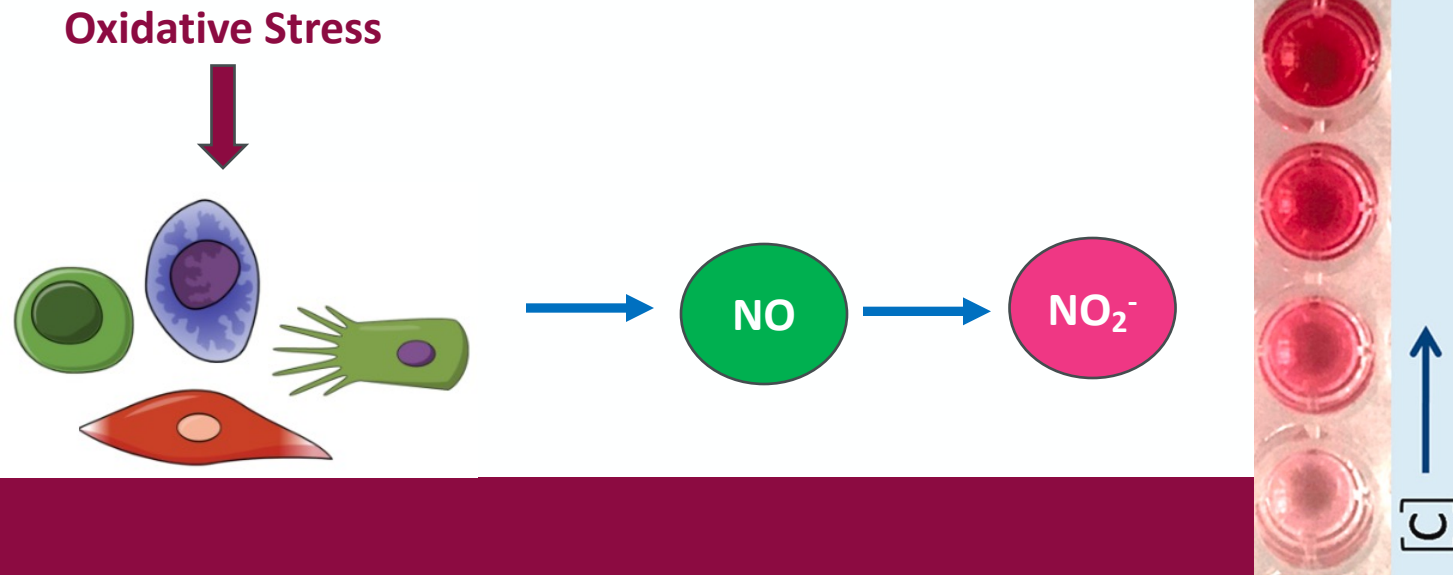
- Caspase-3 and caspase-7 are both activated universally during apoptosis. Caspase-3/7 Activity are measured for cell apoptosis.



Human Cell Line Toxicity Study

Oxidative Stress Assays

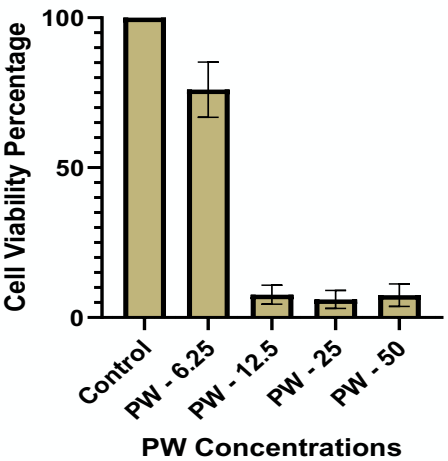
- Nitric oxide is an important physiological messenger and effector molecule in many biological systems
- Measures nitrite, a primary breakdown product of nitric oxide



