Sagacious Seminars

Briana Foley - U.S. EPA Foley.Briana@epa.gov

What makes a seminar sagacious?

- The power of storytelling
- Story construction
- Fundamentals of slide and poster design
- Data visualization
- Accessibility
- Examples
- Discussion / Q&A

When is a scientist a storyteller?

When you want the audience to care about your work.

The cognitive and emotional engagement of the audience aids in content comprehension and retention.

Research not effectively communicated is research not completed. Don’t waste the fruits of your labor through poor communication.

Relevant Communications Research

Energy Research & Social Science 101 (2023) 103100

Contents lists available at ScienceDirect

Energy Research & Social Science

journal homepage: www.elsevier.com/locate/erss

Perspective

'Telling tales': Communicating UK energy research through fairy tale characters

Carolynne Lord [a,*], Katherine Ellsworth-Krebs [b,1], Torik Holmes [c]

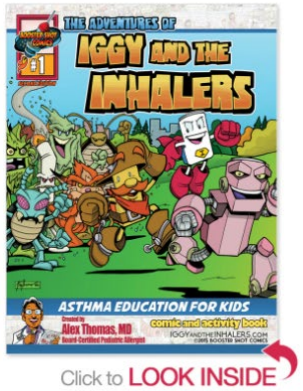
[a] Centre for Research into Energy Demand Solutions (CREDS), Sociology Department, Lancaster University, Lancaster LA1 4YW, United Kingdom
[b] Institute for the Contemporary Arts, Lancaster University, Lancaster LA1 4YW, United Kingdom
[c] Sustainable Consumption Institute, University of Manchester, M13 9PL, United Kingdom

"It would be easy to interpret this work as a trivialization of research or, even, a patronization of potential readers," Lord said in a statement. "This is not our intention. **The point is that communicating through specialist language is not adequately conveying the message to the communities that it needs to reach.** We need to start communicating our work in more accessible ways."

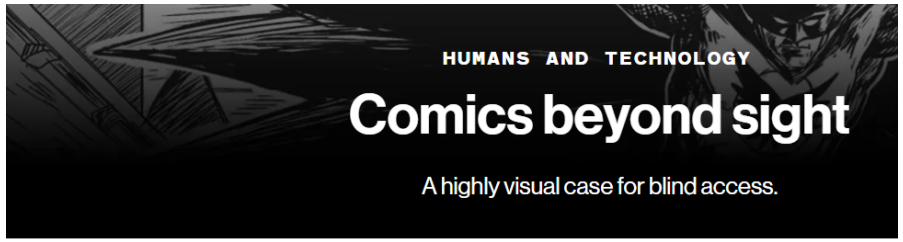
https://subscriber.politicopro.com/article/eenews/2023/06/06/fairy-tales-speak-to-the-wicked-problems-of-climate-change-00100296

Graphic medicine

Using storytelling, through comics, to improve public health outcomes by effectively communicating important concepts that may be scientifically complex, emotionally difficult or socially awkward.

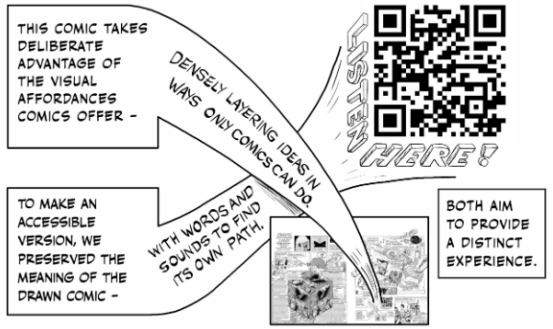


https://iggyandtheinhalers.com/



by **Nick Sousanis** and **Emily Beitiks**
June 28, 2023

For an audio adaptation with descriptive text and for annotations, visit: https://spinweaveandcut.com/mitcomic/



https://www.technologyreview.com/2023/06/28/1074341/comics-beyond-sight

NIH Office of Research on Women's Health
Diverse Voices Seminars:

https://www.youtube.com/watch?v=Cy9P-UpcYMc

Optimizing your voice and language

- Before your talk, do a vocal warm-up!
- Tone – The audience will match your energy.
- Cadence
  - Plot, logic, emotion, humor – are all critical to storytelling and require inflection, flow, timing and ordering.
- Stories with data are just stories with data. The story comes first, so focus on that when building presentations.
- **Clear language is effective language.** Use simple words so that you don't alienate or confuse your audience.
- Avoid acronyms!

Optimizing your message

Effective communication is about **editing down** and **stripping away content** to get to the focus of what is **relevant to the audience**.

*A real-world communication example of this is Axios' use of **Smart Brevity** to effectively disseminate news.*

Focus and Tailoring

01 

Know what your data are saying.

02 

Determine what your audience **NEEDS** to hear.

03 

Carefully examine what you **REALLY** want to say to meet audience **NEED**.

You want to start with a **Grabber** (hook, tweet, one-liner)

Something that entices the audience to select into the interaction

Short Form:

Poster
Elevator Pitch
LinkedIn Post
Ted Style Talk

Long Form:

Work in Progress Update
Podium Talk
Thesis Defense

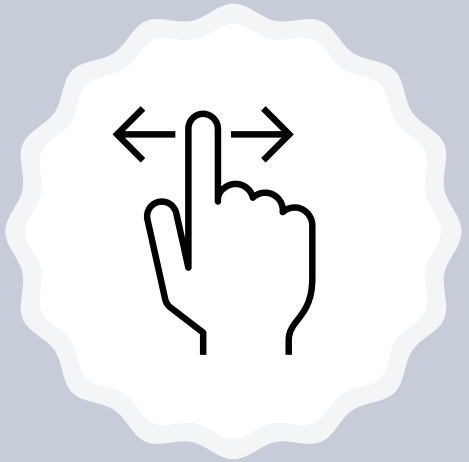
Georges Seurat – Fishermen, 1883

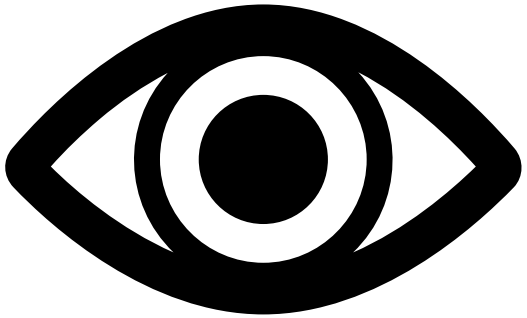
Brain Break!

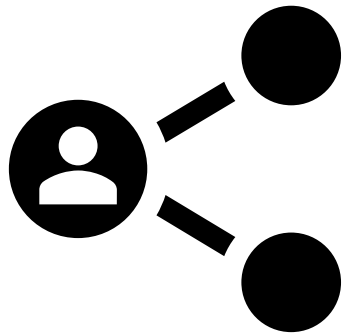
How quickly does audience attention drift?

Four key components to include in content

- Visuals
- Storytelling
- Interactivity
- Shareability







Concept courtesy of Bill Shander

Slide optimization

- Keep it simple and uncluttered.
  - Edit images and diagrams for the current discussion
- Use sans serif fonts.
  - This font has serifs (and it's harder to read).
- High contrast for easy distance viewing.
  - Use black background when showing microscopy
- Short lists with strategic animation are easy to follow.
- Avoid acronyms!
- Each slide is a micro-story within the macro-story that is the presentation.
  - Each micro-story needs to be complete in its content and telling.
- Humans can only consume one thing at a time.
  - Slide and verbal content should align so that you don't divide your audience's attention.

Good Design Optimizes Information Hierarchy

Controlling where you audience is paying attention

Typography matters: size, font, placement and color of text all impact audience attention

- Headers, sub-headers should not be the same size

Humans tend to look at the center of an image first.

Use animations to lead the viewer through the content in slides.

Concept courtesy of Bill Shander

Gestalt Principles of Design

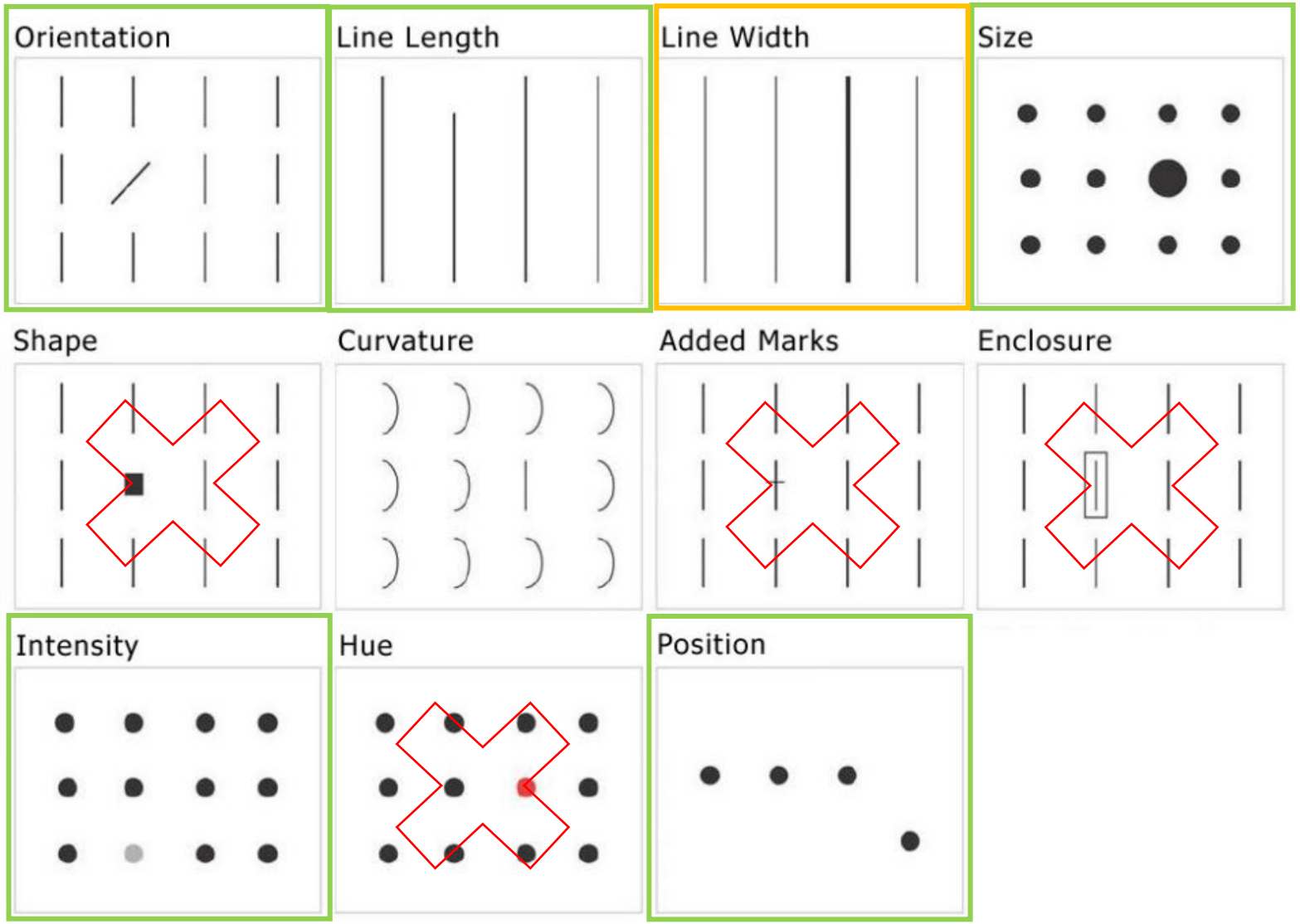
Capitalizing on subconscious (pre-attentive) cognition

