

Amm_transcriptome_results_update2

We investigated the issue of second cloud of female-biased genes in brain tissues, it turned out that 30% transcripts does not have any BLAST hit (88%) with new XX genome.

We removed the non-frog transcripts, and redone all analysis using the updated transcriptome.

A. Tissue specificity index tau

B. Sex-biased gene expression along developmental stages, across adult tissues

C. Transcription degeneration

D. Coding sequence divergence and Faster-X

A. Tissue specificity index tau

Approach:

1. Define intotal 11 tissues, treating somatic tissues with ~~combined sexes~~ and gonad tissues in ~~separate sexes~~ (see Brown & Bachtrog 2014).
2. Use Kallisto to quantify transcript expression, ~~use~~ ^{ppf like} the generated output of TPM matrix.
3. Use tissue specificity index Tau formula below (Mank et al. 2008; Brown & Bachtrog 2014):

$$\tau = \frac{\sum_{i=1}^N 1 - \frac{\log E_i}{\log E_{\max}}}{N - 1},$$

where E is ~~row/normalized~~ expression by tissue per gene

↙ is log part of the ~~log~~ values above?

4. Keep transcripts which ~~the expression~~ are expressed in at least one of the 11 tissues with TPM ≥ 2 .

Figure 1. Histogram distribution of Tau for all transcripts in Ammarnas transcriptome.

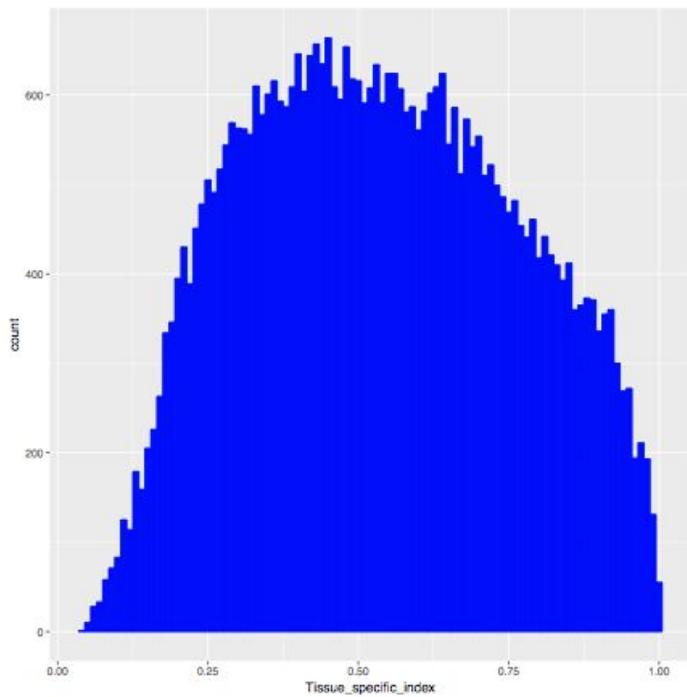


Figure 2. Boxplot of sex-biased genes and the tissue specificity index Tau at G46 stage (sex-biased genes are not including the gene expression from sex-reversed XX male).
we defined after excluding the

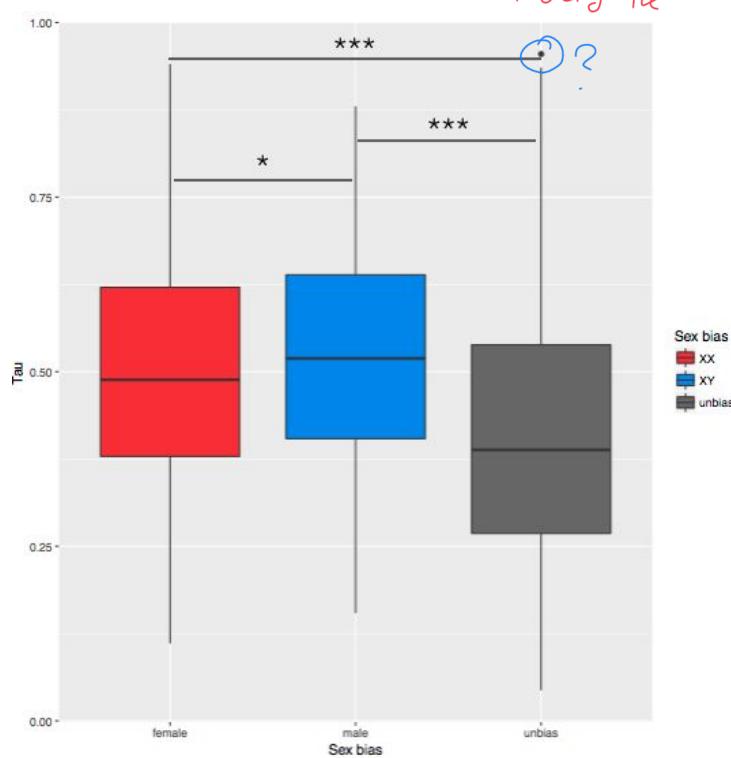


Figure 3. Correlation between ~~absolute values~~ of gene expression ratio $\text{Log2}(\text{XY46}/\text{XX46})$ (include sex-biased and unbiased genes) and tissue specificity index tau.

