# Essays on Bioinformatics and Social Network Analysis

## Statistical and Computational Methods for Complex Systems

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# What motivates my research

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**Statistical and computational methods for bioinformatics and social network analysis**

- We live in a non-*IID* world.
- In some times, the cannot understand a process unless we look at it as a whole.
- There's a reason why we usually assume *IID*.
- *Modern* (as of today) computational tools help us coping with that.

# Contents

## On the prediction of gene functions using phylogenetic trees

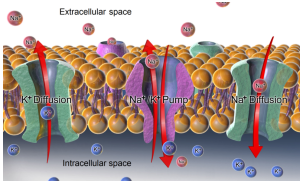
*Joint with*: Paul D Thomas, Paul Marjoram, Huaiyu Mi, Duncan Thomas, and John Morrison

# Genes

Encode the synthesis of genetic products that ultimately are related to a particular aspect of life, for example

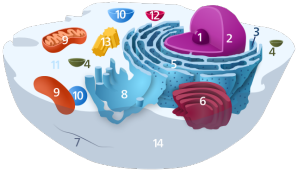
**Molecular function**

Active transport GO:0005215



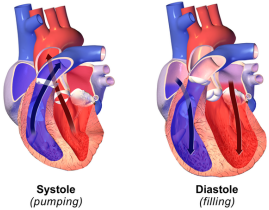

**Cellular component**

Mitochondria GO:0004016



**Biological process**

Heart contraction GO:0060047



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GENEONTOLOGY
Unifying Biology

- The GO project has $\sim$ 44,700 validated terms ▸ more, $\sim$ 7.3M annotations on $\sim$ 4,500 species.
- About $\sim$ 500,000 are on human genes.
- Roughly half of human genes ($\sim$ 10,000 / 20,000) have some form of annotation.
- We know something of less than 10% of known genes (near 1.7M).
- An important effort of the GO has to do with phylogenetics...

**source**: Statistics from `pantherdb.org` and `geneontology.org`

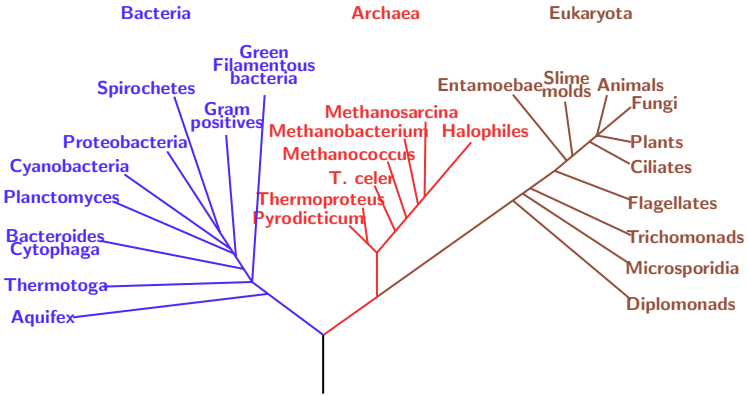



**Figure 1** A phylogenetic tree of living things, based on RNA data and proposed by Carl Woese, showing the separation of bacteria, archaea, and eukaryotes (wiki)

# Phylogenetic Trees: The PANTHER classification system

- The PANTHER project (part of GO) provides information about evolutionary structure of 1.7 million genes
- These genes are grouped in 15,524 phylogenetic trees (families)
- A single family can host multiple species

