Improved Promoter Reporter Technology For Understanding Gene Regulation

GeneCopoeia, Inc.

Presenter:

Ed Davis, Ph.D.
Senior Application Scientist
GeneCopoeia, Inc.

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Goals of this presentation

- Overview of promoter reporter technology
- GeneCopoeia's vast collection of reporter clones with high-performance reporter technology

Quick survey

Why are you attending this webinar?

Please type your answers (A, B, C, or D) in the question box

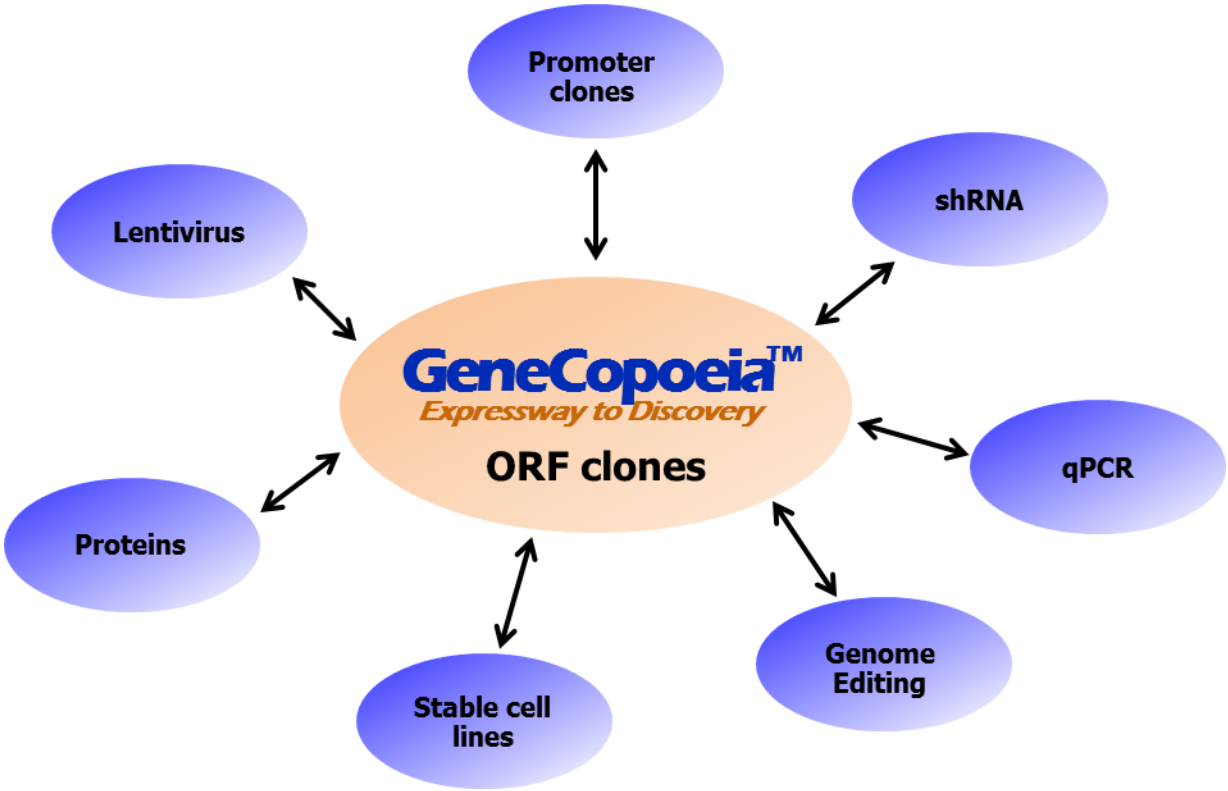
A. I want to use promoter clones & want to learn more.