- Proximity: Things that are close together are cognitively grouped together. It's why scatter plots work.
- Similarity: Cognitively, like things are grouped together regardless of proximity.
  - Symbols in scatter plots
- Parallelism: Humans are good at detecting parallels, but not at quantifying angles.

https://bootcamp.uxdesign.cc/gestalt-psychology-how-design-alters-our-perception-ad306c66bbec

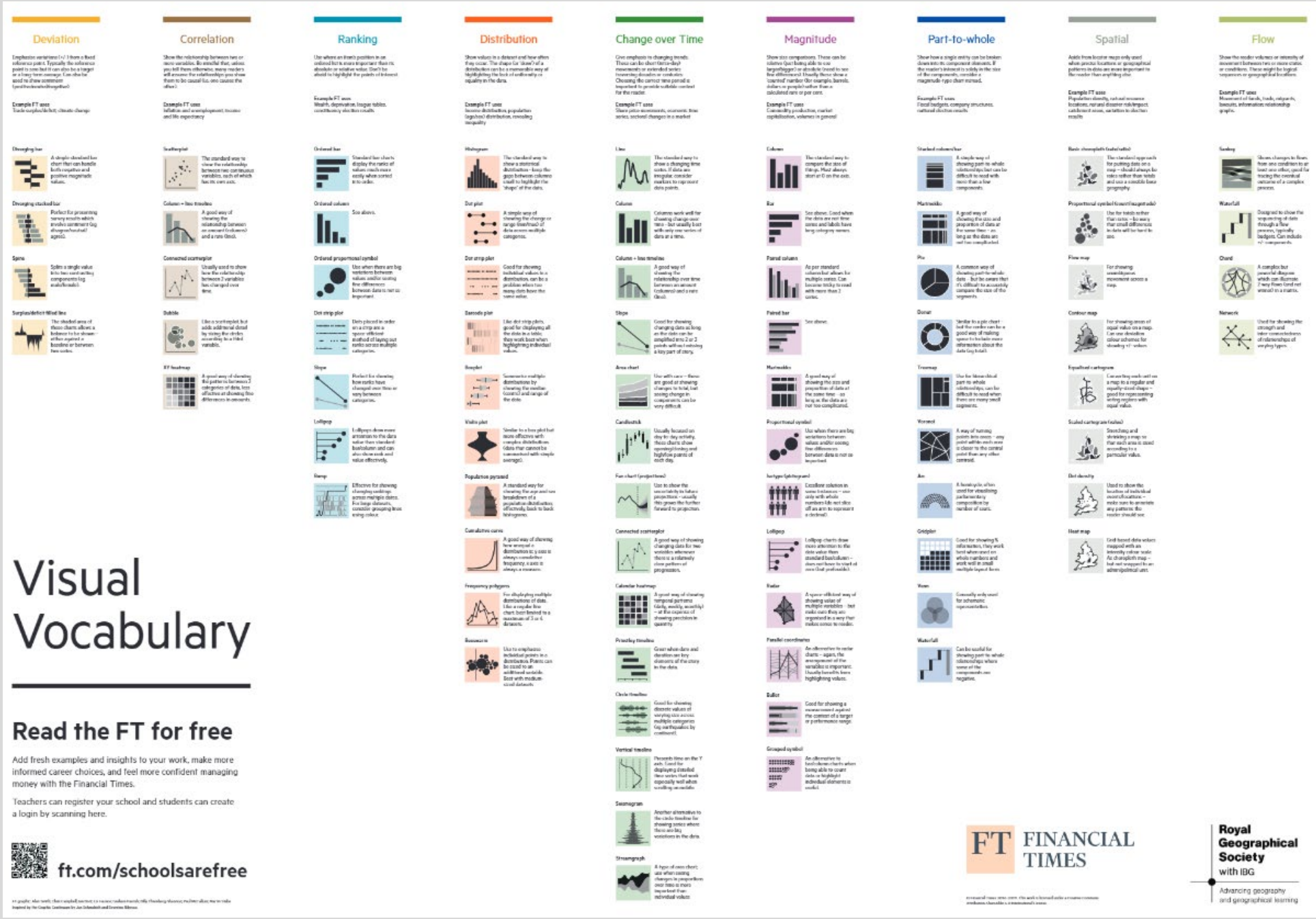
Concept courtesy of Bill Shander

Pre-attentive processing – subconscious visual acuity



Concept and symbol array courtesy of Bill Shander

Choosing the right chart



https://github.com/Financial-Times/chart-doctor/blob/main/visual-vocabulary/FT4schools_RGS.pdf

Brain Break!

Which piece of train track is longer?

https://en.m.wikipedia.org/wiki/Jastrow_illusion

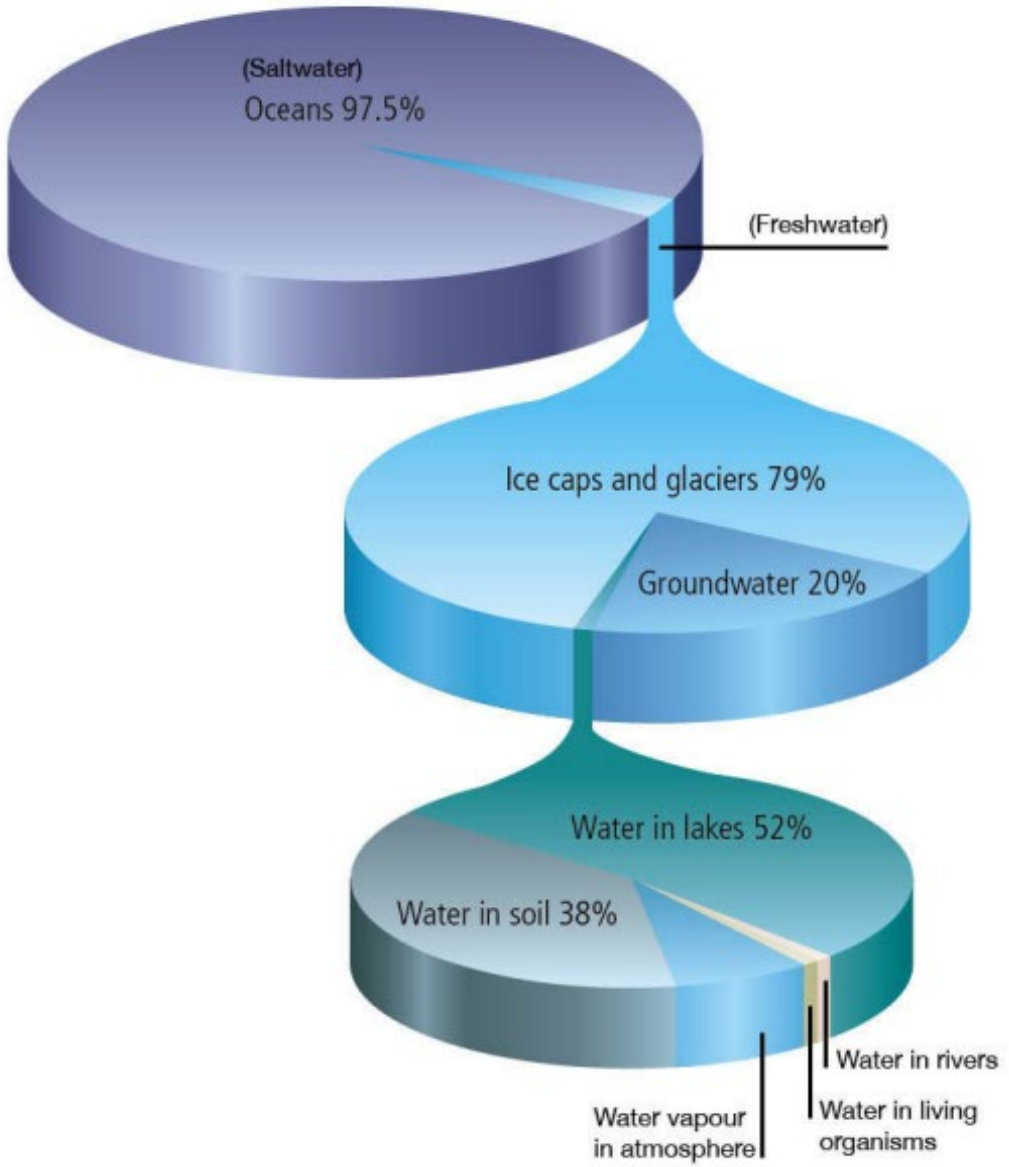
Round shapes / pie charts

- Good at showing percentage of the whole.
- They can make comparisons difficult, especially accurate estimates of quantity.
  - Terrible for enabling distinct value comparison.
  - Humans are very bad at comparing the areas of circles.
- Don’t EVER use double doughnut charts.
  - They cannot be accurately interpreted due visual tricks of arcs.

Concept courtesy of Bill Shander

Round shapes / pie charts continued

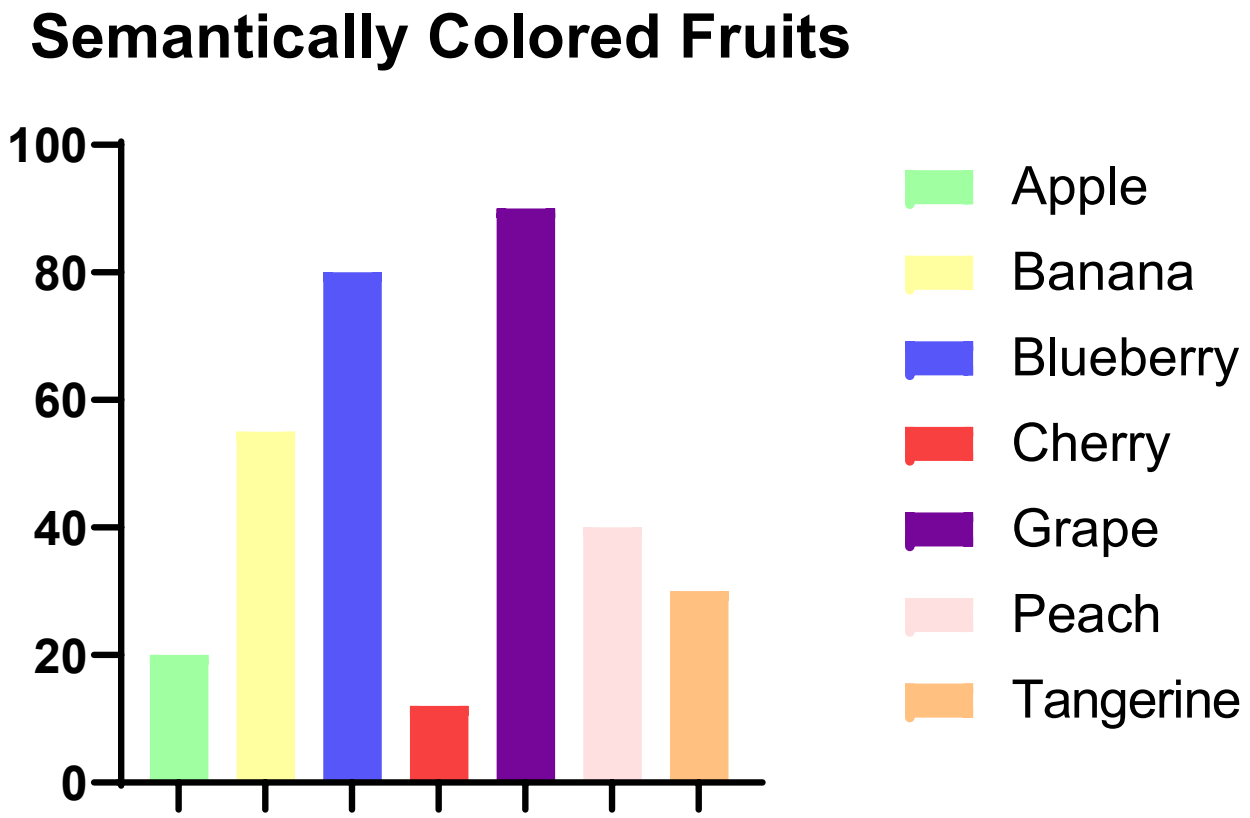
These trickle-down pie charts are visually appealing and easy to interpret.