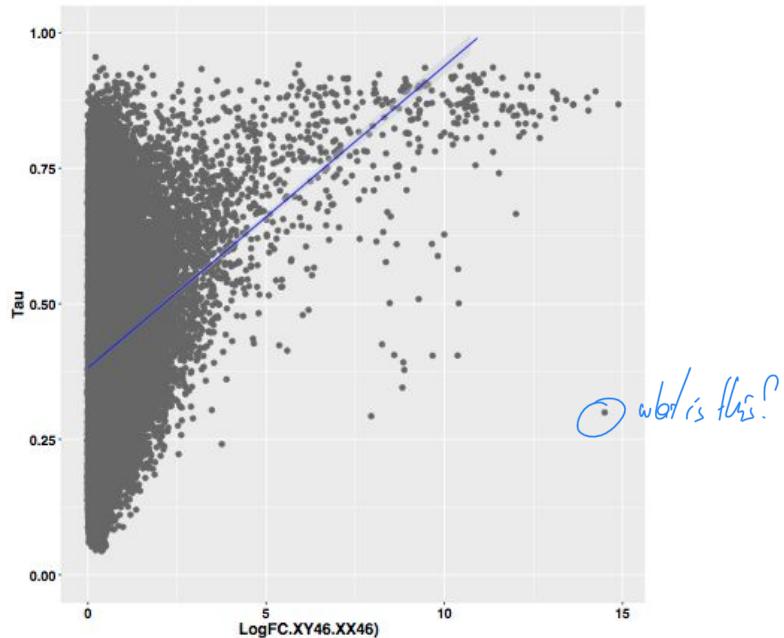


Figure 4. Boxplot of sex-biased genes and the tissue specificity index Tau in gonad tissues.

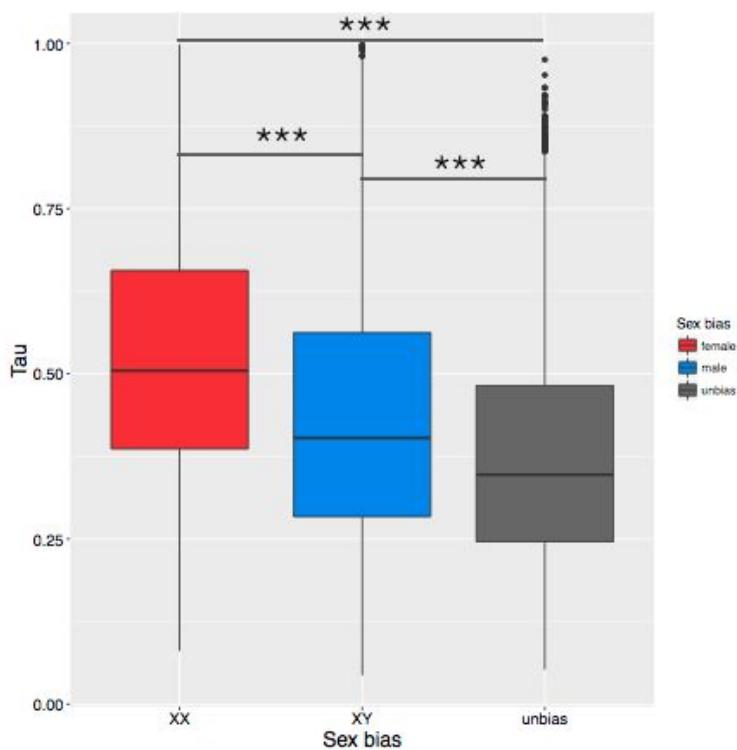
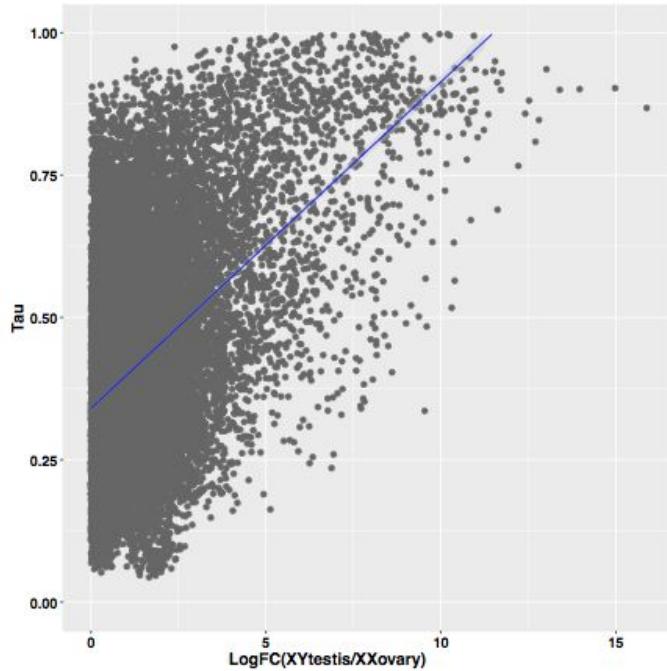