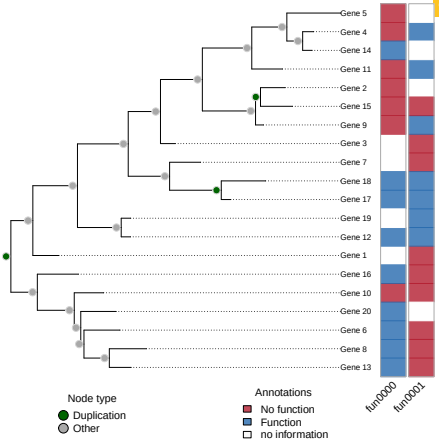



**Figure 2** Simulated phylogenetic tree and gene annotations.

We can use

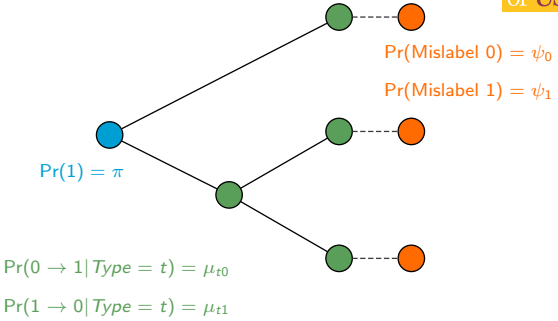
**evolutionary trees**

to inform a model for predicting

**genetic annotations!**

# An evolutionary model of gene functions

- Initial (spontaneous) gain of function.
- Loss/gain of offspring depends on: (a) the state of their parents (**Markov process**), and (b) the type of node more
- We control for human error.




We implemented the model using Felsenstein's' pruning algorithm (linear complexity) in the R package `aphylo`.

other models other view

# The `aphylo` R package

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- Simulation and visualization of annotated phylogenetic trees.
- Pruning algorithm implemented in C++ using the `pruner` template library (by-product).
- Uses meta-programming: users can specify different formulas, including pooled models more.
- The estimation is done using either Maximum Likelihood, Maximum A Posteriory, or MCMC.
- The MCMC estimation is done via the `fmcmc` R package using adaptive MCMC (also implemented as part of this project):
  - Automatic stop via convergence check.
  - Out-of-the-box parallel chains using parallel computing.
  - User-defined transition kernel (in our case, Adaptive Kernel).

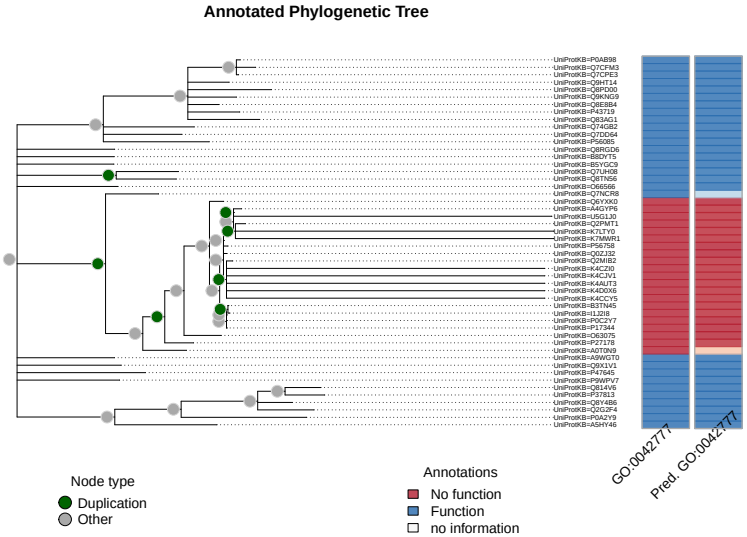
# Prediction with real data

| | Prior | |
|---|---|---|
| | Uniform | Beta |
| Mislab. prob. | | |
| $\psi_0$ | 0.23 | 0.25 |
| $\psi_1$ | 0.01 | 0.01 |
| **Gain/Loss at dupl.** | | |
| $\mu_{d0}$ | 0.97 | 0.96 |
| $\mu_{d1}$ | 0.52 | 0.58 |
| **Gain/Loss at spec.** | | |
| $\mu_{s0}$ | 0.05 | 0.06 |
| $\mu_{s1}$ | 0.01 | 0.02 |
| Root node | | |
| $\pi$ | 0.81 | 0.45 |
| Leave-one-out AUC | | |
| Mean | 0.69 | 0.67 |
| Median | 0.81 | 0.75 |

**Table 1** Parameter estimates using different priors.

- 141 pooled functions (trees) with 7,388 genes with 0/1 annotations.
- Parameter estimates are actually probabilities.
- Data driven results (uninformative prior).
- Biologically meaningful results.
- Took about 5 minutes each.