Illustrated Atlas of Wildlife;
Andrew Davies/Creative Communication

Color

- Use semantic color when relevant
  - EXCEPT with gender or ethnicity.
- Use high contrast colors so they pop off the page.
- Avoid red/green combo when possible



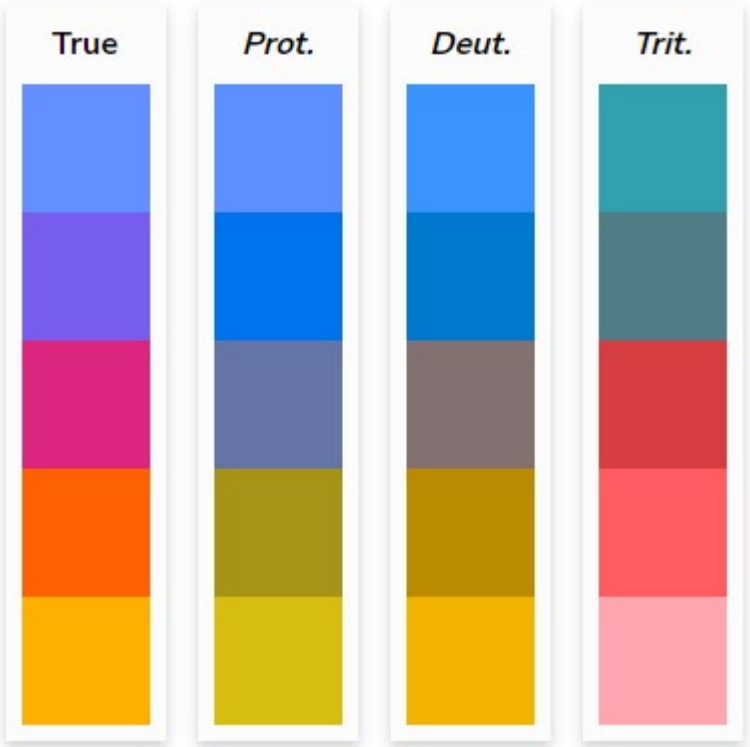
Essential content for accessible data stories

- Headers
- Subheadings
- Charts / graphs
- White Space

More Things to Consider About Accessibility

- Have a title on each slide
- Large fonts, high contrast but not vibrating colors
- Number your slides so that they are easy to reference
- Aim for a colorblind friendly palette
- Use the Microsoft Accessibility Checker when making presentations
- If presenting in Teams, consider using Power Point Live to share your slides
- Describe your whole slide

Palettes that are color-blind accessible



IBM Design Library

Paul Tol

David Nichols - https://davidmathlogic.com/colorblind/#%23D81B60-%231E88E5-%23FFC107-%23004D40

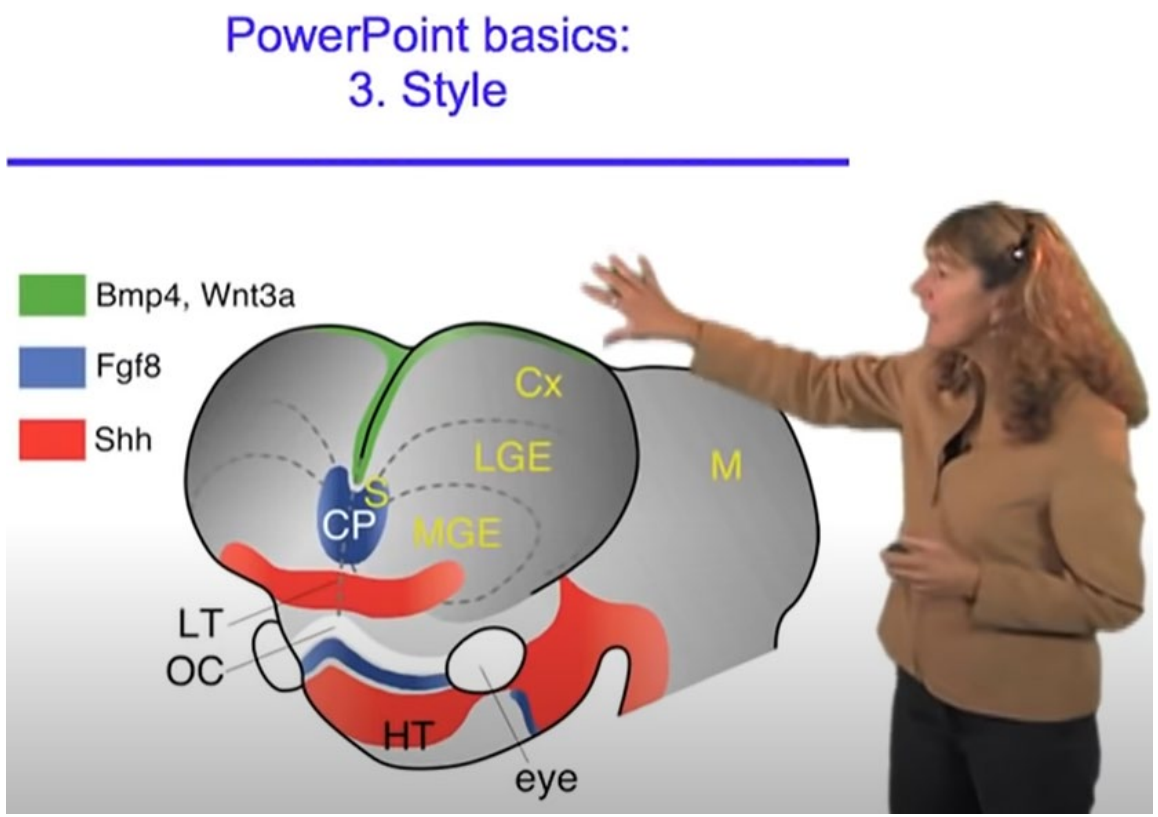
Data vis example courtesy of Bill Shander

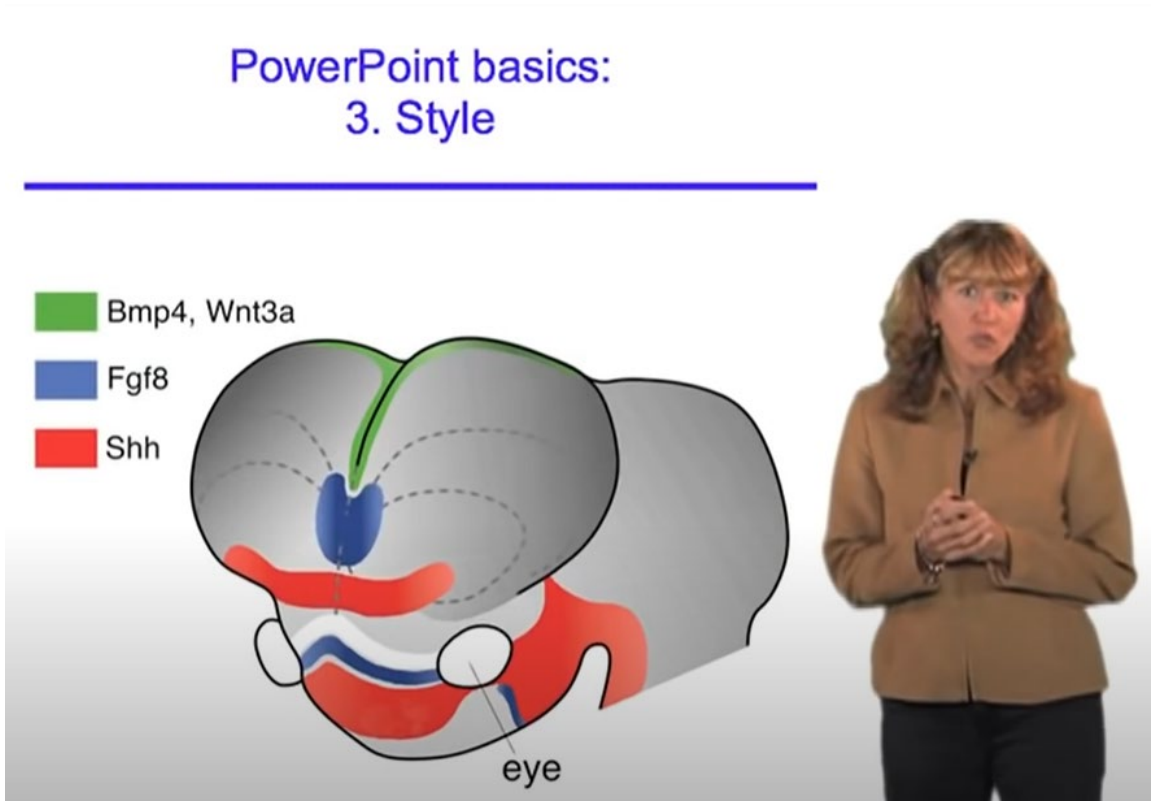
Examples

Choosing the best visuals to support your story.

https://medium.com/analytics-vidhya/recreating-gapminder-in-tableau-a-humble-tribute-to-hans-rosling-53de74b18ec

Simplification for Clarity





Susan K McConnell, PhD https://www.youtube.com/watch?v=Hp7Id3Yb9XQ

Reformatting for Clarity and Impact

Why are trees dropping so many nuts? Climate may drive erratic 'masting'

Bounty of acorns may be a sign of next spring's weather

23 NOV. 2021 · 11:45 A.M. · BY ELIZABETH PENNISI

Lizards may be protecting people from Lyme disease in the southeastern United States