Figure 5. Correlation between ~~absolute values of~~ gene expression ratio $\text{Log2}(\text{male/female})$ (only for sex-biased and unbiased genes) and tissue specificity index tau.



which genes are missing?

ok for correlation on non-mutual data?

better to do Log (LogFC) ?

More analysis will come along the way, now it is just to get some idea on the patterns and trigger further related questions.

B. Sex-biased gene expression along developmental stages, across adult tissues

cleared up transcriptome including only transcripts mapping to frog genome
Approach: the transcriptome with removed-non-frog-transcripts is used, we quantify the transcript abundance with Kallisto.

~~The~~ EdgeR is applied to analyze differential or sex-biased gene expression across developmental stages, as well as adult tissues.

The selection criteria to remove ~~lowly~~ expressed transcripts are: average LogCPM >0 and LogCPM >1 is present in at least half of the tissues per sex.

both 0 and 1?

Figure 6. Number of sex-biased genes throughout development and adult tissues (FDR < 0.05, Log2 >=1).

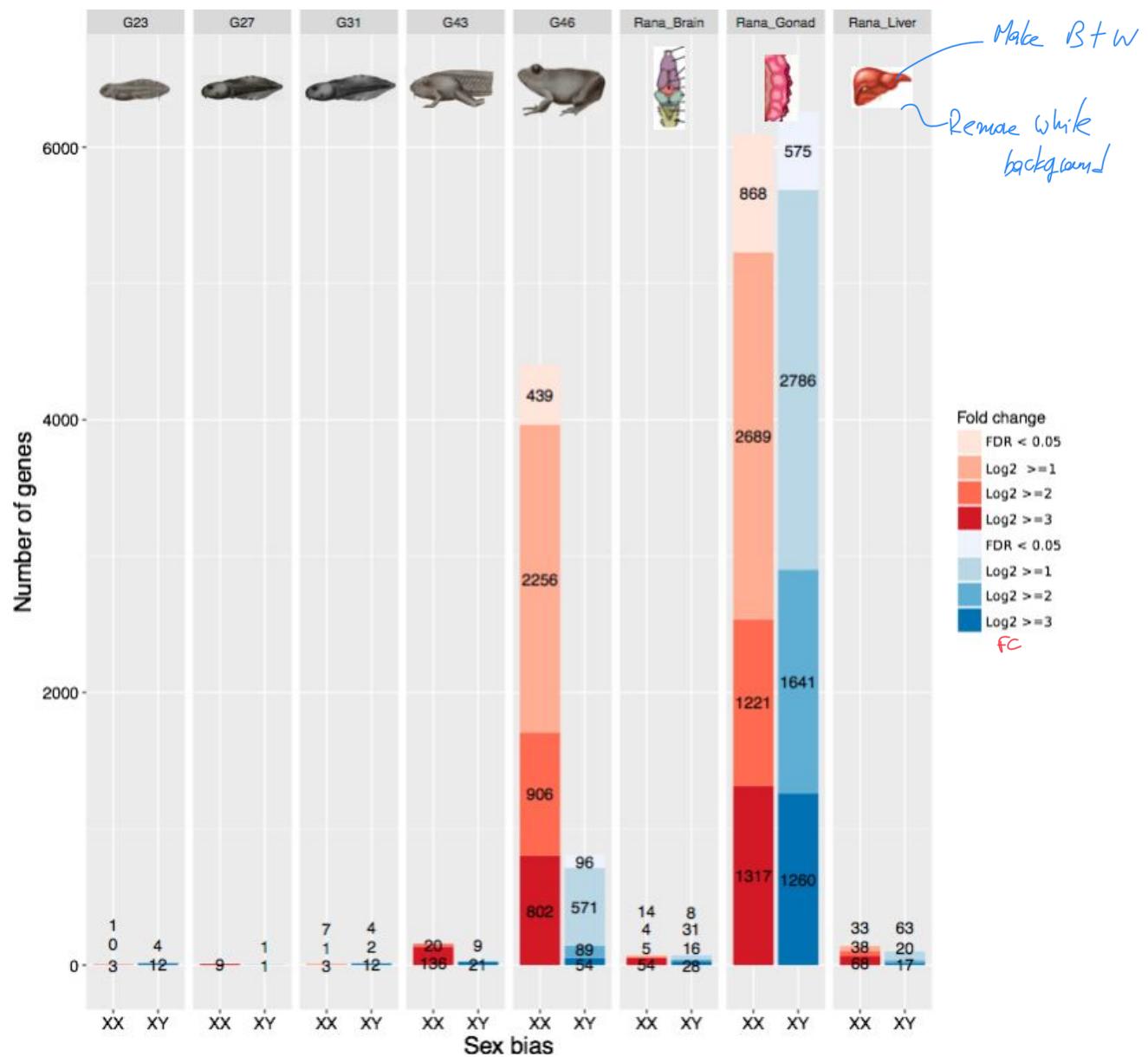


Figure 7. Shared sex-biased genes (a), shared female-biased genes (b) and shared male-biased genes (c) throughout development stages.

