



# Networks Dynamics

## Gene Networks @ Work using Digital Organisms

Reverter & Dalrymple, 2005  
BioInfoSummer, ANU, Canberra



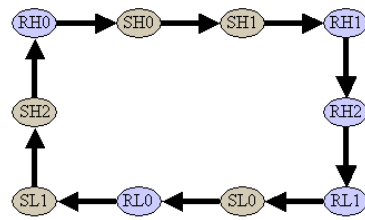
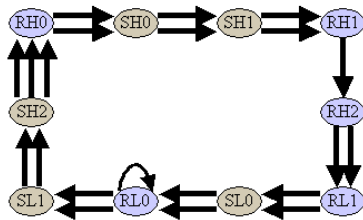
### Network Dynamics

#### Setting the Scene

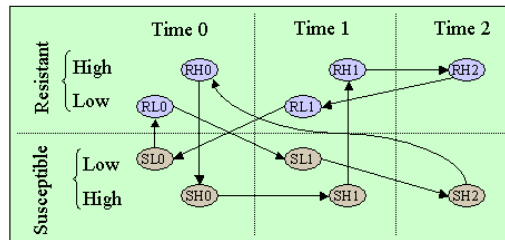
#### Fleece Rot Resistance

Trangie

Armidale



**LEGEND:** Arrows indicate hybridisations and go from the sample labelled with red to the sample labelled with green dye. Clicking of the body of the arrow pops up the 2-page pdf file with summary info for this hybridisation. Clicking of the head pops up the entire raw data. The ppt with full design details is [HERE](#)







### Network Dynamics

#### Setting the Scene

#### Fleece Rot Resistance

	A	B	C	D
1	Clone ID	Analysis	Pattern in Full	Pattern in High
2				
3	CCL007335	Full&High	D_D_F	D_D_F
4	CCL005524	Full&High	D_D_F	D_D_F
5	2_11_H07	Full&High	D_D_F	D_D_F
6	2_07_G03	Full&High	D_D_F	D_F_F
7	2_09_F10	Full&High	D_D_F	F_D_F
8	2_01_B08	Full&High	D_D_F	F_D_F
9	1_10_E01	Full&High	D_D_F	F_D_F
10	2_06_H05	Full&High	D_D_F	F_D_F
11	2_06_G09	Full&High	D_D_F	F_D_F
12	6_03_D05	Full&High	D_D_F	F_D_F
13	2_11_B10	Full&High	D_D_F	F_D_F
14	2_06_G04	Full&High	D_D_F	F_D_F
15	ad-083_24-07-97	Full&High	D_D_F	F_D_F
16	2_06_D01	Full&High	D_D_F	F_D_F
17	1_11_D07	Full&High	D_D_F	F_D_F
18	2_05_H10	Full&High	D_D_F	F_D_F
19	CCL005588	Full&High	D_F_F	D_F_F
20	2_08_F07	Full&High	D_F_F	D_F_F
21	2_02_C01	Full&High	D_F_F	D_F_F
22	CCL005523	Full&High	D_F_F	D_F_F
23	2_11_A08	Full&High	D_F_F	D_F_F
24	CCL002966	Full&High	D_F_F	D_F_F
25	1_03_D05	Full&High	D_F_F	D_F_F
26	2_05_G02	Full&High	D_F_F	F_D_F

297 DE Clones



102 DE Genes



### Network Dynamics

#### Setting the Scene

**Targeting c-Myc-activated genes with a correlation method: Detection of global changes in large gene expression network dynamics**

D. Remondini<sup>1\*</sup>, B. O'Connell<sup>1</sup>, N. Intrator<sup>2</sup>, J. M. Sedivy<sup>3</sup>, N. Morotti<sup>4</sup>, G. C. Castellani<sup>1,5,6,7,8,9</sup>, and L. N. Cooper<sup>1,10,11,12</sup>

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Contributed by L. N. Cooper, March 1