B. I heard about this technology & I'm just curious.

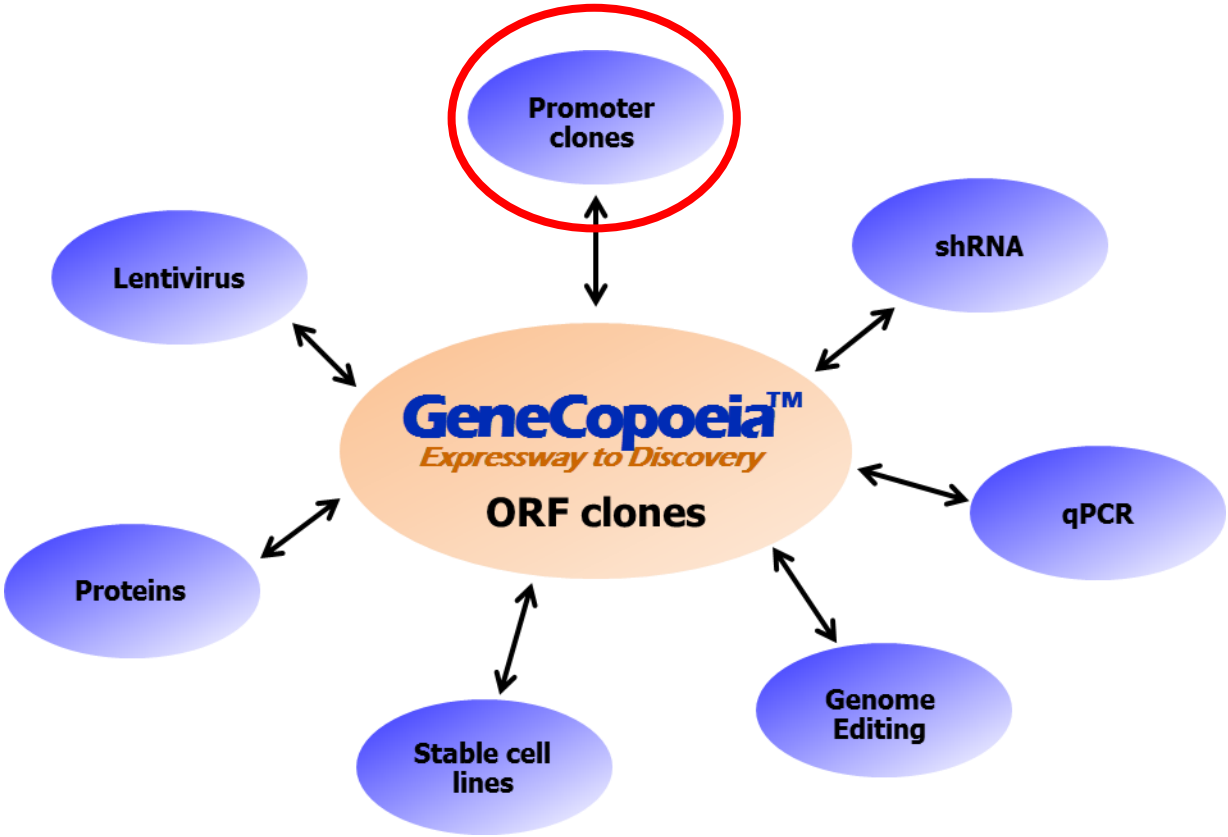
C. I plan to use reporter clones & want to see what your company has to offer.

D. Other (please explain).

GeneCopoeia Products & Services



GeneCopoeia Products & Services



Outline

- Promoter reporter clones: Technologies
  - Standard reporters
  - Dual reporters
- Applications for promoter reporters
- How GeneCopoeia can provide you the promoter reporter clone you need