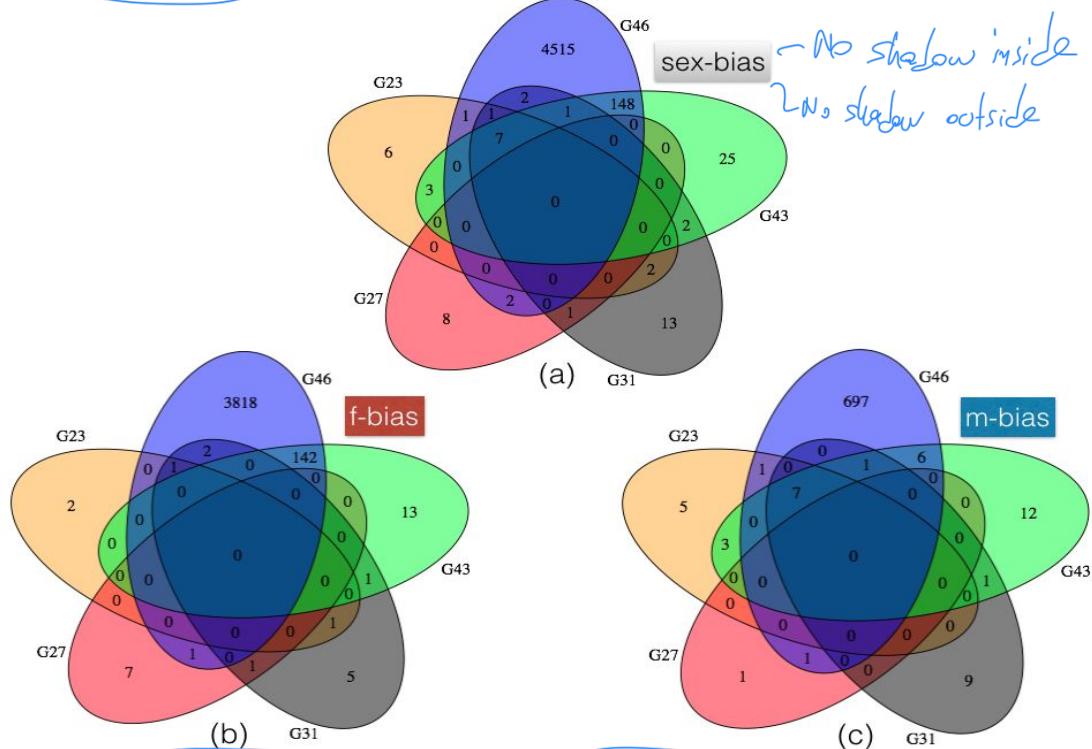


Figure 8. Shared sex-biased genes (a), shared female-biased genes (b) and shared male-biased genes (c) among three adult tissues, brain, gonad and liver.

