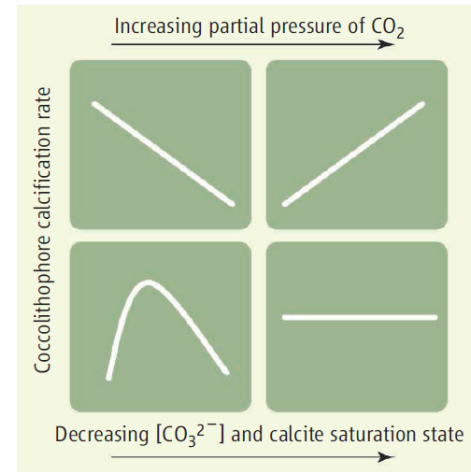
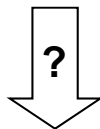
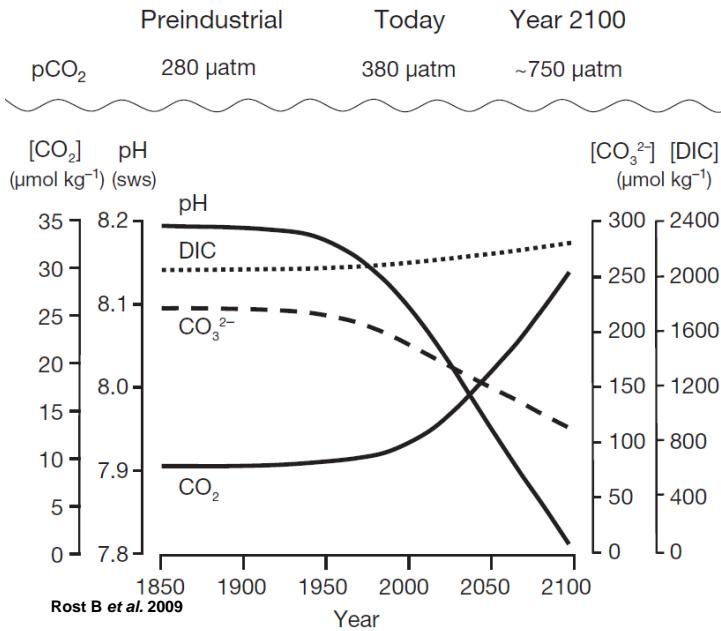


Response of *Emiliana huxleyi* to a High CO₂ World: Assessing the Extent of Genetic Diversity in the Pattern of Gene Expression

Frédéric Verret, Declan Schroeder, Colin Brownlee (MBA)
Stewart Finch, Richard Geider, Christine Raines, David Suggett (University of Essex)
(SOFI Award)

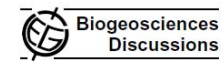


Emiliana huxleyi Response to Increasing pCO₂ is Strain-Specific



Fabry VJ *et al.* 2008

Biogeosciences Discuss., 6, 4361–4383, 2009
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Strain-specific responses of *Emiliana huxleyi* to changing seawater carbonate chemistry