Post-genomics era functional studies

Human Genome Project

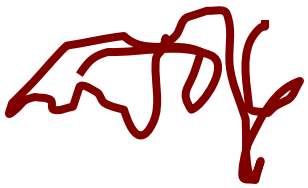
DNA

Post-Human Genome Project:
ORFs and Expression Regulation

mRNA

Reverse Transcription

Protein



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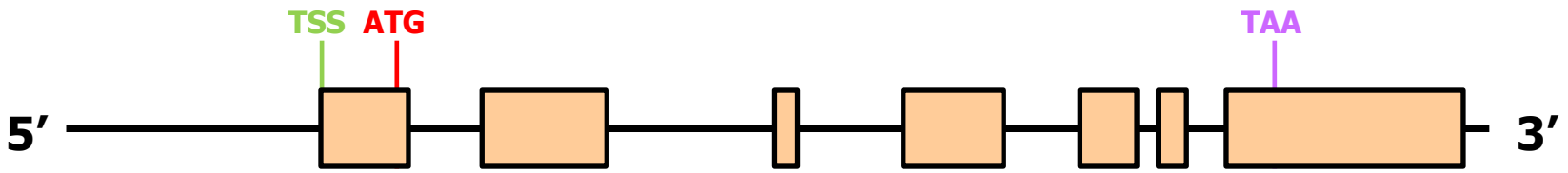
Promoter Clone Collections

- Human promoters
  - 20,000+ promoters
- Mouse promoters
  - 18,000+ promoters

How do we define a promoter?



- Functional: TSS-proximal DNA sequence elements required for native gene expression
- Excludes distal enhancers
- Most mammalian promoters have not been functionally defined

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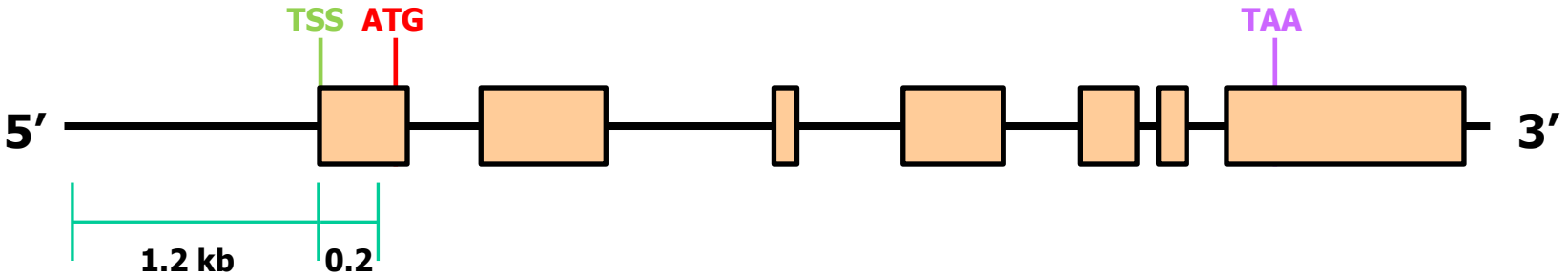
How do we define a promoter?



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Why use promoter clones?

- Study regulatory patterns of gene expression in different tissues
- Determine regulatory proteins that bind promoter sequence
- Determine binding elements for activator and repressor molecules
- Use as biosensors for physiological or environmental stimuli

What is a promoter clone?

- Putative promoter region of interest cloned upstream of reporter ORF in expression vector
- Expression followed either qualitatively (visually) or quantitatively