# Prediction with real data: Leave-one-out



# Prediction with real data: Out-of-sample prediction

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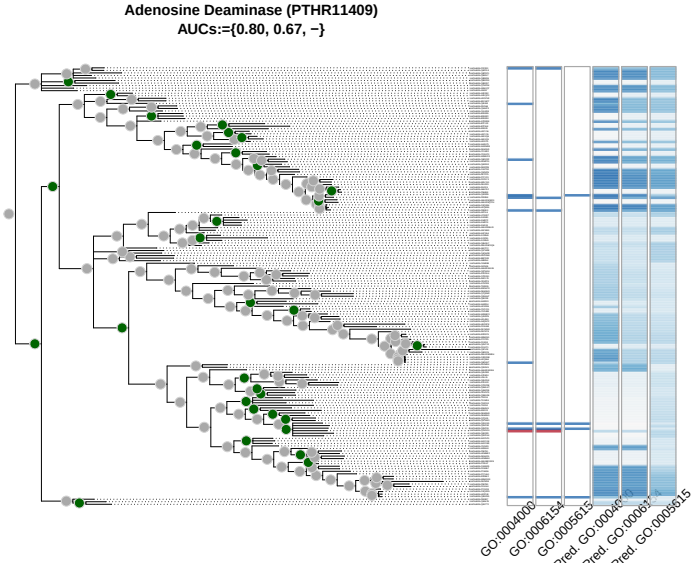

# Paper 1: On the prediction of gene functions using phylogenetic trees

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## Key takeaways

- A parsimonious model for predicting gene functions using phylogenetics.
- Computationally scalable. SIFTER (our benchmark) would take about 66 years (yes, years) to estimate a model for 100 families of size 300, we take about 5 minutes.
- Meaningful biological results.
- Preliminary accuracy results comparable to state-of-the-art phylo-based models.

## Challenges

- Offspring are conditional independent on their parent and
- Functions evolve independently. more

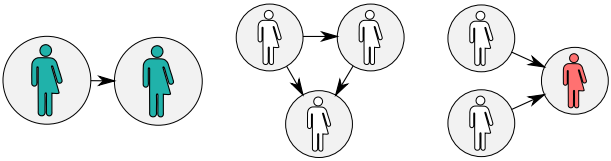
## Exponential Random Graph Models for Small Networks

*Joint with*: Andrew Slaughter and Kayla de la Haye

# What are Exponential Random Graph Models

Exponential Family Random Graph Models, aka ERGMs are:

- Statistical models of (social) networks
- In simple terms: statistical inference on what network patterns/structures/motifs govern social networks



# ERGMs (cont'd)

A vector of model parameters

A vector of sufficient statistics

$$\Pr\left(\mathbf{Y} = \mathbf{y} \mid \theta, \mathbf{X}\right) = \frac{\exp\left\{\theta^{\mathbf{t}} s\left(\mathbf{y}, \mathbf{X}\right)\right\}}{\sum_{\mathbf{y}' \in \mathcal{Y}} \exp\left\{\theta^{\mathbf{t}} s\left(\mathbf{y}', \mathbf{X}\right)\right\}}, \quad \forall \mathbf{y} \in \mathcal{Y}$$

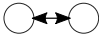
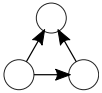
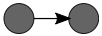
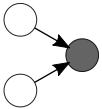
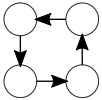
Observed data

The normalizing constant

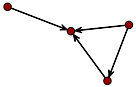
All possible networks

The normalizing constant has $2^{n(n-1)}$ terms!