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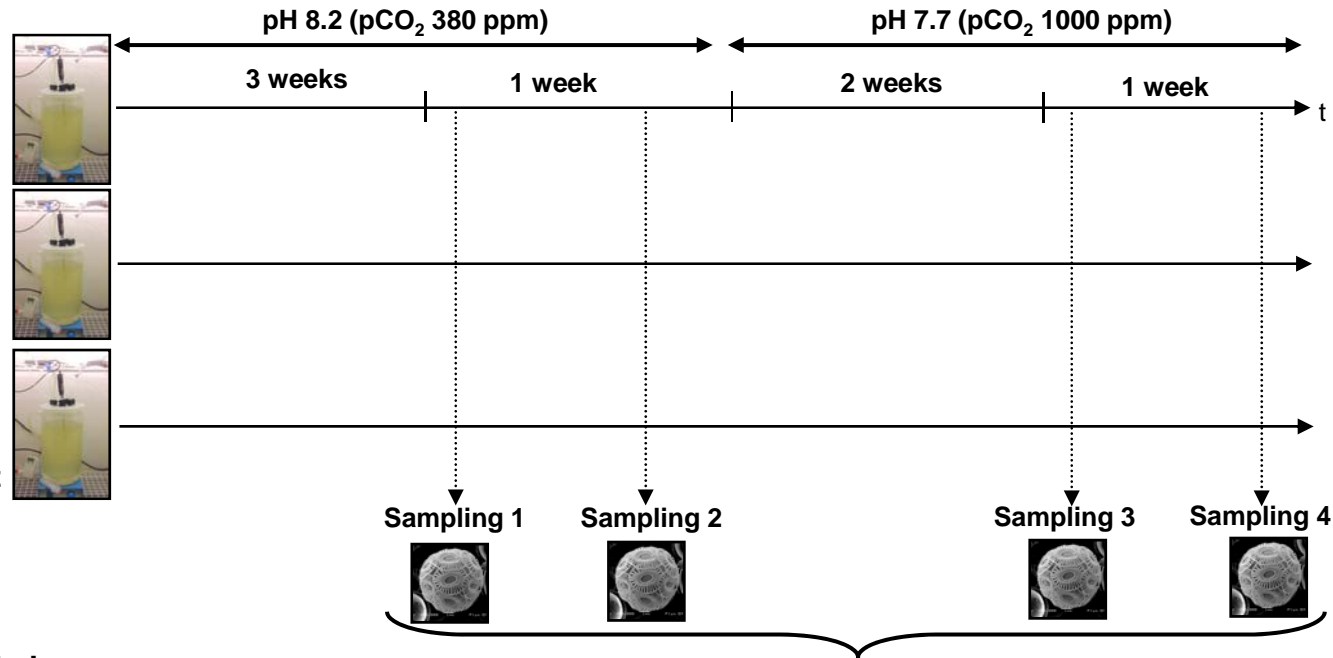
Correspondence to: G. Langer (gerald.langer@awi.de)

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- ➡ Gene expression pattern & physiology related to the strain-specific responses ?
- ➡ Commonality between different *E. hux* strains ?

Experimental Design: Different *E. huxleyi* Isolates Grown in pH-stated Cultures

<i>Coccolithes</i>				
Name	Calcification	morphology	Isolation date	Origin
CCMP1516	++	A	1991	North Pacific
CH25	+	B	1990	North Sea
NZEH	+++	R	1992	South Pacific



- Cultures in controlled environment (light, temperature, nutrients).
- Continuous measurement of pH by pH probe/computer.
- pH stated by computer controlling pumping of CO₂-free /CO₂-enriched air.

Transcriptomic using new technologies:

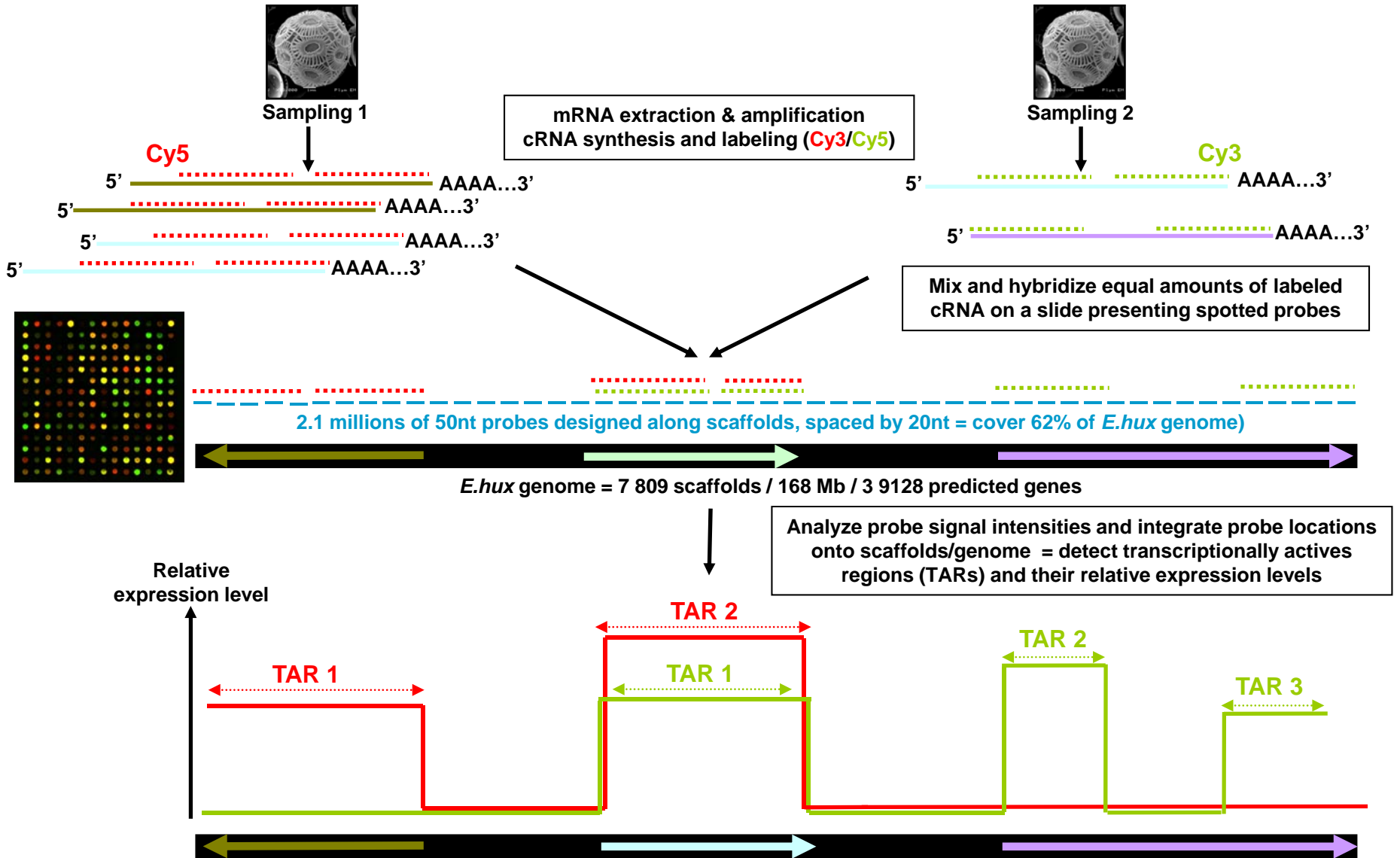
- Whole Genome Tiling microarrays (NimbleGen®)
- Digital Gene Expression (Illumina®)

Physiology:

(Coccosphere Integrity, PIC & POC, Photosynthesis...)

Transcriptomic Analysis using Whole Genome Tiling Microarrays (NimbleGen®)