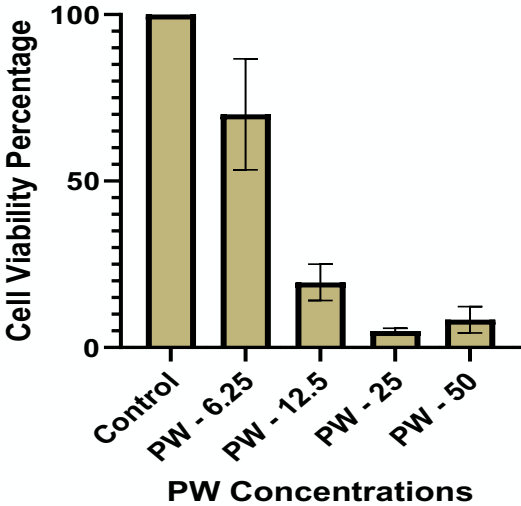
Griess Test

Cell Viability–MTT Assays

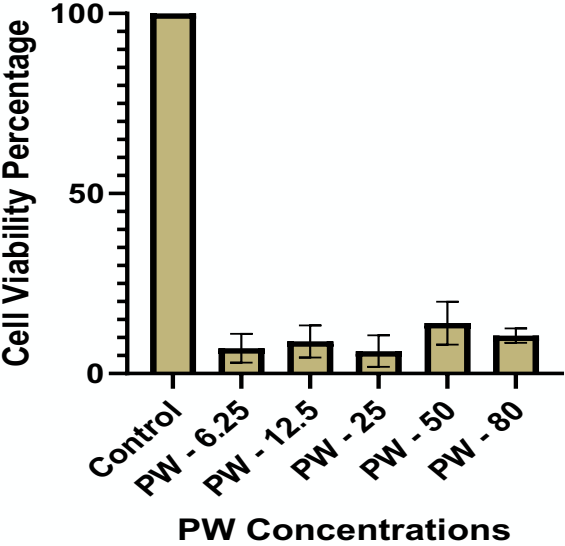
MCF-7 Cell Line - Feed



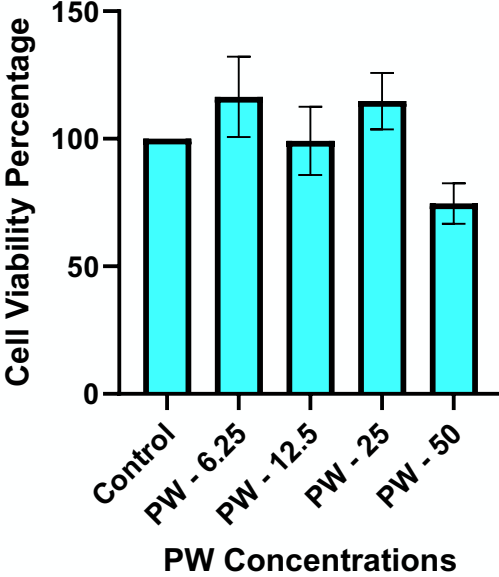
HEK293 Cell Line- Feed



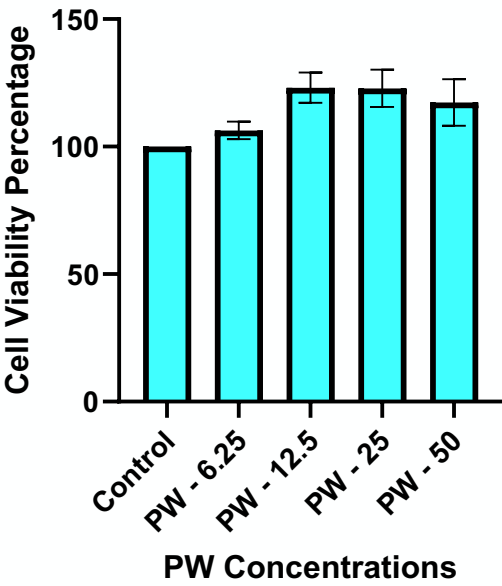
Caco-2 cell line- Feed