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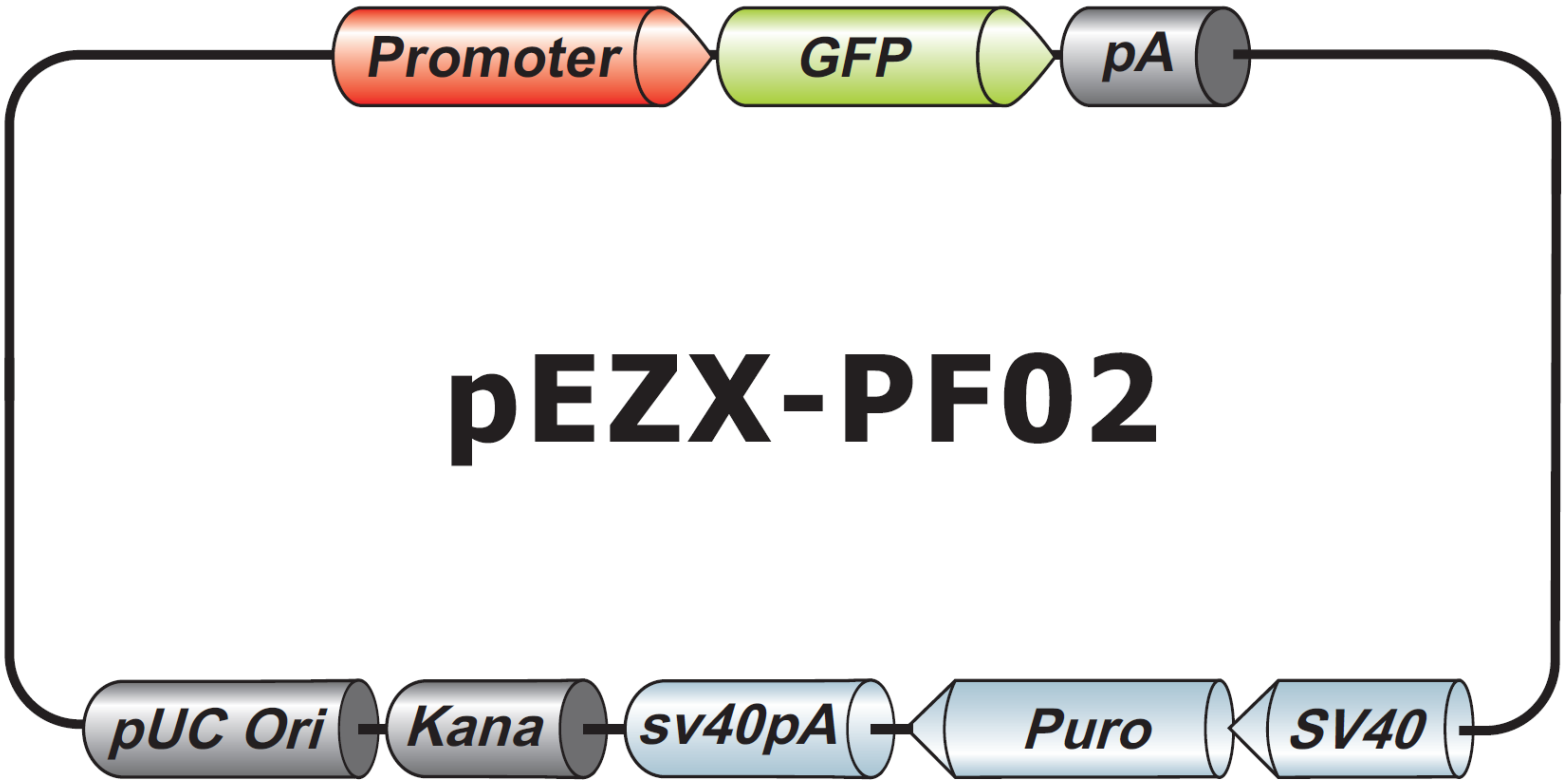
GLuc-ON™ Promoter reporter clones



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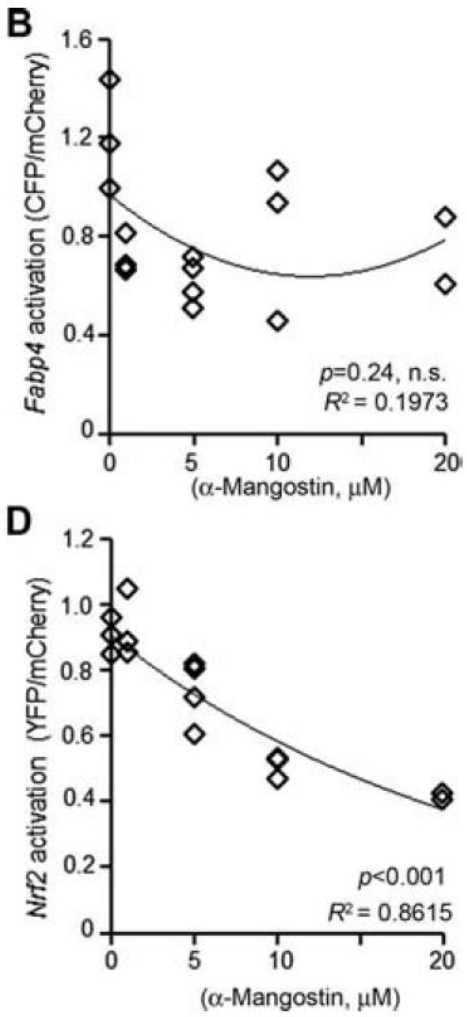
Case study: GLuc-ON™ clones