The reptiles make poor hosts for transmitting the infection

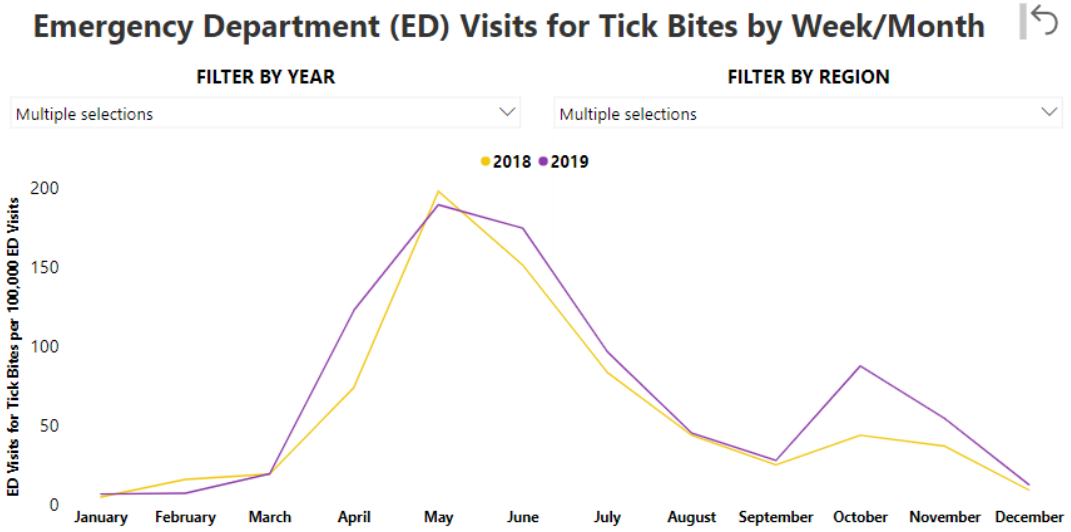
5 FEB 2021 · BY HARINI BARATH

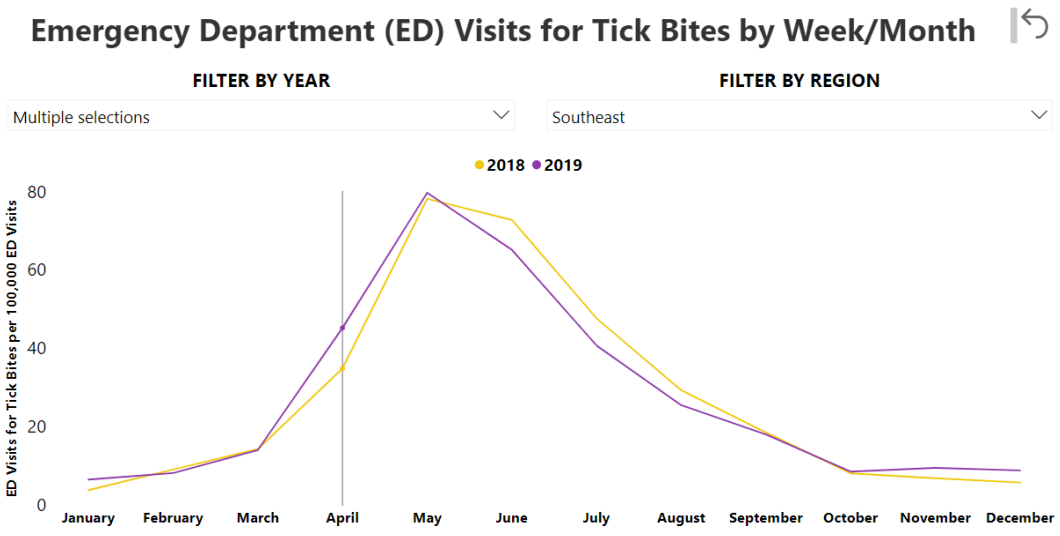
https://www.science.org/content/article/why-are-trees-dropping-so-many-nuts-climate-may-drive-erratic-masting

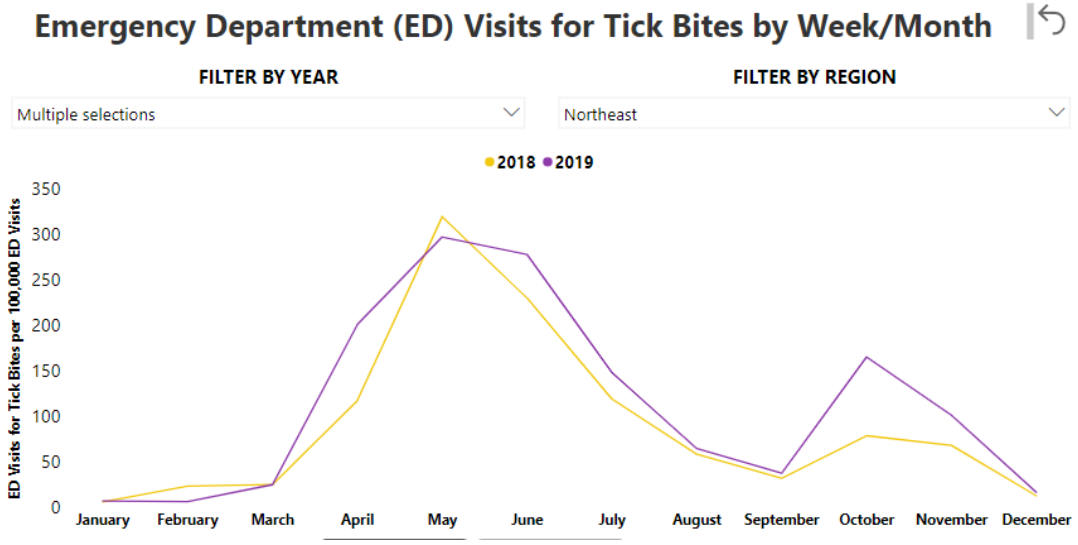
https://www.science.org/content/article/lizards-may-be-protecting-people-lyme-disease-southeastern-united-states

Reformatting for Clarity and Impact

CDC Data in Native Format:



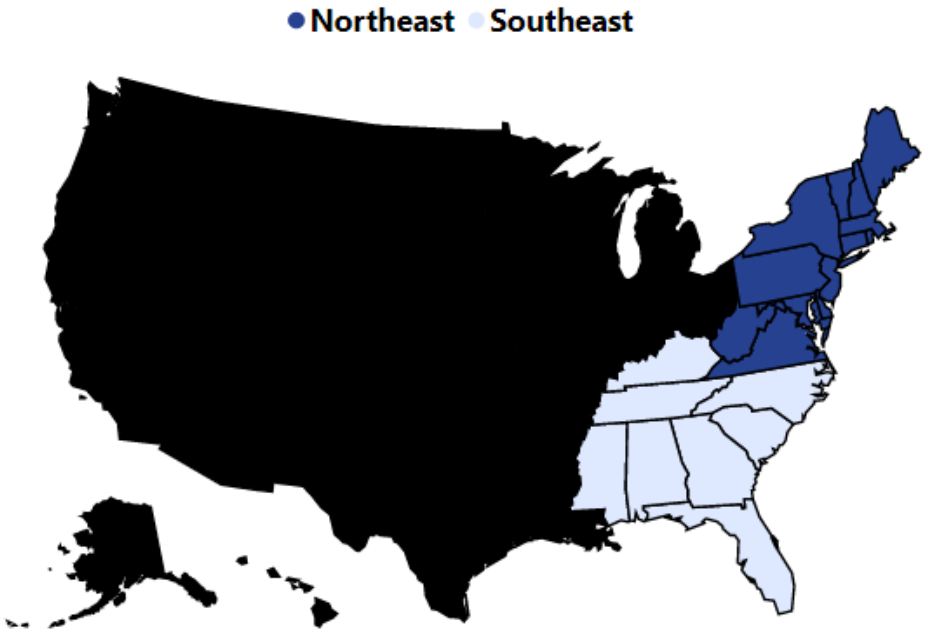


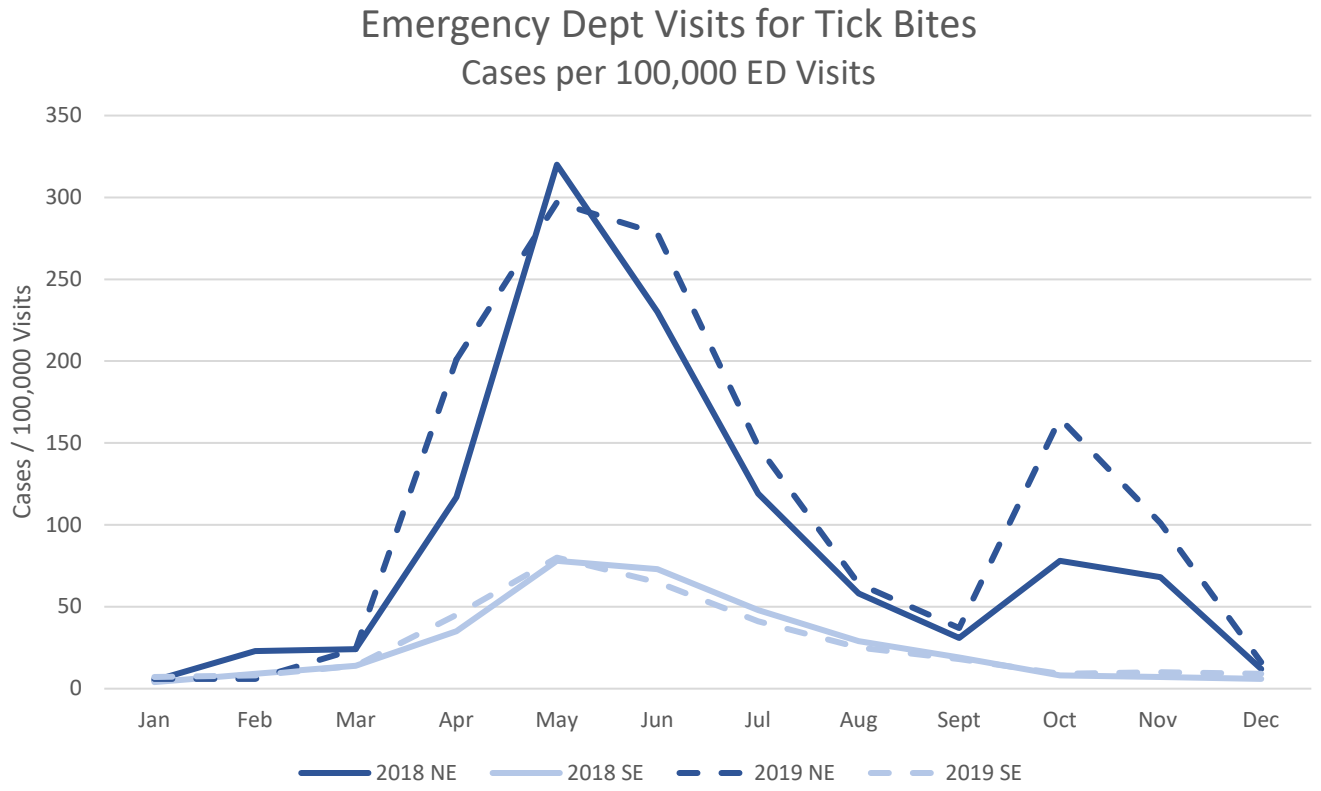


https://www.cdc.gov/ticks/tickedvisits/tick-bite-by-region.html

Reformatting for Clarity and Impact

Emergency Department Visits for Tick Bites



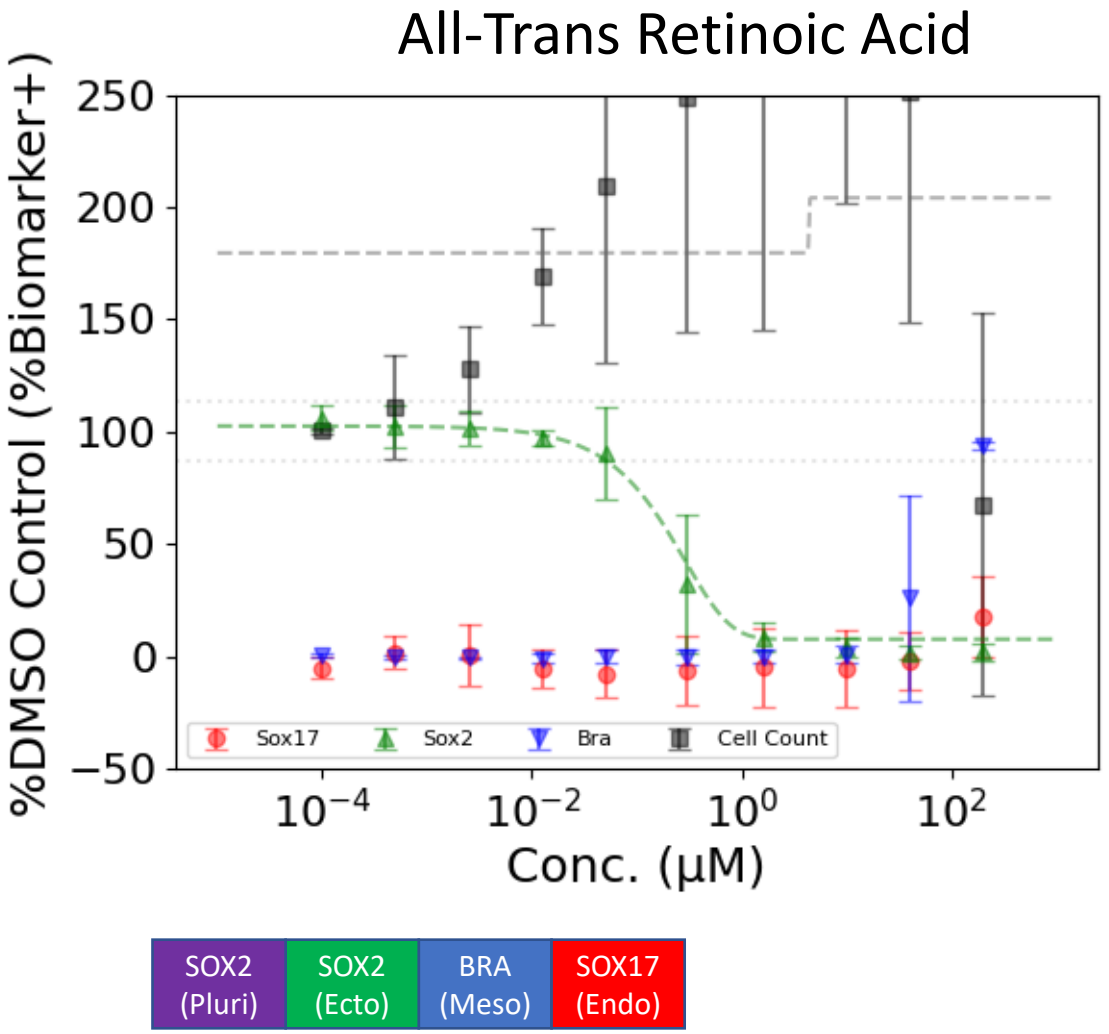


https://www.cdc.gov/ticks/tickedvisits/tick-bite-by-region.html

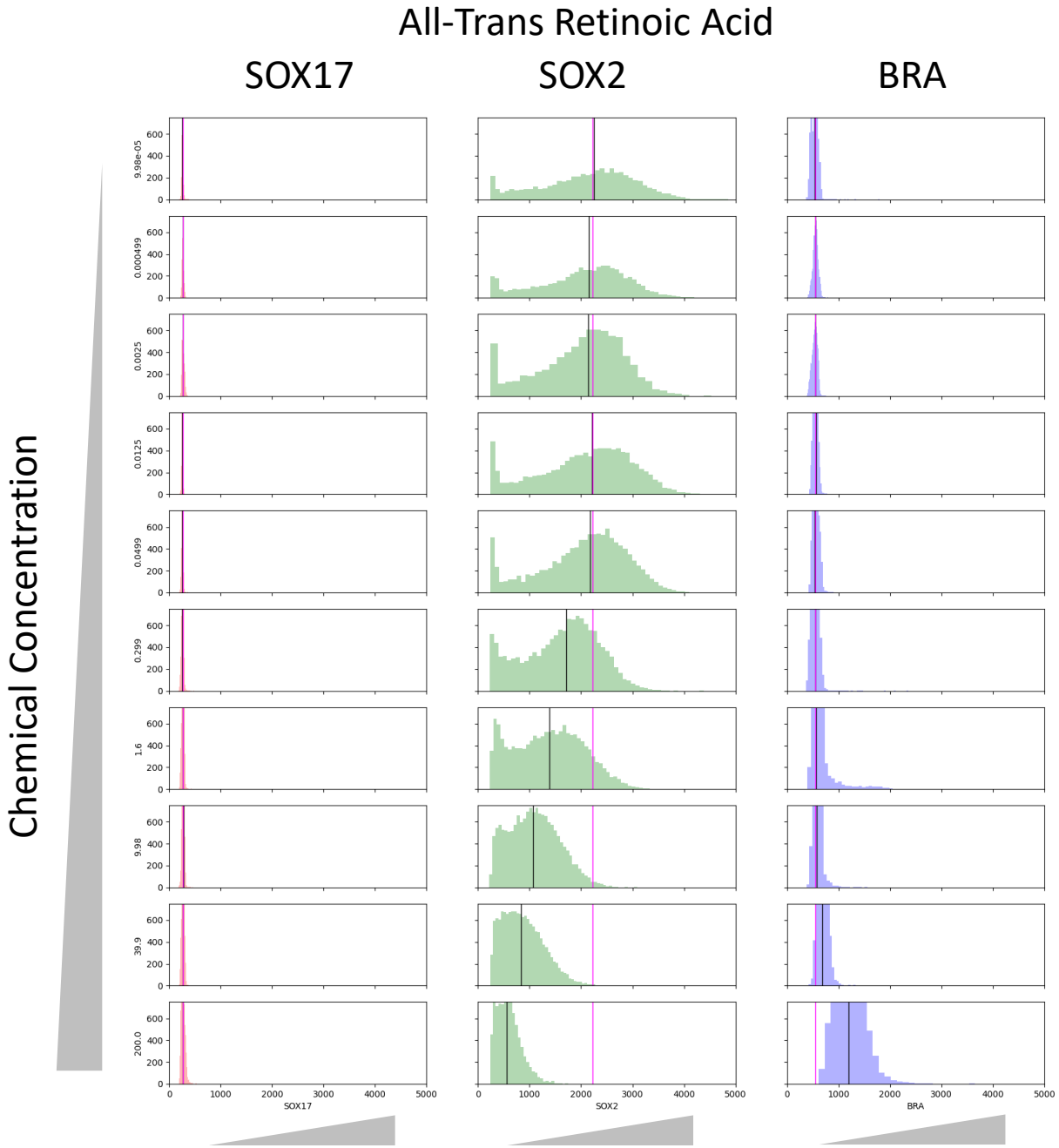
Formatting slides to share complex data sets

- Avoid using two Y-Axes
  - Two graphs, each with one Y-axis, are better than one graph with two Y-axes.
- Simplify chart labels
- Use semantic coloring
- Be aware that the verbal description and time spent absorbing the material increases with complexity of the data visualization.

DevTox GLR Ectoderm Results



SOX2 (Pluri)	SOX2 (Ecto)	BRA (Meso)	SOX17 (Endo)



Gamble, John T.; Oct 2022 BCTD WIP

Ed Hawkins – Global Temperature Change (1850-2020)

The title and **visual appeal** of your poster is your hook!

Development of a 5α-reductase High-throughput Screening Assay for Androgen Steroidogenesis

Briana Foley[1], Wendy Stewart[1], Madison Feshuk[1], Katie Paul Friedman[1], Russell S. Thomas[1], Chad Deisenroth[1]

[1]Center for Computational Toxicology and Exposure, Office of Research and Development, U.S. Environmental Protection Agency, Research Triangle Park, NC 27711, United States

www.epa.gov/research

Briana Foley I Foley.Briana@epa.gov

Introduction

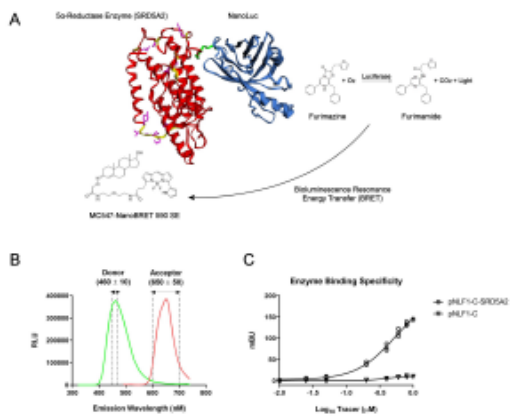
The U.S. EPA employs high-throughput screening assays to identify environmental chemicals that may pose a risk to human health. Many assays are utilized by the Endocrine Disruptor Screening Program to evaluate effects on estrogen, androgen, and thyroid endocrine pathways. Altered androgen hormone biosynthesis contributes to endocrine disruption that may result in impaired reproductive and sexual development. Steroid 5α-reductase enzymes catalyze the conversion of testosterone into the more potent androgen 5α-dihydrotestosterone. Type 2 5α-reductase enzyme (SRD5A2) deficiency is associated with decreased virilization in males and presents an important mode-of-action when evaluating environmental chemical exposure.

Objective

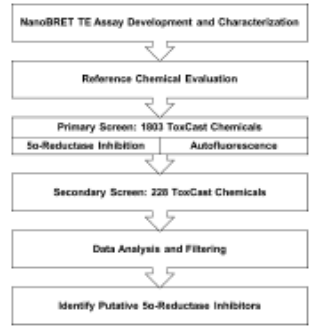
NanoBRET Target Engagement assay technology uses bioluminescence resonance energy transfer (BRET) to directly measure interactions of drugs or chemicals with intracellular protein targets in living cells. The objective of this study was to adapt NanoBRET Target Engagement assay technology for high-throughput screening of SRD5A2 inhibition.

NanoBRET-SRD5A2 Target Engagement Assay Overview

Predicted protein structure of human 5α-reductase isozyme 2 (SRD5A2) fused to NanoLuc luciferase. BRET signaling occurs when the testosterone-fluorophore tracer (MC547-NanoBRET 590 SE) is directly bound to the enzyme (A). Confirmation of the donor luminescence and acceptor fluorescence wavelength emissions (B). Concentration-dependent enzyme binding specificity of tracer to the fusion protein (C). RLU: Relative light units; mBU: milli BRET Units.



Study Workflow



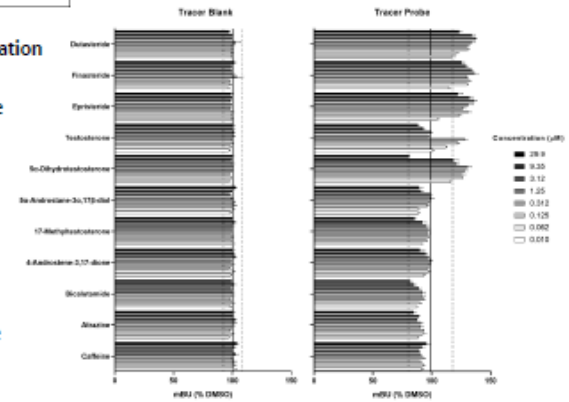
Workflow

NanoBRET Target Engagement assay technology was adapted for inhibition of human 5α-reductase.

