Module 2 Remapping the Body of the World

Evidence worksheet\_04 “Bacterial Rhodopsin Gene Expression”

**Learning objectives:**

* Discuss the relationship between microbial community structure and metabolic diversity
* Evaluate common methods for studying the diversity of microbial communities
* Recognize basic design elements in metagenomic workflows

**General Questions:**

• *What were the main questions being asked?*

* What are the various specific functions and physiological roles of diverse marine microbial PRs
* What are the specific genetic and biochemical roles of the genes involved in the photosystem
* Understand the function and structure of the PR photosystem

• *What were the primary methodological approaches used?*

* Performing a functional screen
* we directly screened large-insert DNA libraries derived from marine picoplankton for visibly detectable PR-expressing phenotypes
* used the copy control system present in our fosmid vector that allowed a controlled transition from one copy per cell to multiple (up to 100 copies)
* found 3 colonies which could express the orange pigment and further analyzed two clones HF1019P19 and HF1025F10 for PR gene expression and function
* why use single copy vs multiple copy
  + fosmids are really large (40kb) having too many copies can increase burden + lead to homologous recombination (lots of insertions and deletions) but need more copies of DNA for functional screening and purification

• *Summarize the main results or findings.*

* Take environmental dna, programmed e coli with it and screened for a phenotype that e coli does not traditionally display
* You can program a host with sufficient information from the environment + have a functional screen to identify the minimal set of genes needed to get that phenotype (implications – if there are fewer genes the chances of HGT, under selective pressure, would be present in multiple different lineages
* Access to information that would otherwise be unavailable
* With retinol and light -> tehre was a pH change
* Different lights – spectrotuning
* Gene was transferable between bacteria and archae -

• *Do new questions arise from the results?*

• *Were there any specific challenges or advantages in understanding the paper (e.g. did the authors provide sufficient background information to understand experimental logic, were methods explained adequately, were any specific assumptions made, were conclusions justified based on the evidence, were the figures or tables useful and easy to understand)?*

* I wasn’t sure of why they had controlled copy vectors (what is the purpose of inserting either one or multiple copies of the fosmid?)