**Significance Analysis and Data Preprocessing.** A two-way full factorial ANOVA was applied to each of the 8,799 probe sets to identify those that significantly changed expression level in time between the two conditions (data set  $N$  versus data set  $T$ ). The significance analysis was based on the general linear model that describes changes in gene expression level  $y$  from the global mean  $\mu$  as caused by the combination of: changes in treatment ( $\beta$ ), i.e., database  $N$  versus database  $T$ ; changes in time ( $\alpha$ ); interaction between time and treatment ( $\gamma$ ); plus some random effects ( $\epsilon$ ):

$$y_{ijk} = \mu + \alpha_j + \beta_i + \gamma_{ij} + \epsilon_{ijk} \quad [1]$$

where the index  $i$  refers to the data set ( $N$  or  $T$ ), the index  $j$  refers to time ( $j = 1, 2, 4, 8, \text{ or } 16 \text{ h}$ ), and the index  $k$  refers to the replication of the experiment for a fixed condition and time ( $k = \text{experiment } 1, 2, \text{ or } 3$ ). Probe sets with a  $P$  value corresponding for convenience). Because the number of genes is larger than the number of time points, Eq. 3 does not have a unique solution. A common approach is to solve it by using the Moore-Penrose generalized matrix inverse  $X_T^+$  of  $X_T$  (a unique pseudoinverse matrix obtained by including additional constraints) via its singular value decomposition (4) such that:

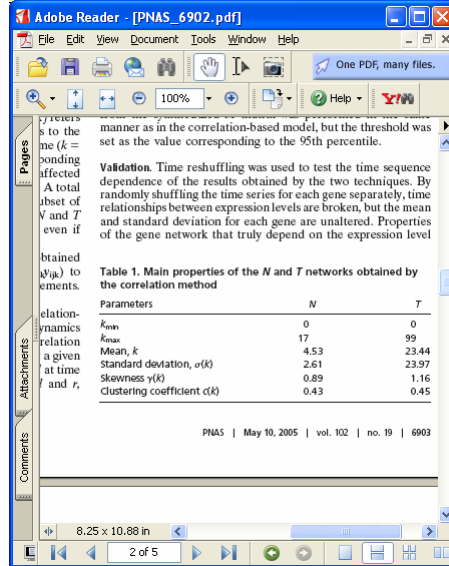
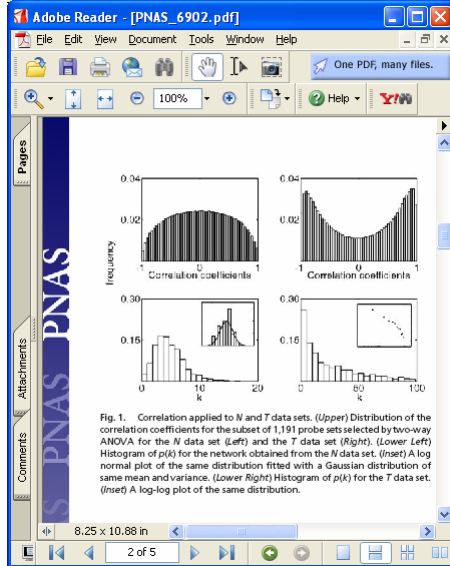
$$M = X_{t+1} X_T^+$$

Because the resulting matrix  $M$  is in general not symmetric, we applied a symmetrization procedure by averaging the corresponding off-diagonal coefficients (other symmetrization techniques lead to similar results in terms of network properties). Computation of the adjacency matrix and gene connectivity from the symmetrized  $M$  matrix was performed in the same manner as in the correlation-based model, but the threshold was set as the value corresponding to the 95th percentile.



Network Dynamics

Setting the Scene

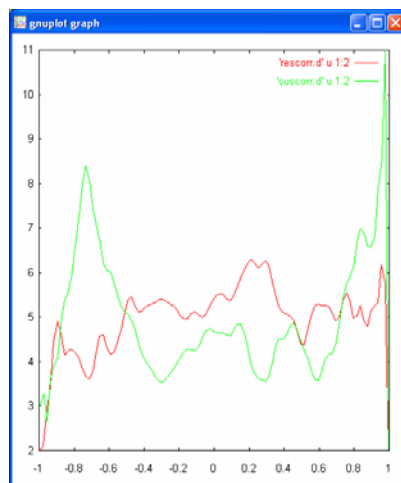


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Network Dynamics

Setting the Scene



	RES	SUS
Mean	34.4	44.2
Std	11.9	18.2
Min.	15	11
Max.	53	70
Clust.	0.34	0.44
Tot.Conn	1,755	2,255
Corr >  .90	439	600