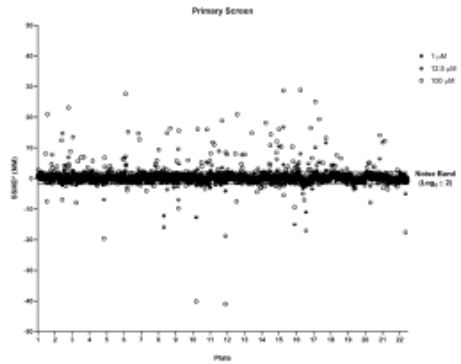
- Reference chemicals were used to evaluate the initial assay performance.
- Primary screening of 1803 ToxCast chemicals was conducted in limited concentration-response format in parallel with autofluorescence screening of the chemical library.
- 228 chemicals from the primary screen were evaluated in a broader multiple concentration-response format for functional inhibition of 5α-reductase.
- Final analysis of 91 chemical hits was performed to classify bioactivity.

Reference Chemical Evaluation

A chemical training set consisting of 5α-reductase inhibitors, enzyme substrate and metabolites, substrate analogs, and negative control compounds were evaluated in concentration-response format in the absence (Tracer Blank) or presence (Tracer Probe) of tracer.



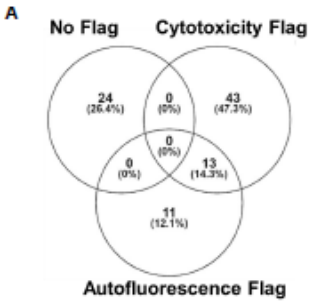
ToxCast Primary Screen

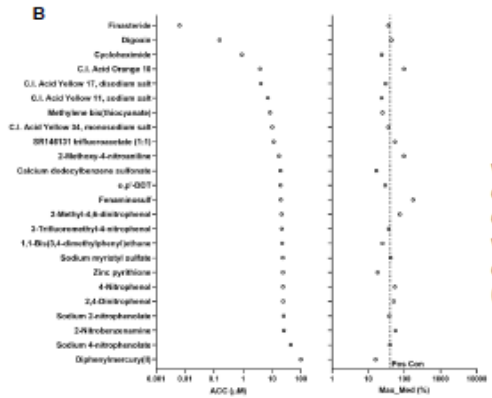


A blinded ToxCast chemical library of 1803 compounds was screened across three concentrations (1, 12.5, 100 μM) in one experimental replicate. Effect size was determined using robust strictly standardized mean difference method-of-moment (SSMD* (MM)).

ToxCast Secondary Screen

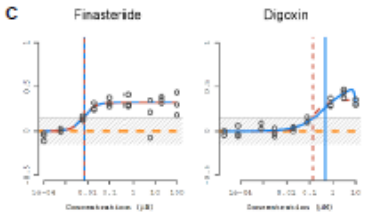
The secondary screen identified 91 total chemicals with potential for inhibition of 5α-reductase. Chemicals were flagged and binned according to potential confounding factors including autofluorescence interference and cytotoxicity (A).





The 24 chemicals identified without any flags were rank-ordered by active concentration at cutoff (ACC) with corresponding maximum observed effect level (Max_Med (%)) (B).

Concentration-response plots for the most potent compounds: Finasteride and Digoxin. (C).



Conclusions

- NanoBRET target engagement assay technology was successfully adapted to directly measure interactions of testosterone substrate with intracellular 5α-reductase enzymes in living cells.
- The NanoBRET-SRD5A2 assay demonstrated high precision, modest dynamic range, and marginal assay quality.
- Few environmental chemicals were identified as potential inhibitors of 5α-reductase enzyme.

The views expressed do not reflect the views or policies of the U.S. EPA.

www.epa.gov/research

High precision, **direct measurement** of steroid - enzyme interactions in **living cells.**

Development of a 5α-reductase High-throughput Screening Assay for Androgen Steroidogenesis

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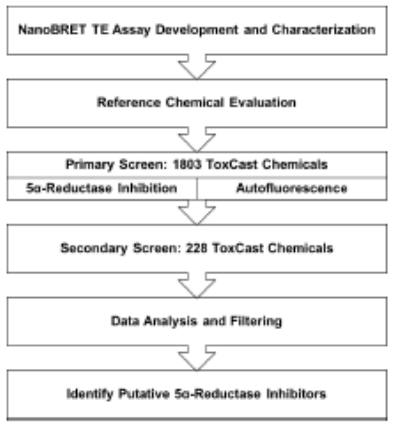
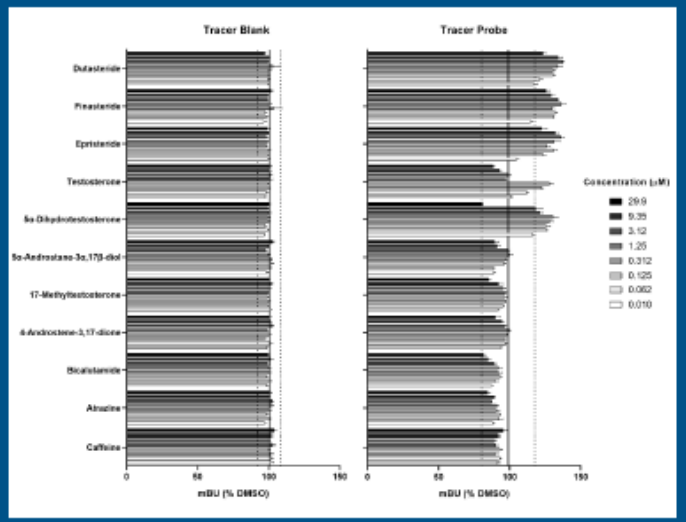
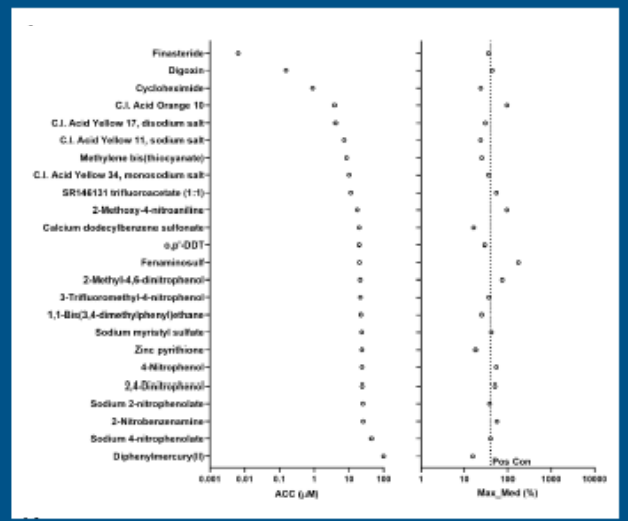
Workflow





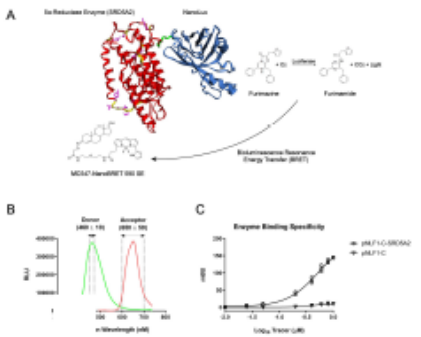
- **Assay response was specific to enzyme substrates and pharmaceutical 5aRIs.**
- A chemical training set consisting of 5α-reductase inhibitors, enzyme substrate and metabolites, substrate analogs, and negative control compounds were evaluated in concentration-response format absence (Tracer Blank) or presence (Tracer Probe) of tracer.



- The 24 active chemicals identified without any flags were rank-ordered by active concentration at cutoff (ACC) with corresponding maximum observed effect level (Max_Med (%)) (B).
- The most potent compounds were pharmaceutical compounds.

NanoBRET-SRD5A2 Target Engagement Assay Overview

BRET signaling occurs when the testosterone-fluorophore tracer (MC547-NanoBRET 590 SE) is directly bound to the enzyme (A). Confirmation of the donor luminescence and acceptor fluorescence wavelength emissions (B). Concentration-dependent enzyme binding specificity of tracer to the fusion protein (C). RLU: Relative light units; mBU: milli BRET Units.



Conclusions

- NanoBRET target engagement assay technology was successfully adapted to directly measure interactions of testosterone substrate with intracellular 5α-reductase enzymes in living cells.
- The NanoBRET-SRD5A2 assay demonstrated high precision, modest dynamic range, and marginal assay quality.
- Few environmental chemicals were identified as potential inhibitors of 5α-reductase enzyme.

U.S. Environmental Protection Agency
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Expanding New Approach Methods for Developmental Toxicity: The DevTox Germ Layer Reporter Platform

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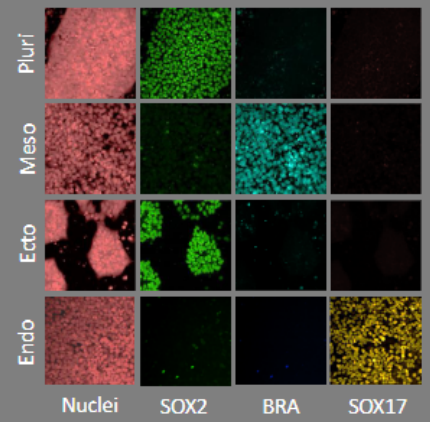
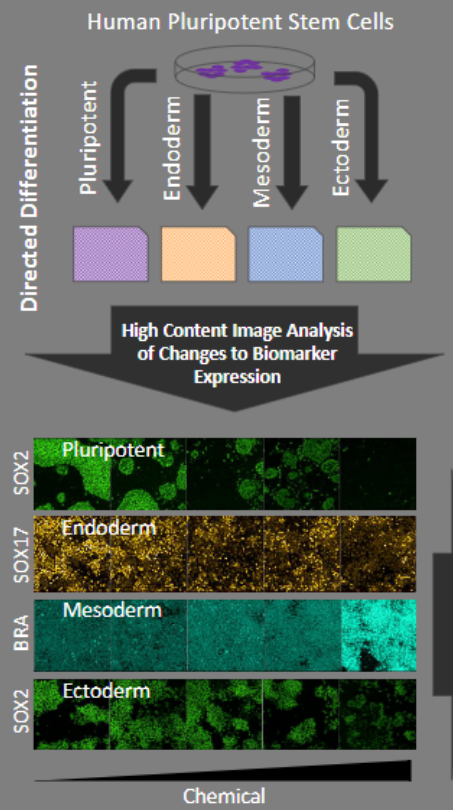
High-Throughput Profiling of Human Developmental Toxicity

RUES2-GLR: transgenic human pluripotent cell line with fluorescent protein reporters.[3]

- SOX2-mCitrine (Ectoderm/Pluripotency)
- BRA-mCerulean (Mesoderm)
- SOX17-tdTomato (Endoderm)

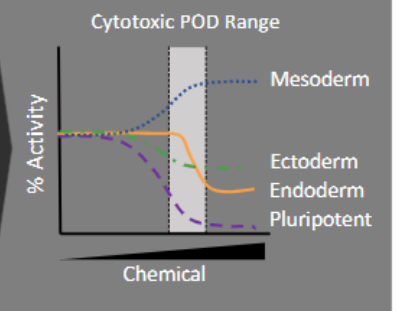


Chemical perturbations to gastrulation pathways are assessed for each lineage-specific biomarker to provide a mechanistic profile specific to each cell state.



To provide complete gastrulation coverage to our adapted Endoderm assay[1,2], additional high-content imaging assays were created for screening developmental toxicants.