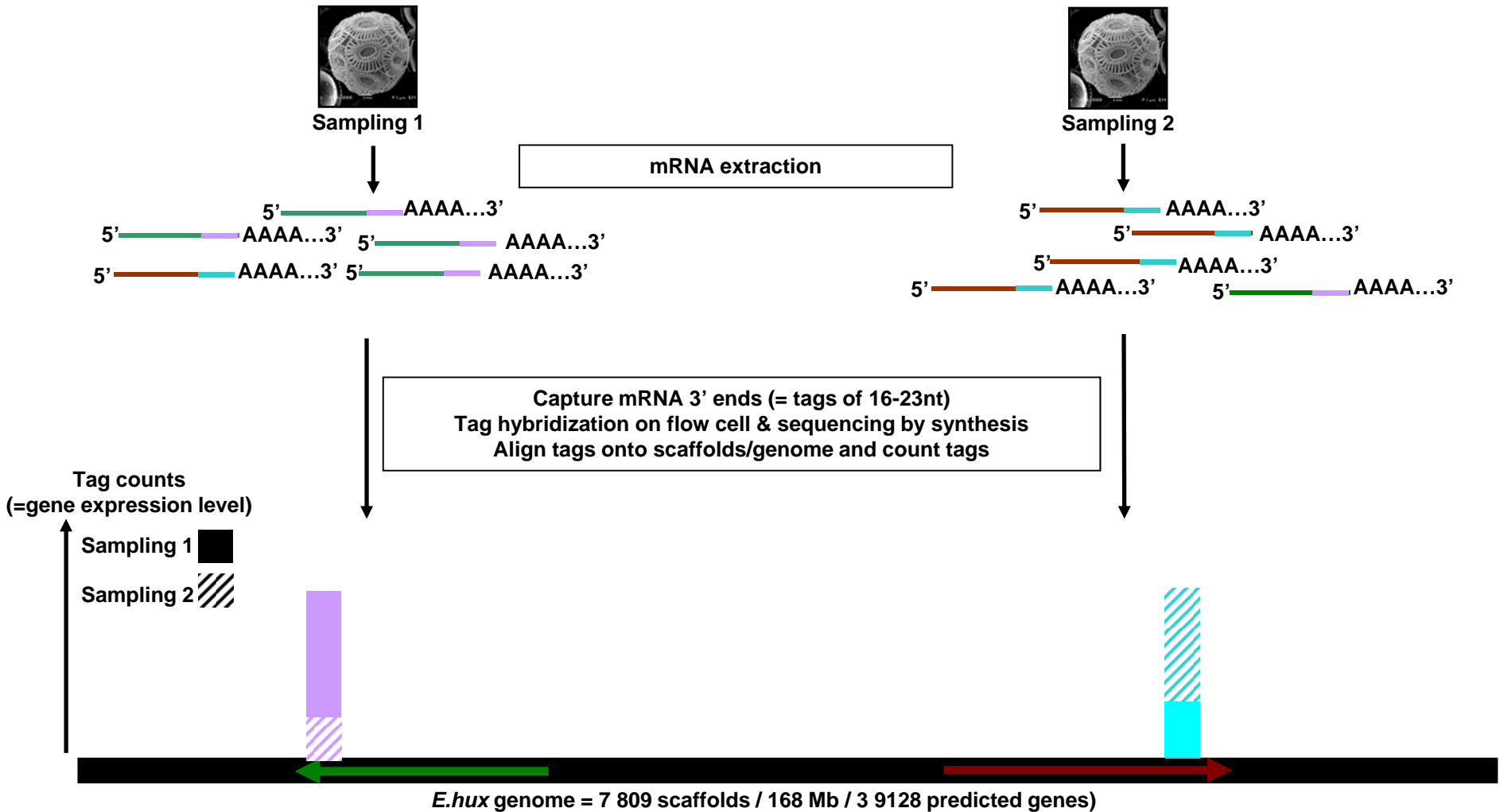
Tiling Microarrays = Hybridization-based approach using high-density probes designed from genome information



- ➡ Advantages: multi probes/gene, identify non predicted TAR.
- ➡ Disadvantages: based on *a priori* genome information, hybridization bias.

Transcriptomic Analysis using Digital Gene Expression (Illumina®)

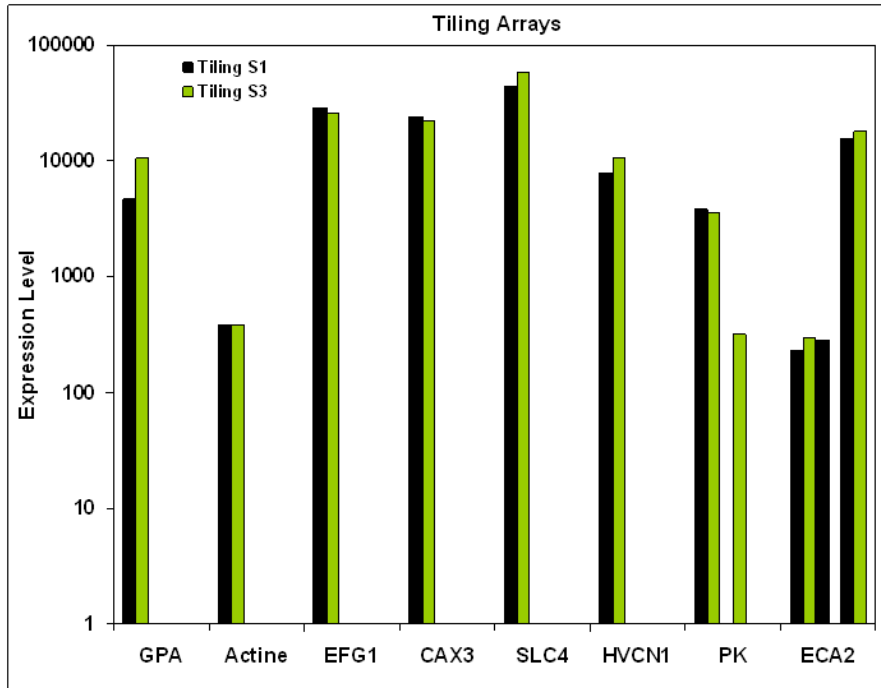
Digital Gene Expression = Tag-based approach by 3' end mRNA sequencing without *a priori* genome information