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### Network Dynamics

#### Setting the Scene

#### Offending Correlations

Gene1	Gene2	Corr in RES	Corr in SUS
CDH1	ODC1	0.758	-0.960
CDH1	ATP5B	0.606	-0.867
CDH1	LOC521081	0.890	-0.791
CDH1	LOC534721	-0.840	0.687
CDH1	c4833423E24	0.917	-0.706
KRT1	ODC1	0.878	-0.768
KRT5	ABCC11	-0.867	0.885
KRT5	LOC506790	0.796	-0.835
ODC1	FBLN1	0.909	-0.585
TCP1	LOC511432	0.723	-0.788
FABP4	LOC534721	0.678	-0.809
FBLN1	ABCC11	-0.911	0.620
FBLN1	OA-MIT	0.864	-0.760
FBLN1	LOC506790	0.955	-0.734
FBLN1	LOC514360	-0.990	0.504
FBLN1	LOC515656	-0.696	0.827
HSPA5	SLC25A5	-0.969	0.622
ITM2B	UREB1	-0.649	0.915

#### Offending Connections

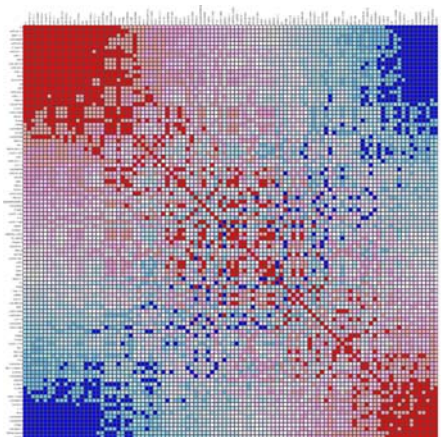
Gene	Conn in RES	Conn in SUS	Diff	Fold
RPL26	15	67	-52	-4.47
LOC511432	16	58	-42	-3.63
KLK10	19	64	-45	-3.37
FABP4	18	60	-42	-3.33
PCDH15	17	54	-37	-3.18
TXNDC	23	66	-43	-2.87
ABCC11	22	63	-41	-2.86
LOC480182	22	59	-37	-2.68
COL3A1	19	47	-28	-2.47
MEIS4	26	63	-37	-2.42
LOC515352	23	53	-30	-2.30
LOC534301	23	53	-30	-2.30
KRT5	20	46	-26	-2.30
KRTAP16.6	21	47	-26	-2.24
LOC507421	21	47	-26	-2.24
ITM2B	22	49	-27	-2.23
LOC506790	28	60	-32	-2.14
LOC534721	30	63	-33	-2.10
CDH1	23	46	-23	-2.00
COL1A2	20	40	-20	-2.00



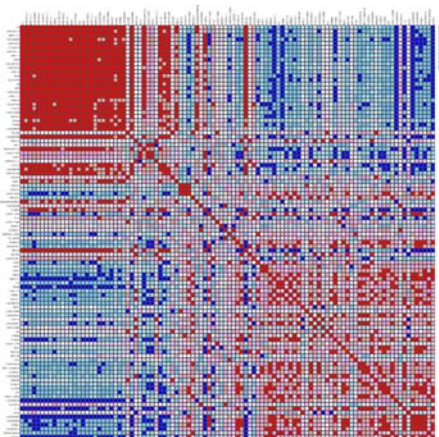
### Network Dynamics

#### Setting the Scene

#### Resistant



#### Susceptible



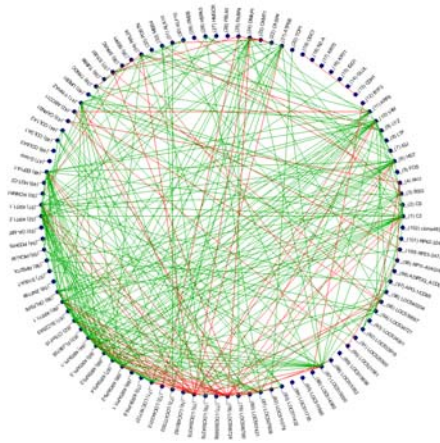
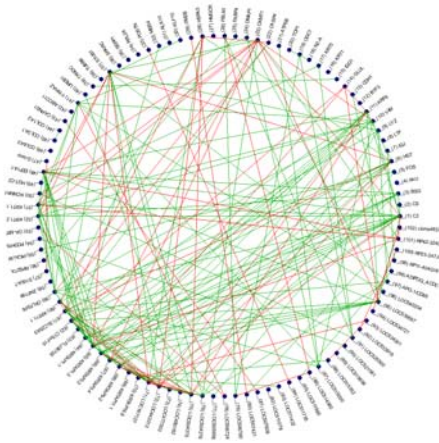