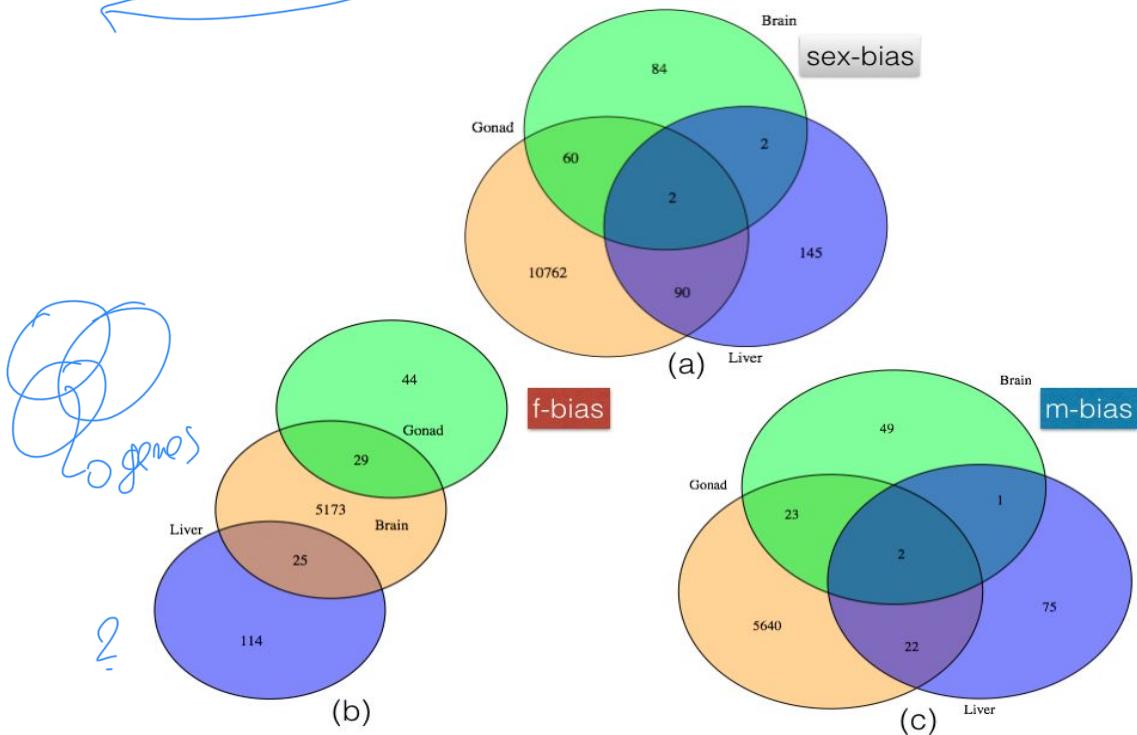


Figure 9. Shared sex-biased genes (a), shared female-biased genes (b) and shared male-biased genes (c) between G43 and gonad tissues which have high sex-biased genes.

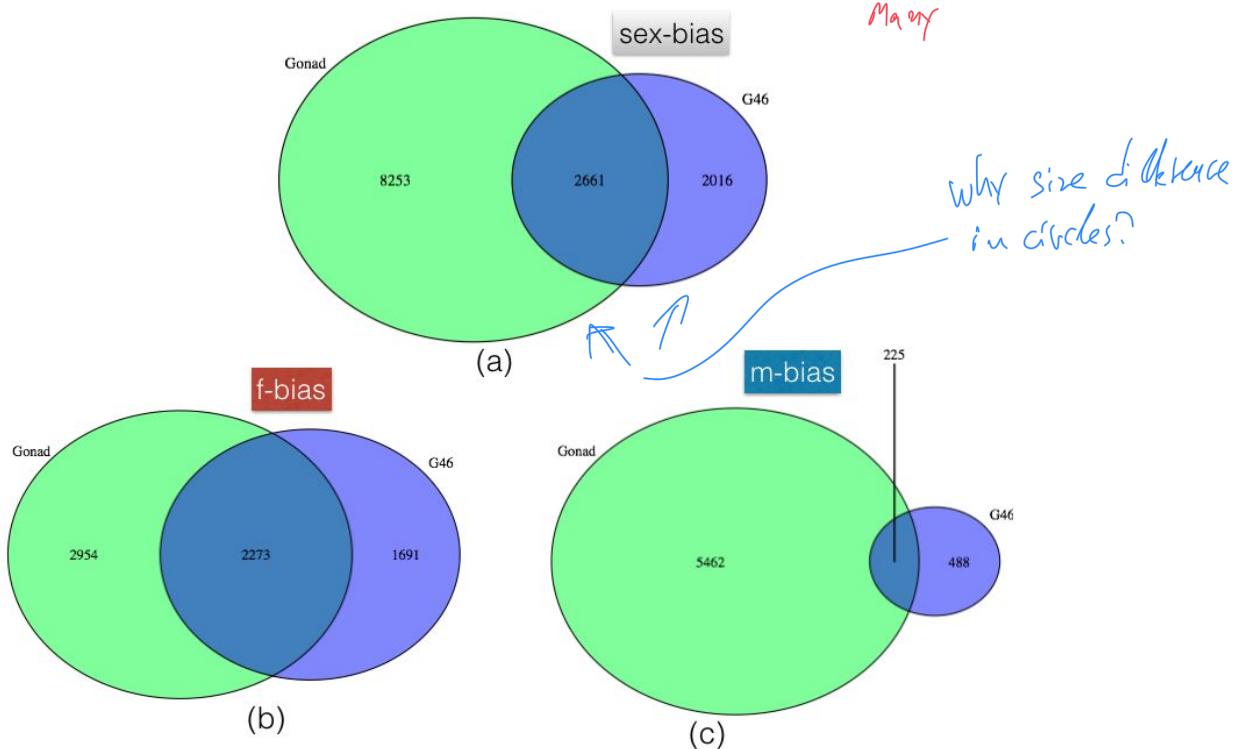
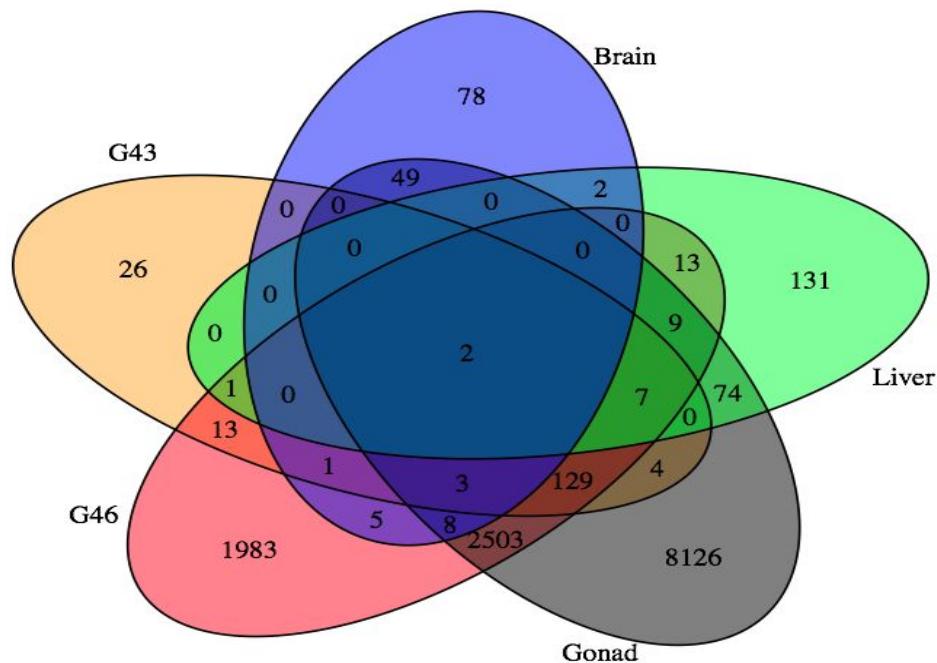