## **Sufficient statistics** have various forms

| Representation | Description |
|---|---|
|  | Mutual Ties (Reciprocity)<br>$\sum_{i \neq j} y_{ij} y_{ji}$ |
|  | Transitive Triad (Balance)<br>$\sum_{i \neq j \neq k} y_{ij} y_{jk} y_{ik}$ |
|  | Homophily<br>$\sum_{i \neq j} y_{ij} \mathbf{1}(x_i = x_j)$ |
|  | Covariate Effect for Incoming Ties<br>$\sum_{i \neq j} y_{ij} x_j$ |
|  | Four Cycle<br>$\sum_{i \neq j \neq k \neq l} y_{ij} y_{jk} y_{kl} y_{li}$ |

In this network



We see 4 **edges**, 1 **transitive triad** and **no mutual ties**.

The probability function of this model would be

$$\mathbb{P}(\mathbf{G} = \mathbf{g} \mid \theta) = \frac{\exp\{4\theta_{edges} + \theta_{ttriads} + 0\theta_{mutual}\}}{\sum_{\mathbf{g}' \in \mathcal{G}} \exp\{\theta^{\mathbf{t}} s(\mathbf{g}')\}}$$

with $\theta = [\theta_{edges} \quad \theta_{ttriads} \quad \theta_{mutual}]^{\mathbf{t}}$

This model has **MLE parameter estimates** of -0.20 (low density), 0.28 (high chance of ttriads), and -Inf (low chance of mutuality) for the parameters `edges`, `ttriads`, and `mutual` respectively.

# ERGMs: State of the Art

Medium-large (dozens to a couple of thousand vertices) networks

- Markov Chain Monte Carlo (MCMC) based approaches like MC-MLE or Robbins-Monro Stochastic Approximation. details
- Maximum Pseudo Likelihood (MPLE)

large-huge networks (up to millions of vertices)

- Parametric bootstrap
- Conditional joint estimation (like snowball sampling, a.k.a. divide and conquer)
- Equilibrium Expectation Algorithm (millions of vertices)

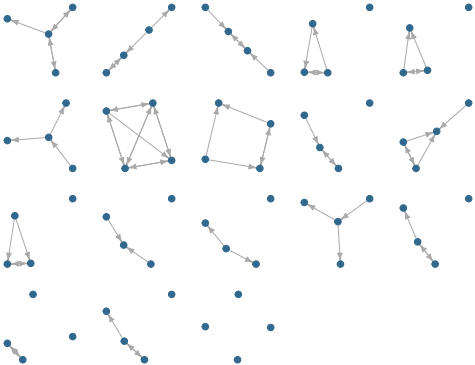
All of these methods are approximations!

# Do we care about small networks?

We see small networks everywhere

- Families and friends
- Small teams
- Egocentric networks
- Online networks (sometimes)
- etc.

Small networks come in samples



# ERGMs for Small Networks

A vector of model parameters

A vector of sufficient statistics

$$\Pr\left(\mathbf{Y} = \mathbf{y} \mid \theta, \mathbf{X}\right) = \frac{\exp\left\{\theta^{\mathbf{t}} \boldsymbol{s}\left(\mathbf{y}, \mathbf{X}\right)\right\}}{\sum_{\mathbf{y}' \in \mathcal{Y}} \exp\left\{\theta^{\mathbf{t}} \boldsymbol{s}\left(\mathbf{y}', \mathbf{X}\right)\right\}}, \quad \forall \mathbf{y} \in \mathcal{Y}$$

Observed data

The normalizing constant

All possible networks

- In the case of small-enough networks, computation of the likelihood becomes computationally feasible.
- This allow us to directly compute **the normalizing constant**.
- Using the exact likelihood opens a huge window of methodological-possibilities.
- We implemented this and more in the `ergmito` R package

Sidetrack...