- Used GLuc-ON™ clones Fabp4 promoter-CFP and Nrf2 promoter-YFP to assay responsiveness to α-Mangostin in adipocytes
- Normalization control: GeneCopoeia mCherry clone

Shen, et al. (2014). Mol. Nutr. Food Res. 58, 239

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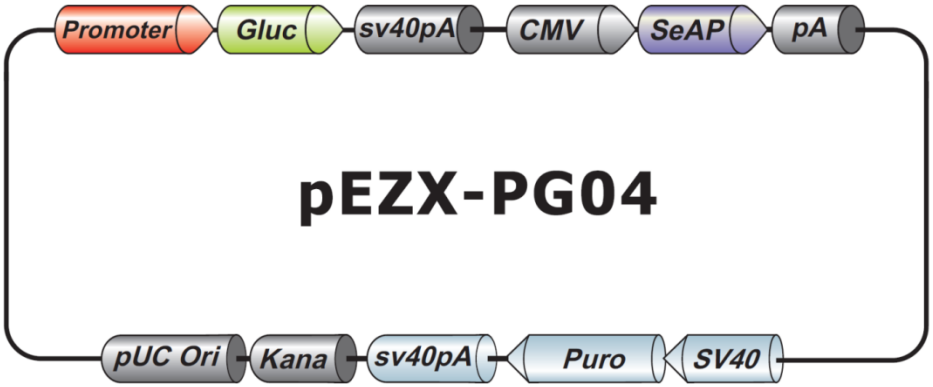
GLuc-ON™ Promoter reporter clones

Limitation of traditional reporters

- Quantitative measurement difficult. Must lyse cells. Cannot measure in real time.

GLuc-ON™ Promoter reporter clones

GeneCopoeia dual secreted reporter promoter clones



Advantages

- Test and control reporters on same vector
- Both reporters secreted. No need to lyse cells
- Activity can be measured multiple times on the same sample
- Gluc is 1,000-fold more sensitive than Fluc or Rluc
- Compatible with Secrete-Pair™ Dual luminescence assay kit

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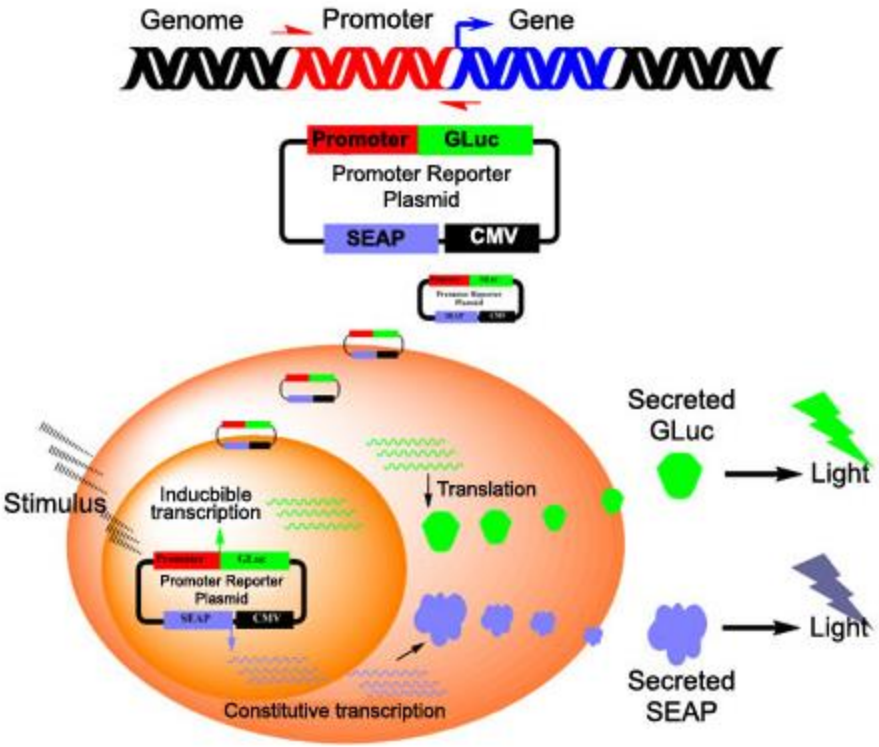
GLuc-ON™ Promoter reporter clones