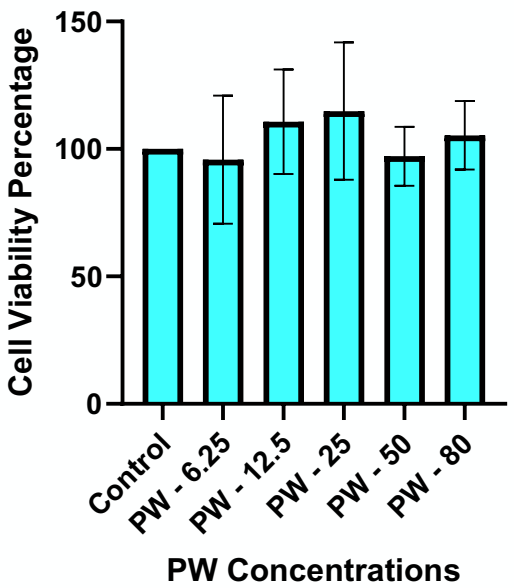
MCF-7 Cell Line - Distillate



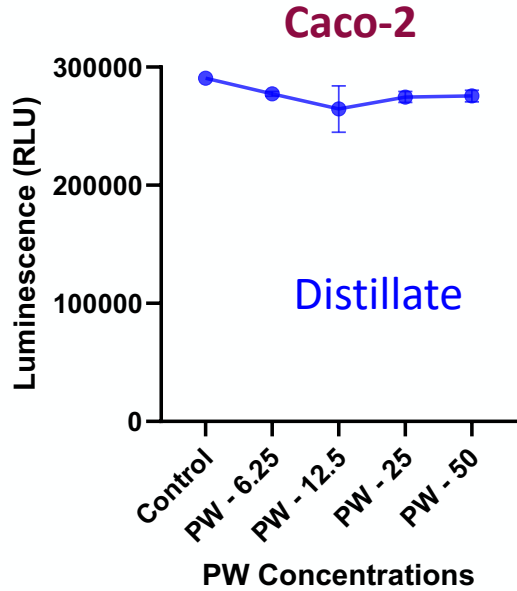
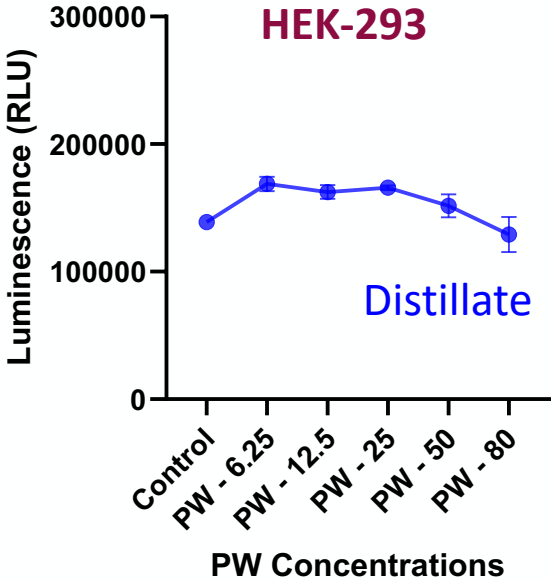
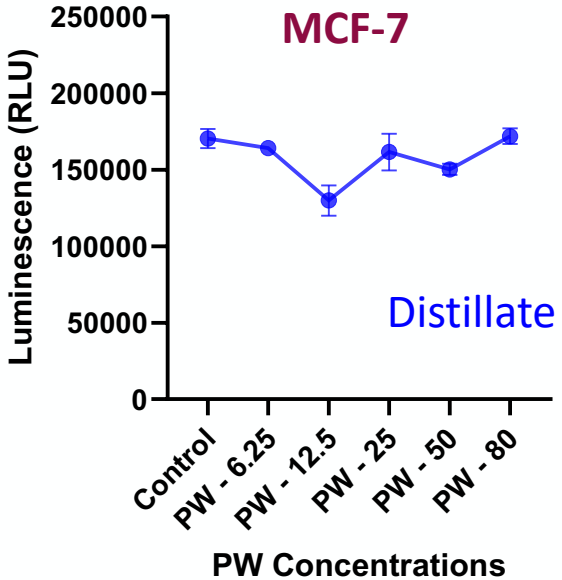
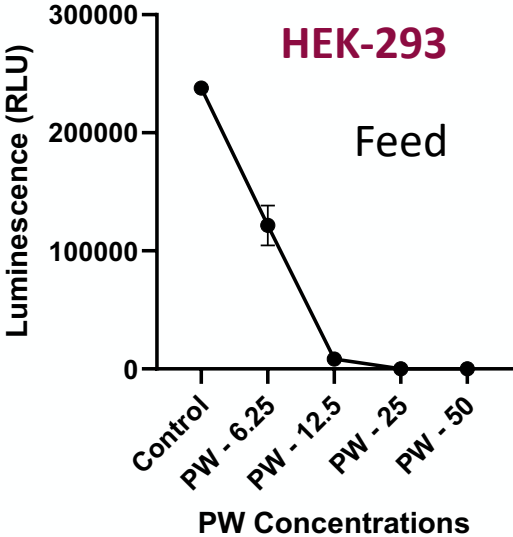
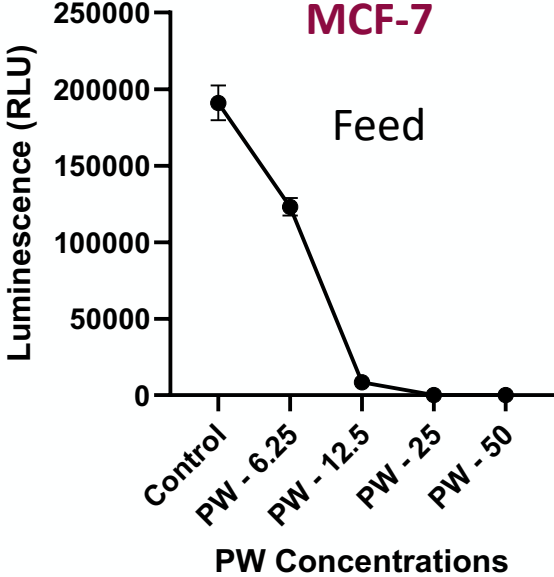
HEK293 Cell Line - Distillate