Figure 10. Shared sex-biased genes between tissues of G43, G46, and adult gonad, liver and brain tissues.

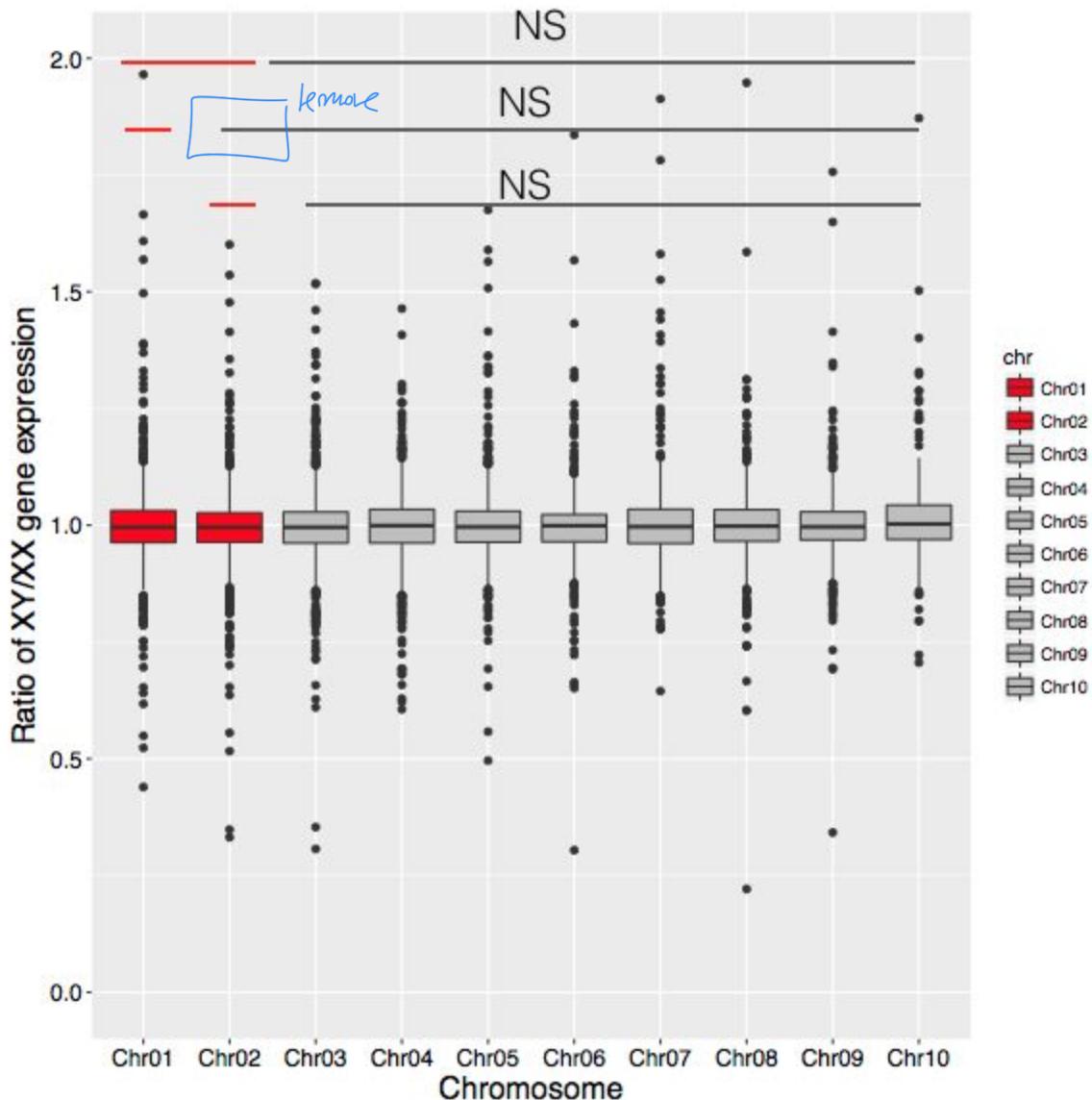


C. Transcription degeneration

To investigate possible transcriptional degeneration ~~at early sex chromosome evolution~~, we compare gene expression ratio of XY individuals with testis and XX individuals with testis, $\text{Log}_2(\text{XY}/\text{XX})$.

Figure 10. BOXPLOT OF Gene expression ratio between individuals of XY with testis and XX with testis. Note, chromosome 1 and 2 are both shown to be linked to sex determination.

both harbour male specific haplotypes



Report for $\text{abs}(\text{LogFC}) \rightarrow$ sexualisation of elutrosome

for $\text{LogFC} [\text{LogFC} > 0] \rightarrow \text{♂ biased only}$

for $\text{LogFC} [\text{LogFC} < 0] \rightarrow \text{♀ biased only}$

D. Coding sequence divergence and Faster-X

Approach: find the longest ORFs per transcript, use 1:1 ortholog with *X.tropicalis* to locate the genome locations for each transcript, finally use PRINK to perform codon alignment and finally calculate dN, dS, dN/dS with Codeml model in PAML.

Figure 11. dN/dS ratio of sex-biased and unbiased genes throughout development and three adult tissues. M = Males biased, F = female biased, U = not sex biased.

*** denotes $P < 0.0005$ 0.001

** denotes $P < 0.005$ 0.01

* denotes $P < 0.07$ 0.05

0.07

What is the statistical test?