GeneCopoeia dual secreted reporter promoter clones



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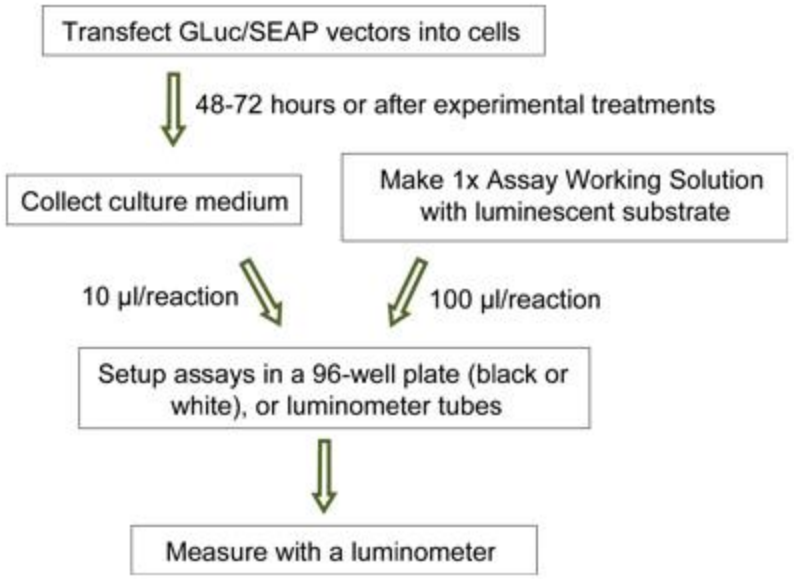
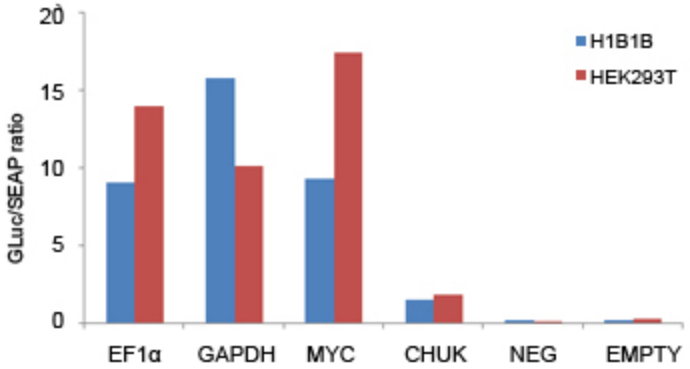
Secrete-Pair™ kit





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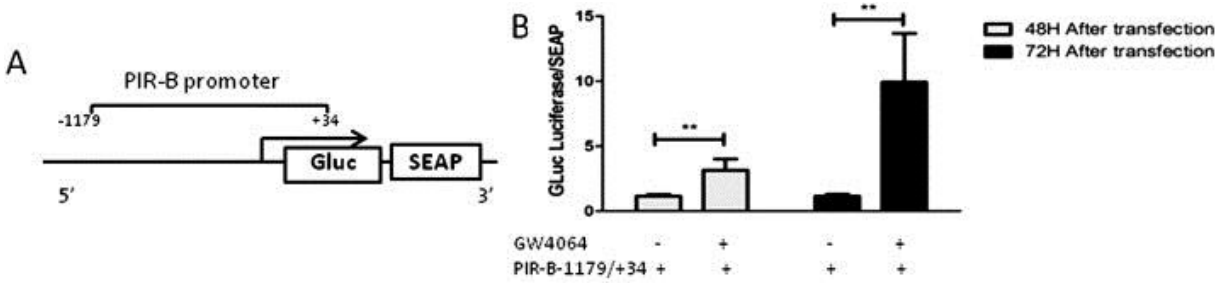
Case study: GLuc-ON™ clones