Caco-2 cell line - Distillate

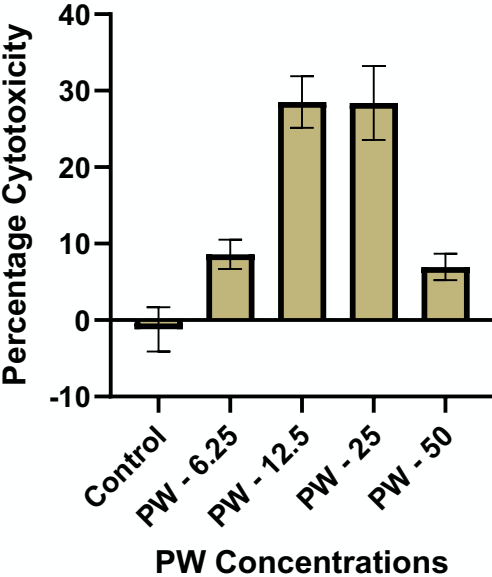


Cell Viability-ATP Assays

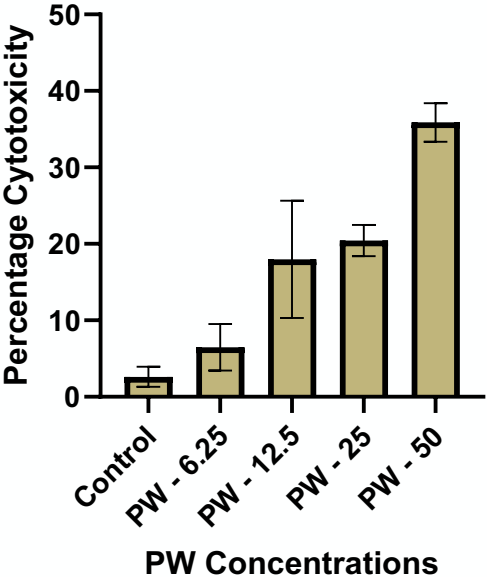


Cell Membrane Damage- -LDH Assays

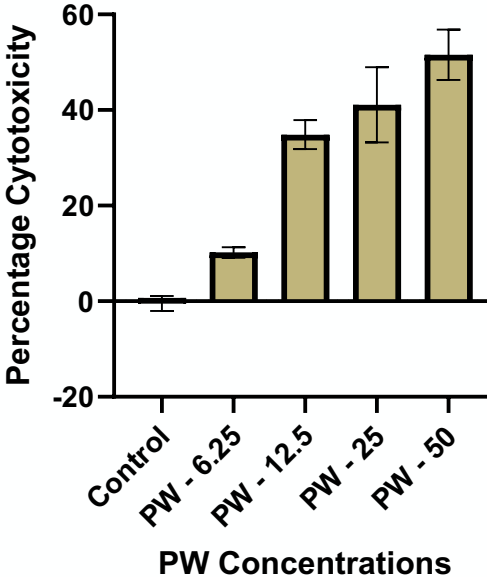
MCF-7 Cells - Feed



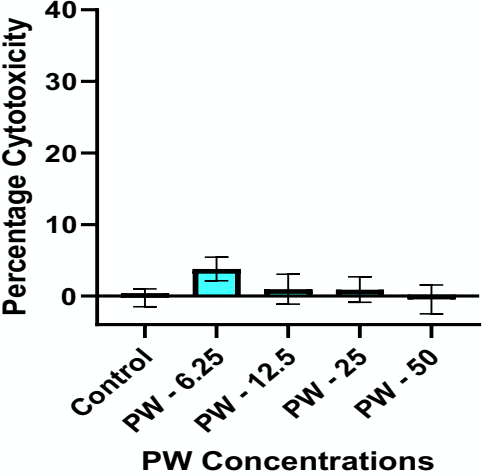
HEK293 Cell Line - Feed



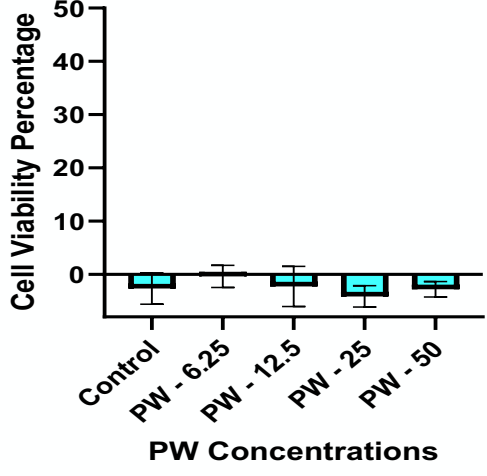
Caco-2 Cell Line - Feed



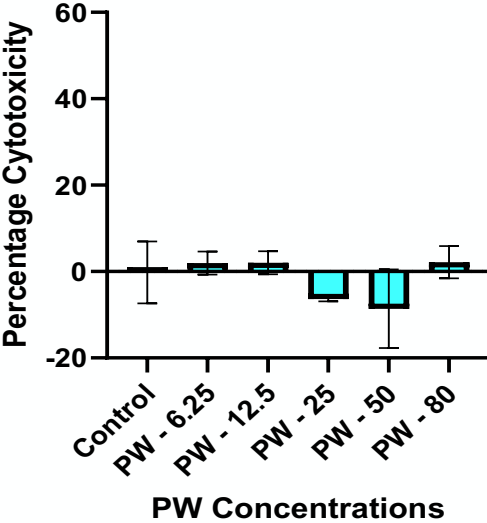