Network Dynamics

Setting the Scene

Resistant

Susceptible



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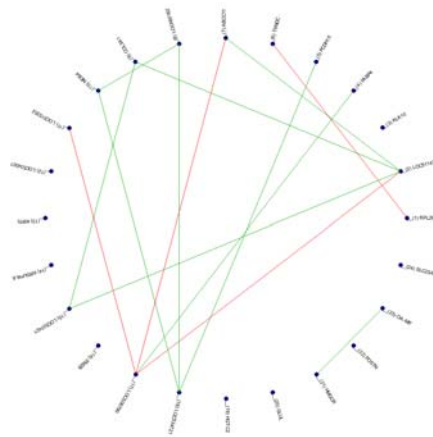
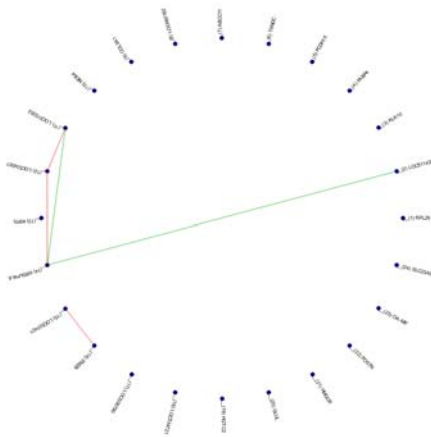
Network Dynamics

Setting the Scene

Resistant

Susceptible

24 Most Offending



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## Network Dynamics

### Introduction

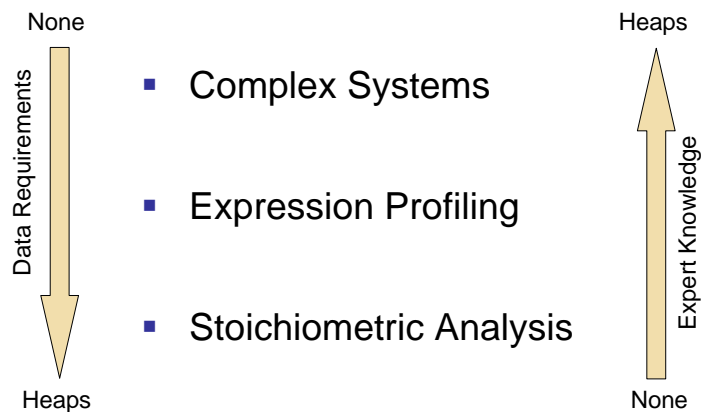
- Building a gene network is a challenge.
- Understanding how the essential genes function within a network is an even bigger challenge.
- Evidence of changes in network topology due to a number of factors.
- Given a network, postulating a hypothesis could be tricky (Type III Error).
- Biologically testing a network could be impossible.



## Network Dynamics

### Introduction

### APPROACHES



**Network Dynamics****Digital Organisms**

Digital Organisms are an elegant way to decipher the gradual evolutionary process found in complex structures that retain features related to earlier ancestral evolutionary steps.

Digital organisms have been shown to provide an increased understanding of fundamental problems including:

1. Why complex organisms have more robust fitness than simple ones (Lenski *et al.*, 1999);
2. The relative contributions of replication and mutation rates to survival (Wilke *et al.*, 2001 → “*Survival of the Flattest*”);
3. The evolutionary effect of productivity on species richness (Chow *et al.*, 2004).

Digital organisms can be understood as model systems to study the kinds of gene interactions that may result in phenotypic variation.

It is anticipated that this knowledge will, in turn, improve our ability to understand common diseases (Moore and Williams, 2005).