- Used GLuc-ON™ Gluc-SEAP dual reporter clone to analyze stimulation of PIR-B expression by FXR (bile acid receptor) gene activation in a mouse monocyte/macrophage cell line
- Used molecule GW4064 to activate FXR
- Assayed Gluc and SEAP activity using the Secrete-Pair™ Dual luminance assay kit



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GLuc-ON™ Promoter reporter clones

How do I order GLuc-ON™ promoter reporter clones from GeneCopoeia?

GLuc-ON™ Promoter reporter clones

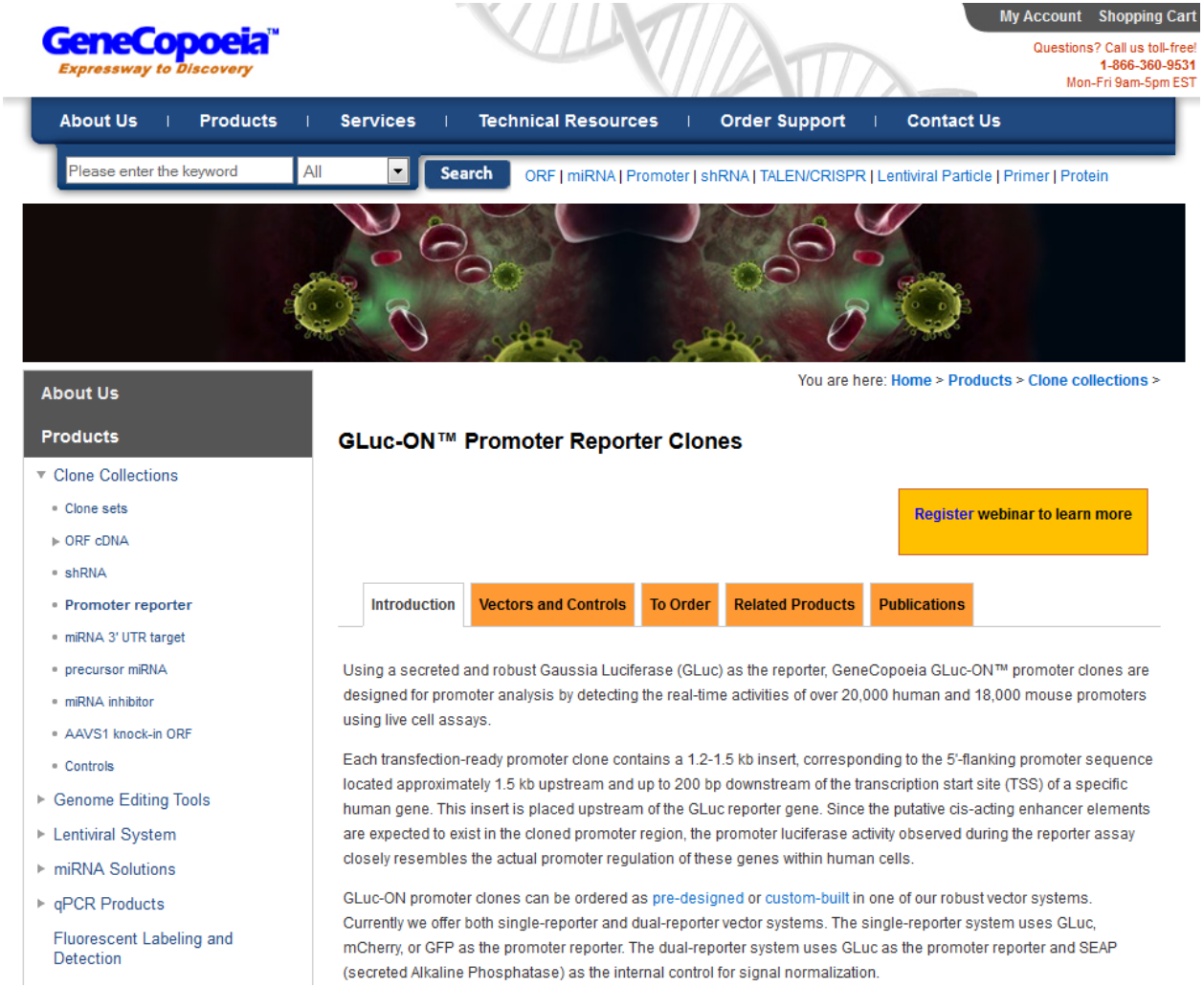
Publications using GLuc-ON™ promoter reporter clones