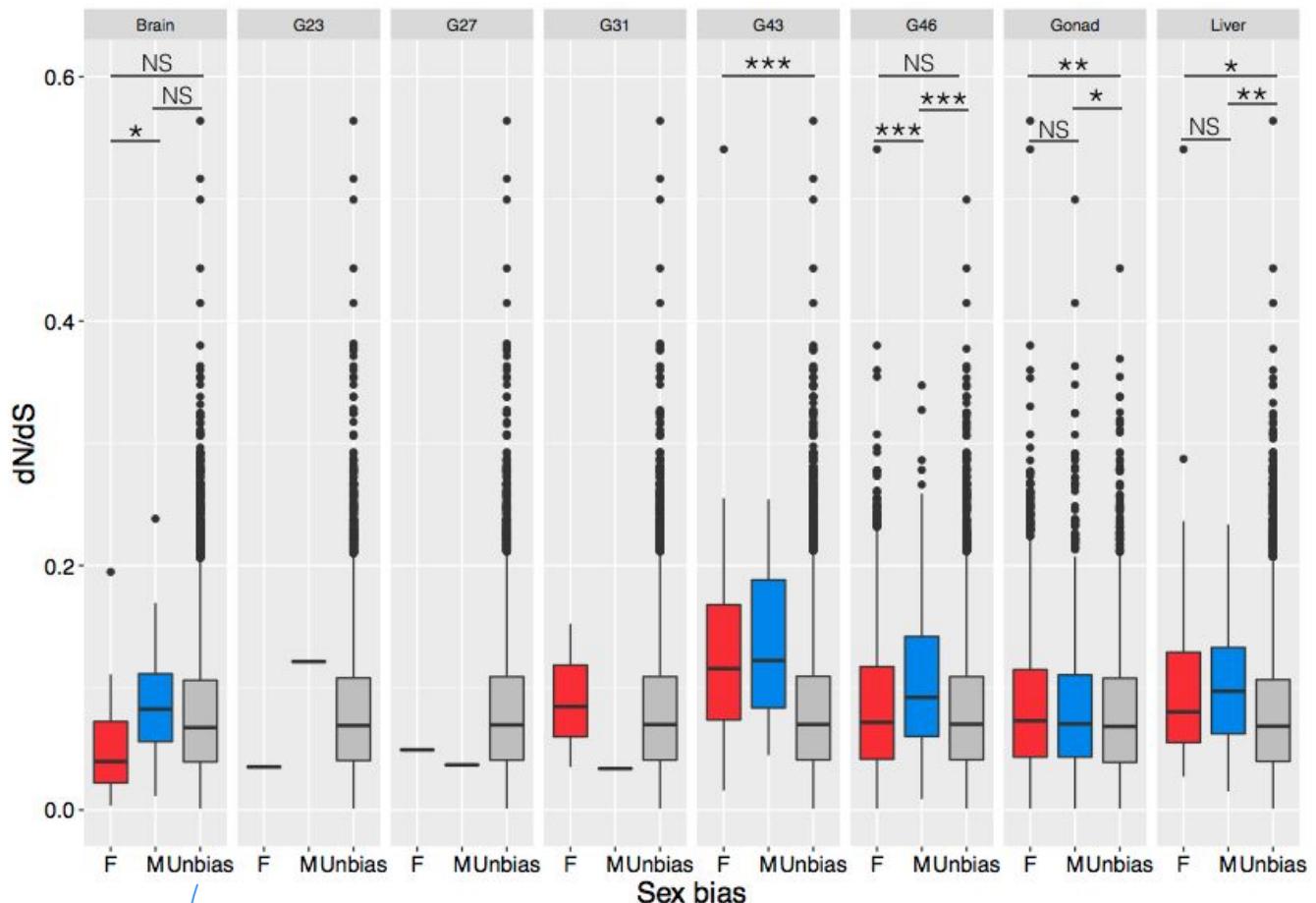


Figure 12. dN (a) and dS (b) values of sex-biased and unbiased genes throughout development and three adult tissues.

*** denotes $P < 0.0005$

** denotes $P < 0.005$

* denotes $P < 0.07$

Supplementary?

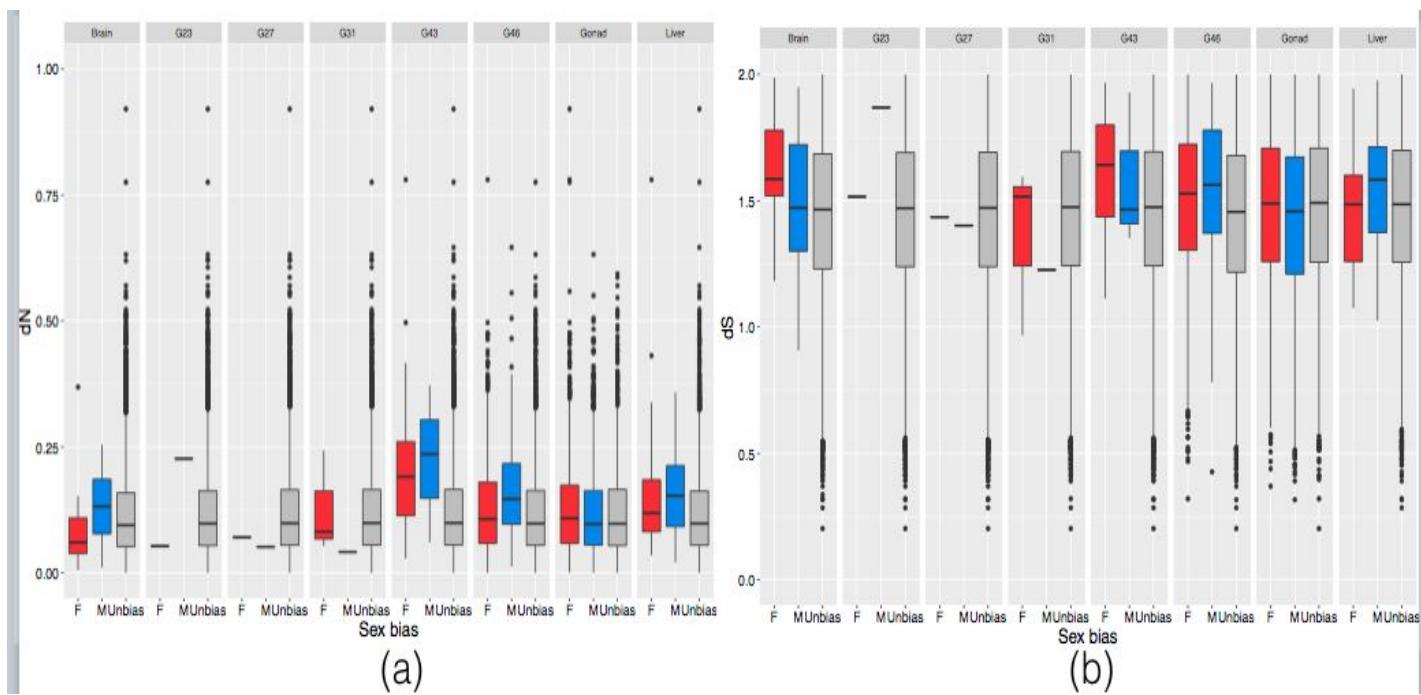


Figure 12. dN/dS ratio of genes on sex chromosomes (chromosome 1 and chromosome 2) and autosomes.