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**Network Dynamics****Digital Organisms****PARAMETERS**

- Genetic Load:

$$GL = \sum_{i=1}^{N_G} \mathbf{m}_i$$

- Phenotype:

$$P = G + E = \sum_{i=1}^{N_G} \mathbf{g}_i + E$$

$$\mathbf{g}_i = \mathbf{m}_i + \sum_{j=1}^{n_i} \mathbf{r}_{ij} \times \mathbf{m}_j$$

$$P = \sum_{i,j} \mathbf{r}_{ij} \times \mathbf{m}_j + E$$

- Descendants:

$$\mathbf{m}_i^n = 0.5 (\mathbf{m}_i^s + \mathbf{m}_i^t) + \sqrt{0.5\sigma^2} \Phi$$

- Connections:

$$\mathbf{r}_{ij}^n = 0.5 (\mathbf{r}_{ij}^s + \mathbf{r}_{ij}^t)$$

$$+ \sqrt{0.5\sigma_r^2} \Phi (1 - |0.5 (\mathbf{r}_{ij}^s + \mathbf{r}_{ij}^t)|)$$

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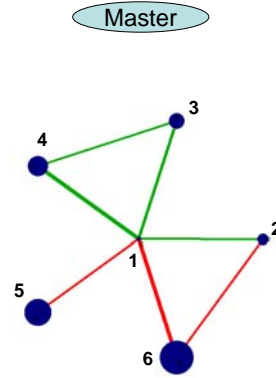
### Network Dynamics

#### Digital Organisms

Normalised Mean Expressions

Correlations

	NME	G1	G2	G3	G4	G5	G6
G1	2	1	.5	.7	.9	-.5	-.9
G2	5	.5	1	0	0	0	-.5
G3	7	.7	0	1	.5	0	0
G4	9	.9	0	.5	1	0	0
G5	12	-.5	0	0	0	1	0
G6	15	-.9	-.5	0	0	0	1