- Zhang, H. *et al.* (2014). The critical role of myeloid-derived suppressor cells and FXR activation in immune-mediated liver injury. *Journal of Autoimmunity*, *in press*.
- Rubie, C. *et al.* (2014). Chemokine receptor CCR6 expression is regulated by miR-518a-5p in colorectal cancer cells. *Journal of Translational Medicine* **12**, 1479
- Klimosch, SN. *et al.* (2013) Functional TLR5 genetic variants affect human colorectal cancer survival. *Cancer Research* **73**, 7232
- Shen, Q. *et al*. (2013). Adipocyte reporter assays: Application for identification of anti-inflammatory and antioxidant properties of mangosteen xanthones. *Molecular Nutrition & Food Res*earch**58**, 239
- Celardo, I. *et al*. (2013). Caspase-1 is a novel target of p63 in tumor suppression. *Cell Death and Disease* **4**, e645
- Zheng, H. *et al*. (2013). Glycogen synthase kinase-3 beta regulates Snail and β-catenin expression during Fas-induced epithelial–mesenchymal transition in gastrointestinal cancer. *European Journal of Cancer* **49**, 2734
- Mills, L.D. *et al*. (2013). Loss of the Transcription Factor GLI1 Identifies a Signaling Network in the Tumor Microenvironment Mediating KRAS-Induced. *Journal of Biological Chem*istry **288**, 11786
- Schank, JR. et al (2013) Tacr1 Gene Variation and Neurokinin 1 Receptor Expression Is Associated with Antagonist Efficacy in Genetically Selected Alcohol-Preferring Rats. Biological Psychiatry Journal **73**, 774
- Petrella, B. et al. (2012) Interleukin-1 beta and transforming growth factor-beta 3 cooperate to activate matrix metalloproteinase expression and invasiveness in A549 lung adenocarcinoma … Cancer Letters, Volume **325**, 220

Summary

- Promoter reporter clones are useful for many applications in the analysis of gene expression
- Traditional promoter clones use fluorescent reporters or firefly/renilla luciferase, which require cell lysis for quantitative measurement
- GeneCopoeia GLuc-ON™ promoter reporter clones offer higher sensitivity and ease-of-use over traditional fluorescent and luminescent reporters
- Customers can easily purchase GeneCopoeia GLuc-ON™ promoter reporter clones by searching our database of nearly 40,000 human and mouse promoters

GLuc-ON™ Promoter clone website



http://www.genecopoeia.com/product/promoter-reporter-clones/?category_name=product&PHPSESSID=sq5o5lla337nk1n3hnc8gmacf7

Upcoming webinar!

CRISPR & TALEN In Mammalian Cells: What Do I Do Next?

Wednesday, September 24, 2014 1:30 pm EDT

Register here:

https://attendee.gotowebinar.com/register/3793592553033800450

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Thank you!

If you have any additional questions, please call

1-866-360-9531 x227

Email: edavis@genecopoeia.com

Or visit us on the web:

www.genecopoeia.com

GeneCopoeia, Inc.

9260 Medical Center Drive Suite 101

Rockville, Maryland USA 20850

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