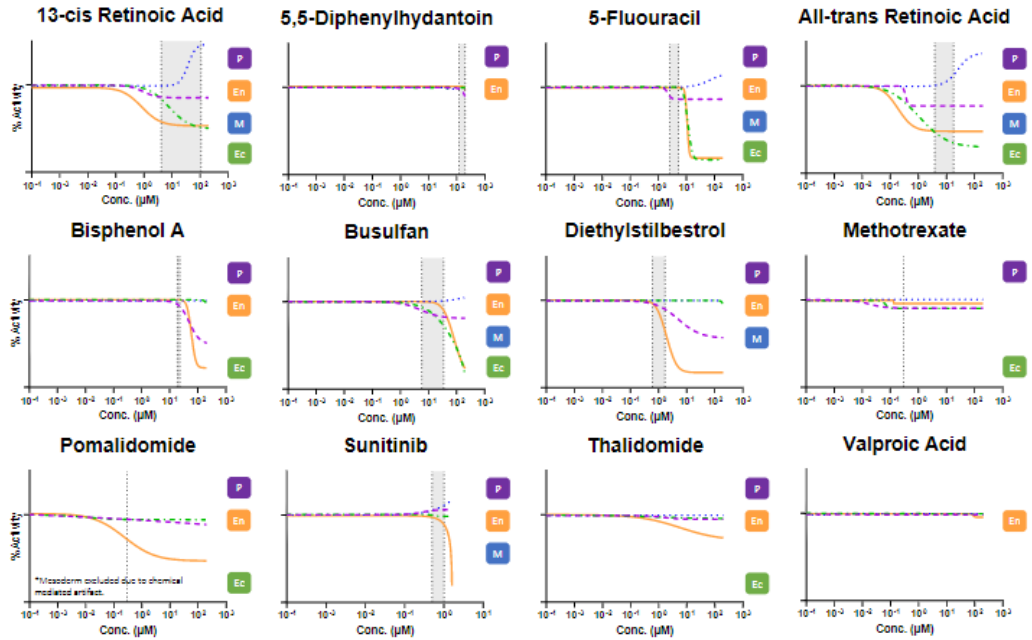
For the platform, human pluripotent stem cells are induced to either of the three germ layer lineages (or maintained at pluripotency) while under chemical exposure in 384-well format.



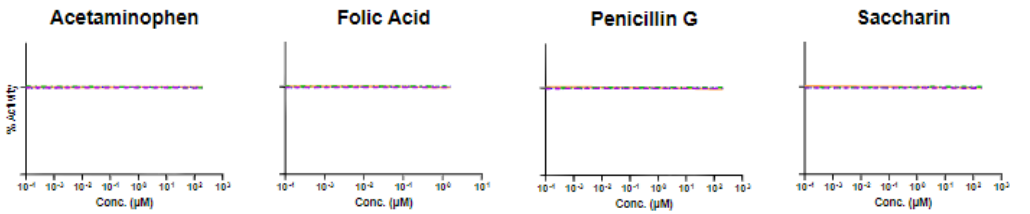
References

1. *Kameoka S. et al. (2014). Toxicol Sci. 137(1):76–90.*
2. *Gamble J. et al. (2022). Toxics. 10(7):392.*
3. *Martyn I et al. (2018). Nature. 558(7708):13–135.*

Reference Developmental Toxicants



Reference Non-Developmental Toxicants



DevTox Bioactivity Profile

P Pluri En Endo M Meso Ec Ecto

- DevTox GLR Endoderm and Pluripotent assays performed best, with at least one of the assays correctly identifying all developmental toxicants.
- Ectoderm assay was not as sensitive, and the Mesoderm assay was the least sensitive with higher variability between biological replicates.
- Overall, the DevTox GLR platform demonstrates reasonable predictivity and may provide insight into lineage-specific effects during gastrulation.

Performance Metrics

	Pluri	Ecto	Meso	Endo
rZ' Factor	0.8	0.9	0.9	0.8
Inter r%CV	6.4	4.3	4.5	5.3
Intra r%CV	17.7	8.9	26.5	6.4
Sensitivity	92%	67%	55%	92%
Specificity	100%	100%	100%	100%
Balanced Acc.	96%	83%	77%	96%

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Human Brain Organoid Model to Study Developmental Neurotoxicity

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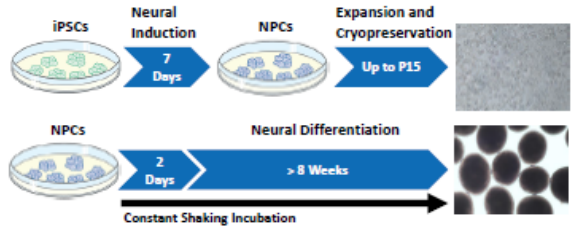
[2]Center for Computational Toxicology and Exposure, US EPA

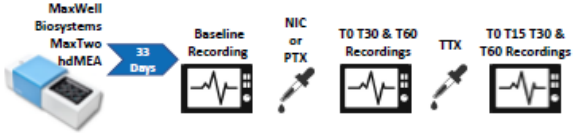
Background

Human *in vitro* models of the developing brain are important for studying neurodevelopment and the potential developmental neurotoxicity (DNT) of environmental and pharmacological compounds. Exposure to these compounds during neurodevelopment can potentially cause adverse effects such as morphological alterations and/or functional changes in the developing brain. Here, we sought to characterize the responses of a three-dimensional (3D) brain organoid model to three common pharmacological agents: nicotine (NIC), picrotoxin(PTX), tetrodotoxin (TTX). Based on literature, we hypothesis PTX and NIC will increase network activity while TTX will decrease network activity.

Methods



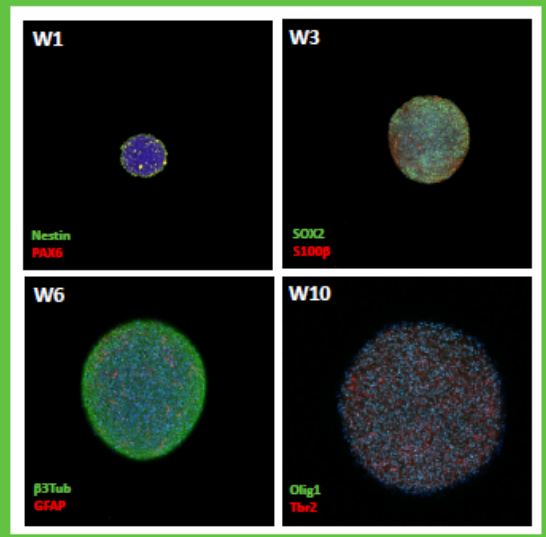
In this study, we establish and characterize an induced pluripotent stem cell (iPSC)-derived brain organoid model containing mature neurons and glial cells originally developed at Johns Hopkins University. To produce these organoids, iPSCs are first induced into neural progenitor cells (NPCs) which can be further cultured, expanded and cryopreserved. A single-cell suspension of NPCs is cultured for 2 days under constant gyratory shaking (88 rpm) before starting neural differentiation. Organoids can be cultured long-term (> 8 weeks) by changing the differentiation media every other day.



Neural organoids (3- & 4- weeks-old) are plated on a high-density microelectrode array (hdMEA) for 33 days to measure the spontaneous electric field potentials produced. After a baseline recording, 2 wells were treated with Nicotine (NIC) and 4 wells were treated with Picrotoxin (PTX) at final concentrations of 300µM and 25µM respectively. 10-minute recordings were taken after 0-, 30-, and 60-minutes. Then, Tetrodotoxin (TTX) was added to all 6 wells at a final concentration of 1µM and recordings were taken after 0-, 15-, 30-, and 60-minutes.

Disclaimer: The views expressed in this presentation are those of the authors and do not necessarily reflect the views or policies of the U.S. EPA. This project was supported, in part, by an appointment to the Research Participation Program at the Office of Research and Development administered by the Oak Ridge Institute for Science and Education through an interagency agreement with the U.S. Environmental Protection Agency.

Human brain organoid model on high-density microelectrode array shows decrease in network activity when exposed to Nicotine.



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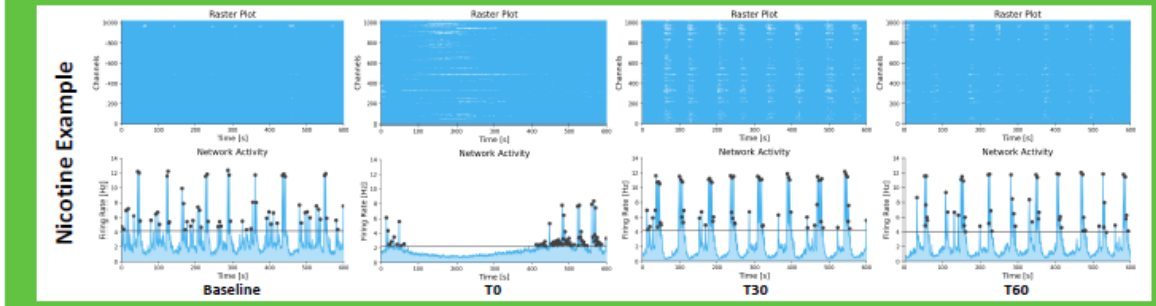

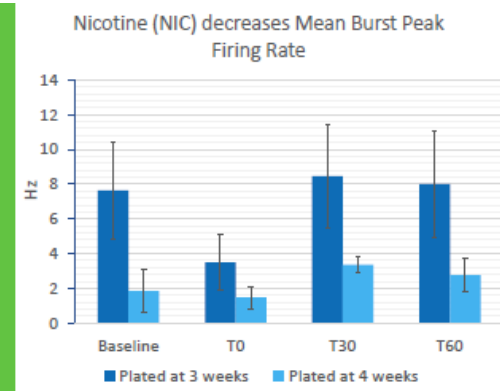

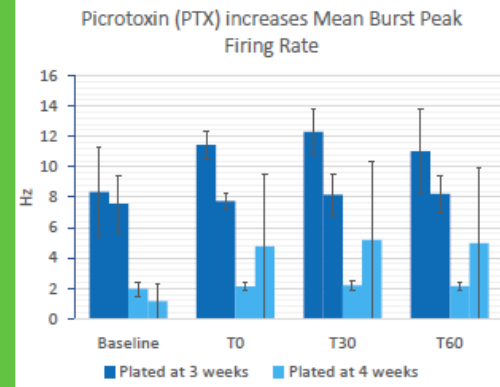

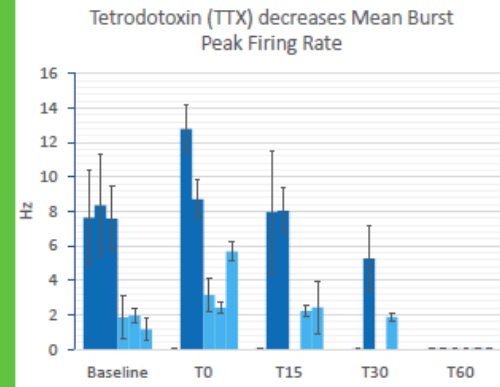

Conclusion and Future Work

This study demonstrates that neural organoids mimic the complex structure and development of the human brain and respond to blocking of $GABA_A$ receptors (PTX), voltage-gated sodium channels (TTX), and importantly activation of nicotinic acetylcholine receptors (NIC). The latter response is not robust in 2D rodent models. Future studies will develop an exposure protocol relevant to neurodevelopment and assess effects of additional compounds, including neonicotinoid insecticides and per- and polyfluoroalkyl substances (PFAS).