- ➡ **Advantages:** not based on *a priori* genome information, identify non predicted genes, high sensitivity, no hybridization bias.
- ➡ **Disadvantages:** only one tag per gene, low specificity/mapping capacity due to short tags length.

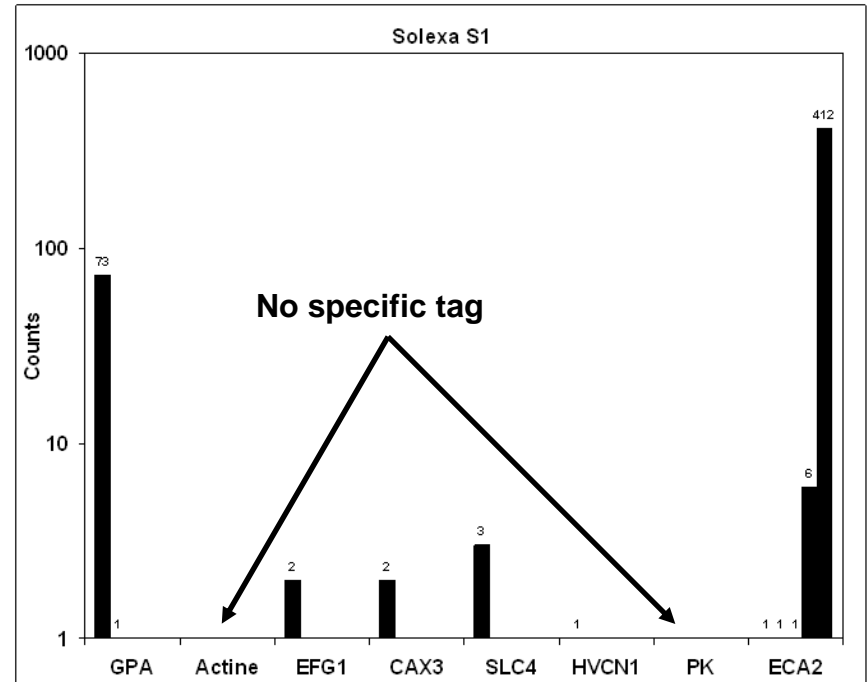
Whole Genome Tiling Arrays vs Digital Gene Expression

Transcriptome of *E.hux* grown at standard pCO₂:



Whole Genome Tiling Arrays on cultures in triplicate sampled at 2 time points:

- 60 000 TARs detected (39 128 predicted genes).
- Good reproducibility between technical replicates (dyes swap & hybridizations in triplicate) and between biological replicates.



Digital Gene expression on one culture sampled at 1 time point:

- 13.4 millions tags sequenced = 700 000 unique tags.
- Only 23% of the unique tags can be perfectly aligned onto the genome!
- Only 9% of the aligned tags have a unique location (probably due to the short tags length, repetitive sequences, orthologues).
- Some house keeping genes do not have a specific tag (e.g. Actin, Pyruvate Kinase....but are detected using whole genome tiling arrays).

➔ Whole genome tiling arrays look more adequate to analyze *E.hux* transcriptome...will be used for next experiments (low/high pCO₂ & inter-isolate comparisons)