**ito, ita**: From the latin -*ī̄ttus*. suffix in Spanish used to denote small or affection. e.g.:

*¡Qué lindo ese perr**ito**! / What a beautiful little dog!*

*¿Me darías una tac**ita** de azúcar? / Would you give me a small cup of sugar?*

Special thanks to George Barnett who proposed the name during the 2018 NASN!

# The ergmito

In general

- Implements estimation of ERGMs using exact statistics for small networks.
- Meta-programming allows specifying likelihood (and gradient) functions for pooled models.
- Includes tools for simulating and post-estimation checks.
- Getting ready for CRAN!

Other features

- Vectorized calculation of sufficient statistics.
- Scales up nicely (hundreds of small networks) saving space and computation (when possible).
- Highly tested (90% coverage with more than one hundred tests).

# ergmito example

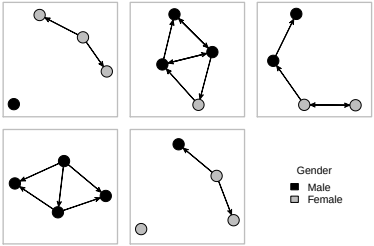



**Figure 3** Random sample of 5 networks simulated using the ergmito package

| | Bernoulli | Full model |
|---|---|---|
| Edge-count | $-0.69^{*}$ | $-1.70^{**}$ |
| | (0.27) | (0.54) |
| Homophily (on Gender) | | $1.59^{*}$ |
| | | (0.64) |
| AIC | 78.38 | 73.34 |
| BIC | 80.48 | 77.53 |
| Log Likelihood | -38.19 | -34.67 |
| Num. networks | 5 | 5 |

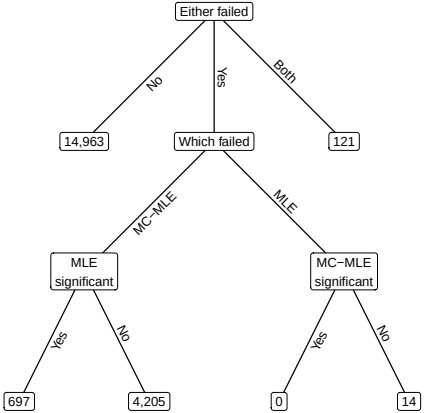
Standard errors in parenthesis. $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$

**Table 2** Fitted ERGMitos using the fivenets dataset.

We performed a large simulation study more comparing MC-MLE (ergm) with MLE (ergmito).

# Paper 2 Simulation Studies: Error rate

- The MC-MLE implementation failed $\sim$ 5,000/20,000 times
- In $\sim$700 of those cases ergmito (MLE) reported a significant effect
- I no case that MLE failed MC-MLE reported an effect.



# Paper 2 Simulation Studies: Empirical Type I error

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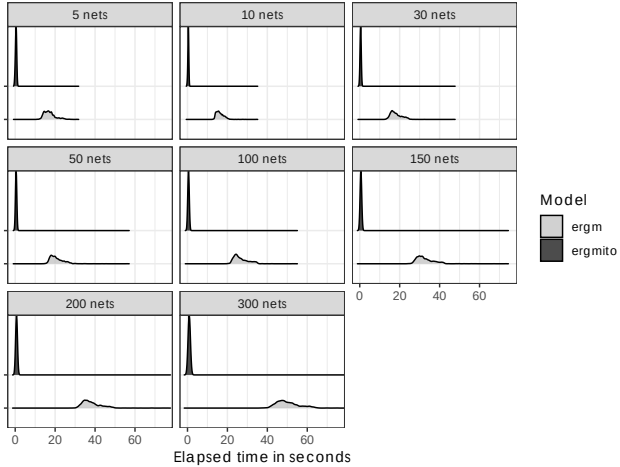
| Sample size | P(Type I error) | | $\chi^2$ |
|---|---|---|---|
| | MC-MLE (`ergm`) | MLE (`ergmito`) | |
| 5 | 0.084 | 0.057 | 11.71 *** |
| 10 | 0.070 | 0.045 | 12.46 *** |
| 15 | 0.084 | 0.066 | 5.55 * |
| 20 | 0.074 | 0.060 | 3.58 |
| 30 | 0.057 | 0.052 | 0.67 |
| 50 | 0.046 | 0.044 | 0.17 |
| 100 | 0.048 | 0.048 | 0.00 |