Optimization of an animal-component free system for high-throughput phenotypic profiling of human neural progenitor cells

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Abstract # 3793
Poster # P466

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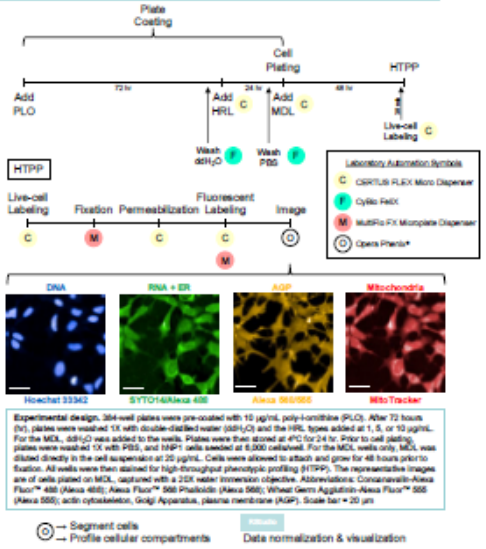
Background

- A worldwide effort to refine and/or reduce the use of animals for toxicity testing has prompted the development of new approach methods (NAMs).
- As such, our laboratory adapted a previously established high-throughput phenotypic profiling (HTPP) NAM for use with hNP1 human neural progenitor cells (Culbreth et al., 2022; DOI: 10.3389/ftox.2021803987)
- This approach, however, was optimized using a mouse-derived laminin (MDL) growth substrate, and thus still relies on animal products.
- Therefore, we wanted to determine whether substitution of the MDL with a human recombinant laminin (HRL) would produce similar phenotypic profiles in the hNP1 cells.
- Identification of an appropriate HRL type would allow for an animal component-free approach to developmental neurotoxicity (DNT) chemical hazard evaluation
- We selected four HRL types to examine:

Species	Manufacturer	Number	Description
Mouse	Sigma	L2020	supports growth and differentiation of many cell types
Human	BioLamina	LN111	general attachment protein for many cell types in vitro
		LN211	supports growth, survival, and differentiation of multiple cell types
		LN511	natural laminin for mouse embryonic stem cells
		LN521	natural laminin for pluripotent stem cells

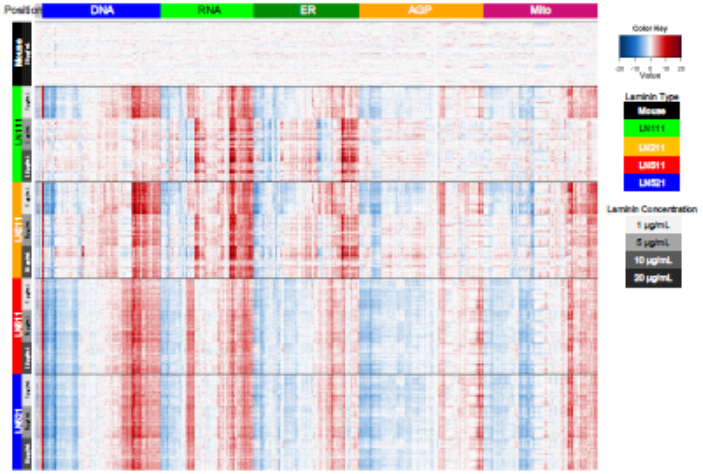
Experimental Design



Experimental design. 384-well plates were pre-coated with 10 µg/mL poly-l-ornithine (PLO). After 72 hours (hr), plates were washed 1X with double-distilled water (ddH$_2$O) and the HRL types added at 1, 5, or 10 µg/mL. For the MDL, ddH$_2$O was added to the wells. Plates were then stored at 4°C for 24 hr. Prior to cell plating, plates were washed 1X with PBS, and hNP1 cells seeded at 6,000 cells/well. For the MDL wells only, MDL was diluted directly in the cell suspension at 20 µg/mL. Cells were allowed to attach and grow for 48 hours prior to fixation. All wells were then stained for high-throughput phenotypic profiling (HTPP). The representative images are of cells plated on MDL, captured with a 20X water immersion objective. Abbreviations: Concanavalin-Alexa Fluor™ 488 (Alexa 488); Alexa Fluor™ 568 Phalloidin (Alexa 568); Wheat Germ Agglutinin-Alexa Fluor™ 555 (Alexa 555); actin cytoskeleton, Golgi Apparatus, plasma membrane (AGP). Scale bar = 20 µm

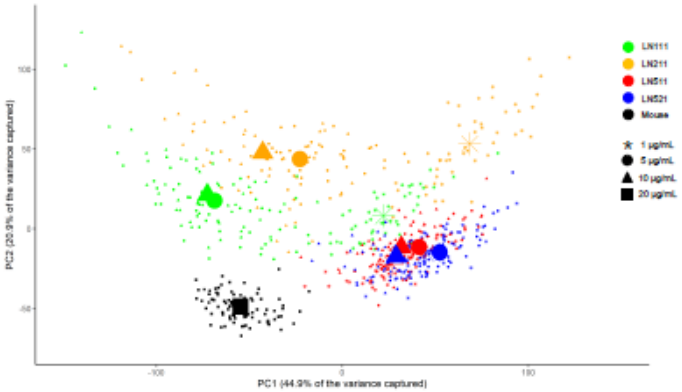
LN111 human recombinant laminin (HRL) is a comparable substrate to mouse-derived laminin (MDL) for growth of hNP1 human neural progenitors in the HTPP approach

HRL phenotypic profiles were qualitatively distinct from MDL



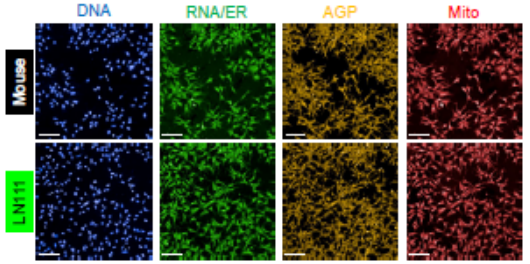
HTPP heat map. Cell-level data were exported to generate the heat map in R statistical software. Data represent three biological replicates (i.e., independent cultures) which includes 16 technical replicates/laminin type and concentration per biological replicate. The rows depict individual wells and are organized by laminin type and concentration; the columns depict individual features and are organized by channel. Heat map coloring represents the size and direction of the phenotypic effect relative to MDL [Mouse (20 µg/mL)]. Abbreviations: actin cytoskeleton, Golgi Apparatus, plasma membrane (AGP); mitochondria (Mito); microgram (µg); milliliter (mL)

HRL phenotypic profiles did not cluster with MDL



HTPP PCA. Cell-level data were exported to generate the PCA in R statistical software. Data represent three biological replicates (i.e., independent cultures) which includes 16 technical replicates/laminin type and concentration per biological replicate. On the x-axis is the first principal component (PC1) which captures 44.9% of the variance in the data, and on the y-axis is the second principal component (PC2) which captures 20.9% of the variance in the data. Enlarged shapes indicate the centroid for each laminin type and concentration.

LN111 produced the more similar phenotypic profile to MDL



Representative images. hNP1 cells were plated at 6,000 cells/well on MDL [Mouse (20 µg/mL)] or LN111 (5 µg/mL). Images were captured on the Opera Phenix High-Content Screening System with a 20X water immersion objective and Harmony® v4.9 software. Cell nuclei (blue) were captured in the DNA channel. Nucleoli and endoplasmic reticulum (green) were captured in the RNA or ER channels, respectively. The actin cytoskeleton, Golgi apparatus, and plasma membrane (yellow) were captured in the AGP channel, and mitochondria (red) were captured in the Mito channel. Abbreviations: microgram (µg); milliliter (mL). Scale bar = 100 µm

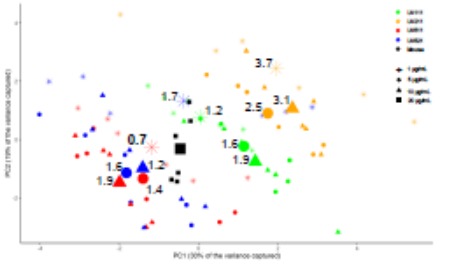
The shorter distance between centroids indicated LN111 is most comparable to MDL

Laminin Type	Concentration	Centroid Distance
Mouse	20	-
LN111	1	95.7
	5	67.8
	10	72.3
LN211	1	160.1
	5	97.9
	10	97.3
LN511	1	85.7
	5	103.1
	10	94.1
LN521	1	105.2
	5	112.5
	10	89.4

The distance between centroids of each laminin type and concentration relative to mouse were calculated as: centroid distance = $sqrt{(PC1_{HRL} - PC1_{MDL})^2 + (PC2_{HRL} - PC2_{MDL})^2}$. Concentration units are in micrograms/milliliter (µg/mL).

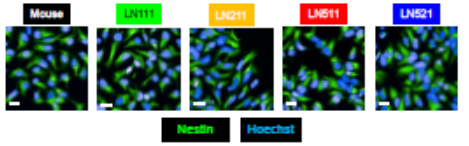
HRL types exhibited only minor differences in whole transcriptome profiles and neural marker expression relative to MDL

Laminin type had minimal impact on gene expression



Whole transcriptome PCA. Normalized count data were exported to generate the PCA in R statistical software. Data represent two biological replicates (i.e., independent cultures) which includes 3 technical replicates/laminin type and concentration. On the x-axis is the first principal component (PC1) which captures 33% of the variance in the data, and on the y-axis is the second principal component (PC2) which captures 19% of the variance in the data. Enlarged shapes indicate the centroid for each laminin type and concentration. The number associated with each centroid is the distance relative to mouse, calculated as: centroid distance = $sqrt{(PC1_{HRL} - PC1_{MDL})^2 + (PC2_{HRL} - PC2_{MDL})^2}$. Abbreviations: microgram (µg); milliliter (mL)

hNP1 cells expressed nestin regardless of laminin type



Neural marker expression. hNP1 cells plated on respective laminin types were immunostained with Nestin (green), a neural progenitor cell marker. Cell nuclei were fluorescently labeled with Hoechst 33342 (blue). Images were captured on the Opera Phenix High-Content Screening System with a 20X water immersion objective and Harmony® v4.9 software. For the HRL types, all representative images are at 5 µg/mL. Abbreviations: microgram (µg); milliliter (mL) Scale bar = 20 µm

Conclusions

- The phenotypic profiles of hNP1 cells plated on HRL did not wholly resemble that of MDL.
- LN111 produced the most similar phenotypic profile to MDL, and therefore could be applied as a substitute growth substrate in the HTPP approach.
- While LN111 and MDL were comparable, this does not necessarily indicate hNP1 cells grown on either substrate will entirely recapitulate relevant human biology.
- Most historical DNT NAMs chemical data, however, were acquired from assays optimized for MDL; thus, use of LN111 offers greater comparative power to established datasets.

Acknowledgments and Funding

The authors would like to thank Joseph Sundy for guidance on the centroid distance analysis and Kelly Carstens, Derik Haggard, and Logan Everett for assistance with the HTTr data analysis. The U.S. EPA through its Office of Research and Development (ORD) provided funding for this research. The views expressed on this poster, however, are those of the authors and do not necessarily reflect U.S. EPA policies. Reference to commercial products does not constitute endorsement.

Additional Resources

Financial Times Visual Vocabulary is a useful tool for determining what kind of graph to use:

https://github.com/Financial-Times/chart-doctor/blob/main/visual-vocabulary/FT4schools_RGS.pdf

Visual Capitalist uses high quality data visualizations for effective data storytelling:

https://www.visualcapitalist.com/

Visual Examples of Chart Designs by Software

https://www.visualisingdata.com/resources/

Thank you for your time & attention!

Questions?

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