MCF -7 Cell Line - Distillate



HEK293 Cell Line - Distillate

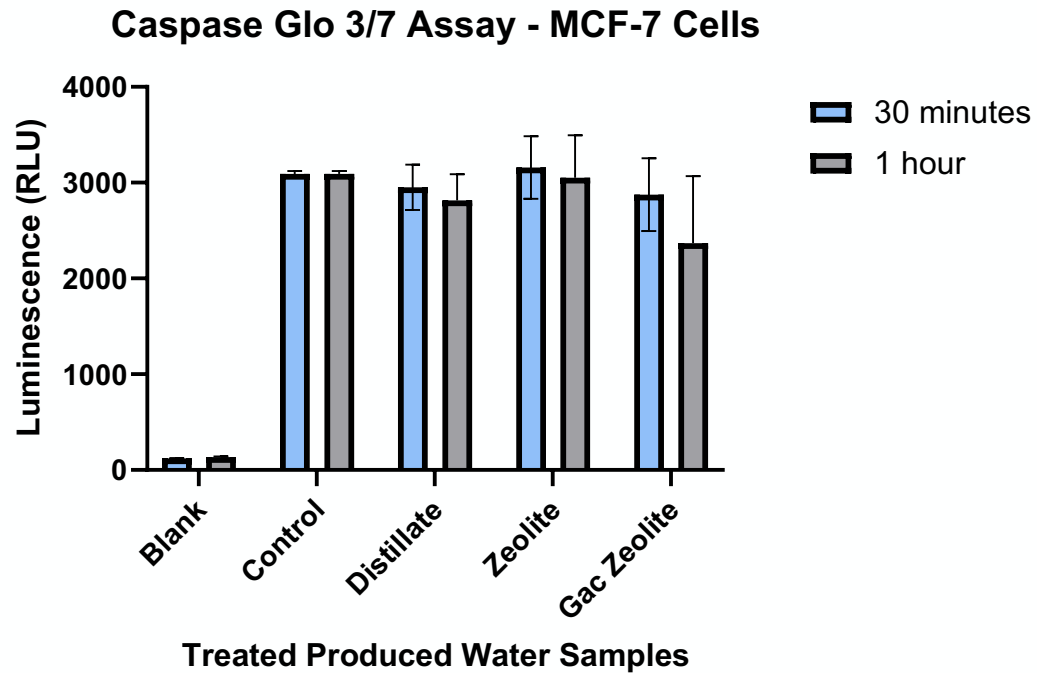


Caco-2 - LDH Test - Distillate

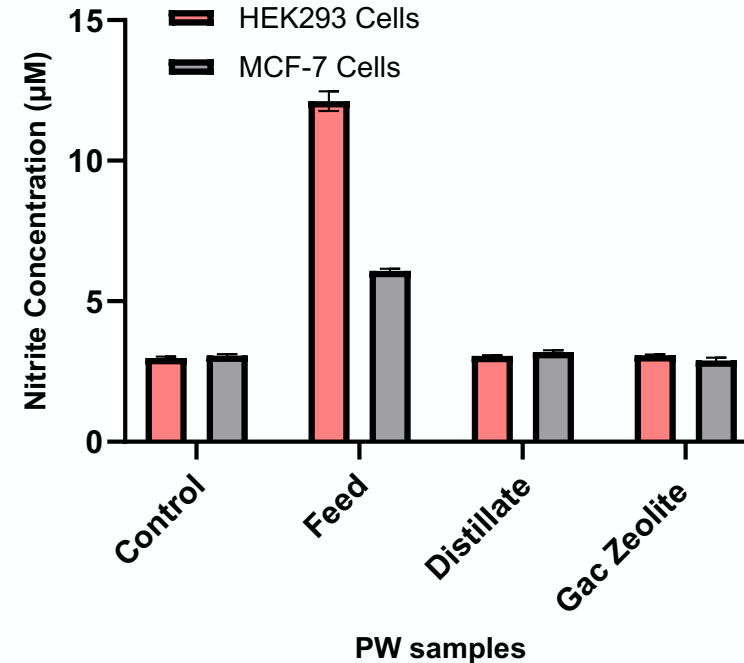


Other Cell Health Indicators

Cell Apoptosis



Cell Oxidative Stress



Nitrite is a marker of NO production

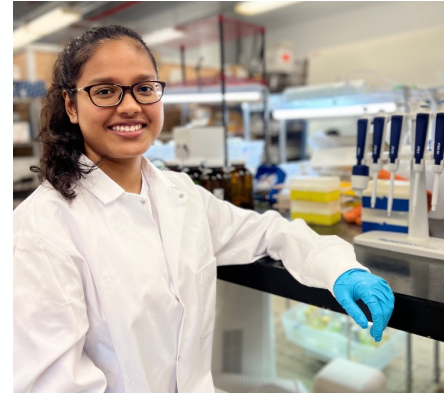
Conclusions

- All tested human cells are not sensitive for contaminants in the PW distillate
- Cell viability and apoptosis have no significant differences with the control samples
- There is no observed cell membrane damage and cell oxidative stress caused by the distillate.

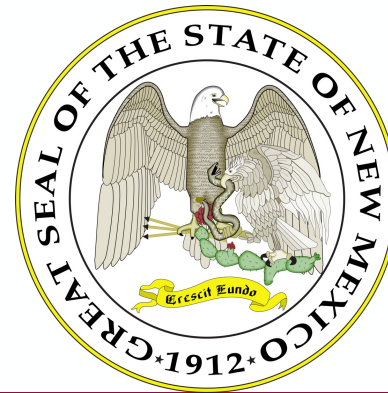
Acknowledgement



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Senuri Wijekoon Mudiyansele



— BUREAU OF —
RECLAMATION