**Table 3** Empirical Type I error rates. The $\chi^2$ statistic is from a 2-sample test for equality of proportions, and the significance levels are given by *** $p < 0.001$, ** $p < 0.01$, and * $p < 0.05$.

# Paper 2 Simulation Studies: Elapsed time




more results

# Paper 2: Exponential Random Graph Models for Small Networks

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## Key takeaways

- New extension of ERGMs using exact statistics for small networks (families, teams, etc.)
- Performance: Same (un)bias, Lower Type I error rates, (way) faster.
- Opens the door the new methods, e.g. Mixed effects, LRT, etc.

## Challenges

- Computationally, we can do better in terms of speed/memory.
- Have a good way of assessing goodness-of-fit.
- Explore extending this method for (very) large networks.

# Future Research

# Future Research: phylogenetic models

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- Make the model hierarchical when pooling trees
  - Different mutation rates per class of tree/function
  - Can be complicated to fit/justify (how many classes?)
- Use a framework similar to Exponential Random Graph Models:

$$\mathbb{P}\left(\mathbf{X} = \{x_{n1}, x_{n2}, \ldots\} \;\middle|\; x_{\mathbf{p}(n1,\ldots)}\right) = \frac{\exp\left\{\mu^T s(\mathbf{x}|x_{\mathbf{p}(\cdot)})\right\}}{\sum_{\mathbf{x}'} \exp\left\{\mu^T s(\mathbf{x}'|x_{\mathbf{p}(\cdot)})\right\}}$$

  - A generalization of the model.
  - Extends to account for joint dist of functions+siblings.
  - Can incorporate additional information such as branch lengths.
  - Yet computationally more compact compared to SIFTER (finite number of parameters).

# Future Research: phylogenetic models

Imagine that we have 3 functions (rows) and that each node has 2 siblings (columns)

| | | | **Transitions to** | |
|---|---|---|---|---|
| | | | Case 1 | Case 2 |
| **Parent** | A | $\begin{bmatrix} 0 \end{bmatrix}$ | $\begin{bmatrix} 0 & 0 \\ 1 & 0 \\ 0 & 1 \end{bmatrix}$ | $\begin{bmatrix} 1 & 0 \\ 0 & 0 \\ 1 & 0 \end{bmatrix}$ |
| | B | $\begin{bmatrix} 1 \end{bmatrix}$ | | |
| | C | $\begin{bmatrix} 0 \end{bmatrix}$ | | |
| **Sufficient statistics** | | | | |
| # Gains | | | 1 | 2 |
| # only one offspring changes | | | 1 | 0 |
| # Swaps (0→1, 1→0) | | | 2 | 4 |

In SIFTER, for modelling 3 functions, we need $2^{2\times 3} = 64$ parameters.

# Future Research: ERGMitos

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**Goodness-of-fit**

- Is something that will need to be addressed at some point.
- The problem is not easy as we need to deal a discrete distribution.
- Two key questions: What sufficient statistic to look at? what test?

**ERGMs for large networks**

- There is still no standard way to estimate ERGMs for large networks.
- Most attempts are still depending on simulation methods.
- We could use the Snowball Sampling framework together with ERGMitos. (... I would call this ERGMote)

# Concluding Remarks

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- Paper 1: Phylogenetic models of gene functional evolution
  - Parsimonious and biologically meaningful.
  - Computationally scalable.
  - Performance comparable to state-of-the-art alternatives.
  - **Next steps:** Use ERGMs framework to break assumptions.
- Paper 2: ERGMs for small networks
  - An extension to a well studied models for social networks.
  - Small size allows exact calculations.
  - Opens the door to a large set of methodological innovations.
  - **Next steps:** GOF or extensions to large networks?

**Accomplishments during the development of this work**

- 6 journal publications (Journal of Open Source Software, Stata Journal, Journal of health and social behavior, Translational behavioral medicine, Social Science & Medicine)
- 11 packages/libraries built (ergmito, similR, gnet, fmcmc, slurmR, aphylo, polygons, pruner, netplot, rphyloxml, jsPhyloSVG)

# Essays on Bioinformatics and Social Network Analysis

## Statistical and Computational Methods for Complex Systems

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November 18, 2019

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# Thanks!

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Yu, Chun et al. (Jan. 2018). "Assessing the Performances of Protein Function Prediction Algorithms from the Perspectives of Identification Accuracy and False Discovery Rate". In: International Journal of Molecular Sciences 19.1, p. 183. ISSN: 1422-0067. DOI: 10.3390/ijms19010183. URL: http://www.mdpi.com/1422-0067/19/1/183.

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Example of GO term

| | |
|---|---|
| **Accession** | GO:0060047 |
| **Name** | heart contraction |
| **Ontology** | biological_process |
| **Synonyms** | heart beating, cardiac contraction, hemolymph circulation |
| **Alternate** | IDs None |
| **Definition** | The multicellular organismal process in which the heart decreases in volume in a characteristic way to propel blood through the body. Source: GOC:dph |

**Table 4** Heart Contraction Function. source: amigo.geneontology.org

You know what is interesting about this function?

go back

These four species have a gene with that function... and two of these are part of the same evolutionary tree!

Felis catus pthr10037

Oryzias latipes **pthr11521**

# Predicting gene functions

There various approaches for this, some to highlight

- Text analysis like in Pesaranghader et al. 2016
- Protein-protein interaction networks like in Oliver 2000; Piovesan et al. 2015.
- Phylogenetic based like SIFTER Barbara E. Engelhardt et al. 2011, 2005.
  - Parameters to estimate: $2^{2P}$, where $P$ is the number of functions.

(a nice literature review in Jiang et al. 2016; Yu et al. 2018) go back

# An evolutionary model of gene functions (algorithmic view)

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**Data:** A phylogenetic tree, $\{\pi, \mu, \psi\}$(Model probabilities)

**Result:** An annotated tree

```
for n ∈ PostOrder(N) do
    Nodes gain/loss function depending on their parent;
    switch class of n do
        case root node do
            Gain function with probability π;
        case interior node do
            if Parent has the function then Keep it with prob. (1 − μ1);
            else Gain it with prob. μ0;
    end
    Finally, we allow for mislabeling;
    if n is leaf then
        if has the function then Mislabel with prob. ψ1;
        else Mislabel with prob. ψ0;
end
```

go back

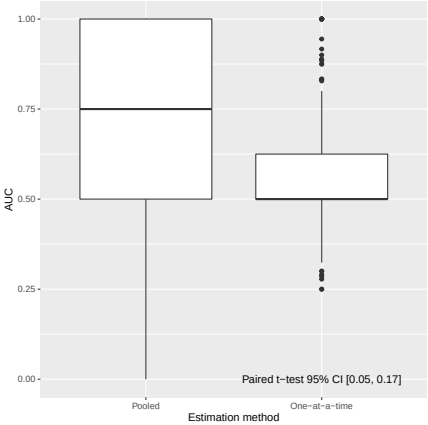



**Figure 4** Comparing LOOCV AUC when performing predictions using either the estimates from the pooled model or each trees' own set of estimates obtained when fitting the model individually go back.

# Speciation

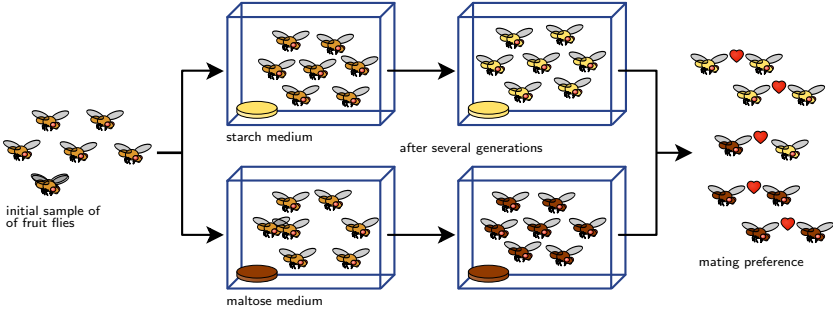



**Figure 5** Dodd 1989: After one year of isolation, flies showed a significant level or assortativity in mating (wikimedia)

go back

# Duplication

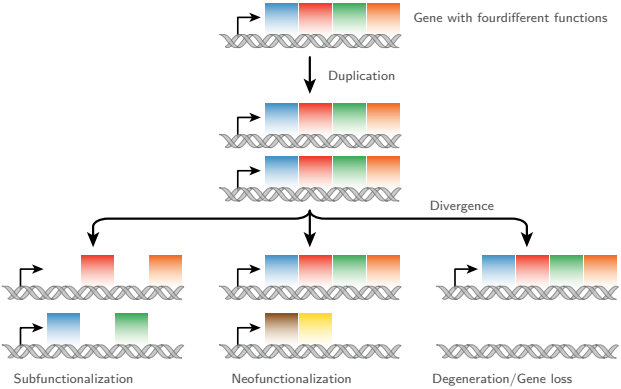



**Figure 6** A key part of molecular innovation, gene duplication provides opportunity for new functions to emerge (wikimedia)

go back

# ERGMs: The MC-MLE approach

One of the most popular methods for estimating ERGMs is the MC-MLE approach (citations here)

This consists on the following steps

1. Start from a sensible guess on what should be the population parameters (usually done using pseudo-MLE estimation)
2. While the algorithm doesn't converge, do:
   - 2.1 Simulate a stream of networks with the current state of the parameter, $\theta_t$
   - 2.2 Using the law of large numbers, approximate the ratio of likelihoods based on the parameter $\theta_t$, this is the objective function
   - 2.3 Update the parameter by a Newton-Raphson step
   - 2.4 Next iteration

go back

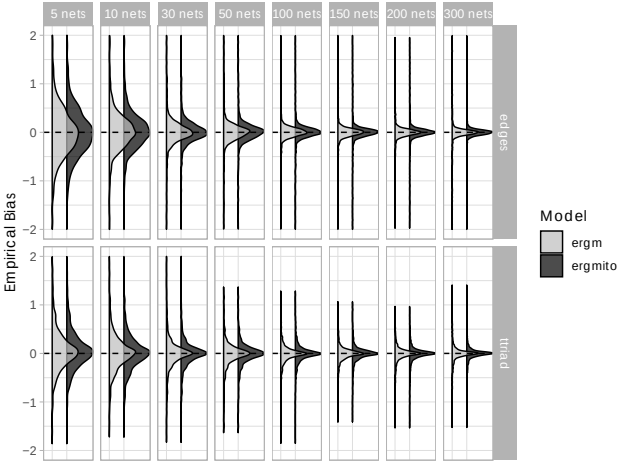
# Paper 2 Simulation Studies

We performed a simulation study with the following features:

- Draw 20,000 samples of groups of small networks
- Each group had prescribed: (model parameters, number of networks, sizes of the networks)
- Each group could have from 5 to 300 small networks
- We estimated the models using MC-MLE and MLE.

go back

# Paper 2 Simulation Studies: Empirical Bias




go back