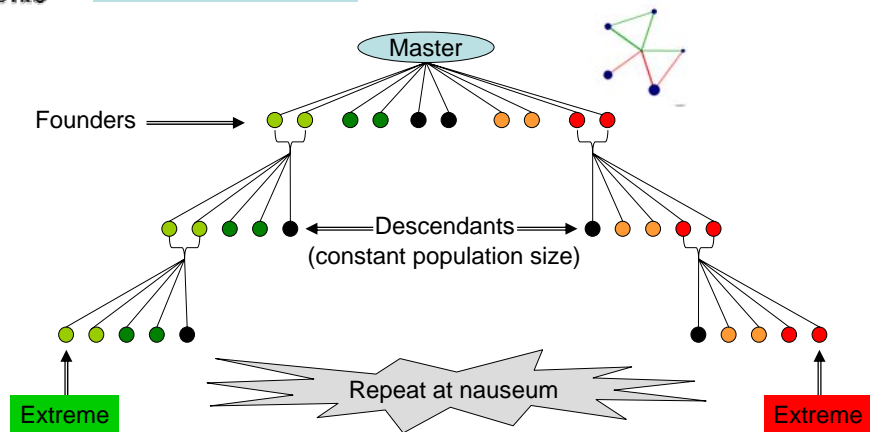


Master Genetic Load = 50.0  
 Master Phenotype = 45.4

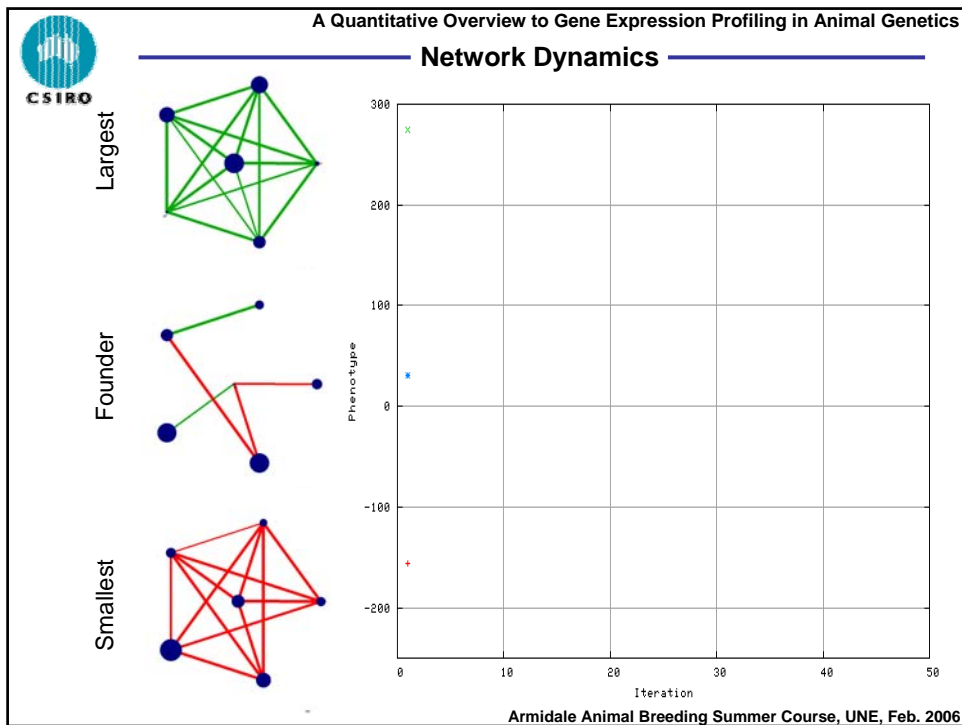
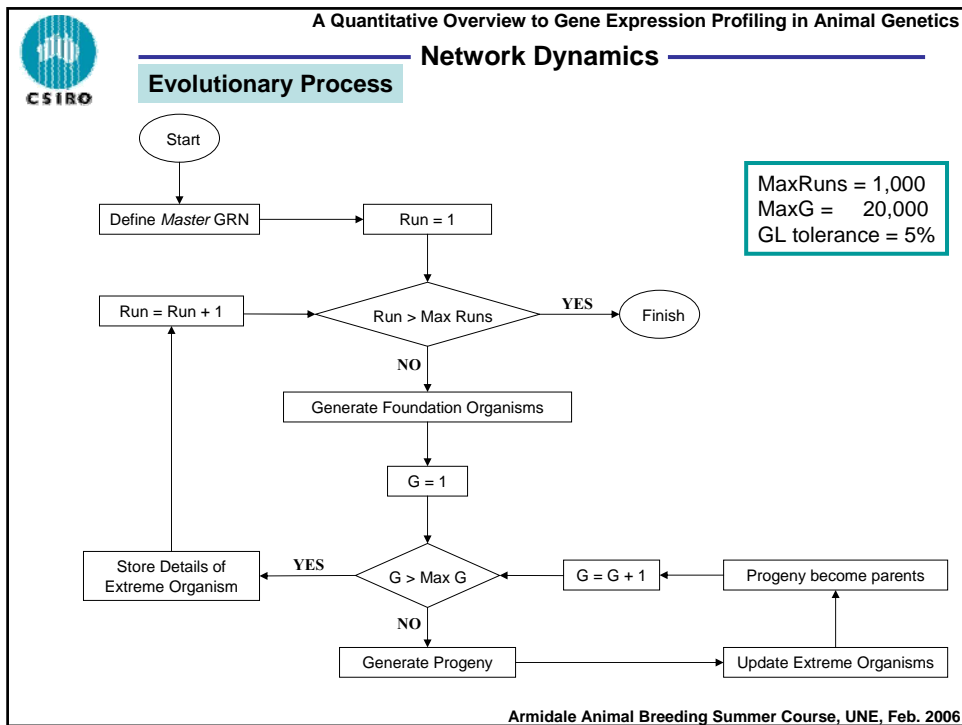


### Network Dynamics

#### Evolutionary Process



- What global changes are required to generate an extreme phenotype?
- What are the minimal changes to generate a massive change in a target gene?
- What extreme changes in phenotype can be seen after knocking out a given gene?



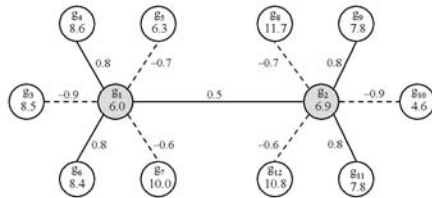


### Network Dynamics

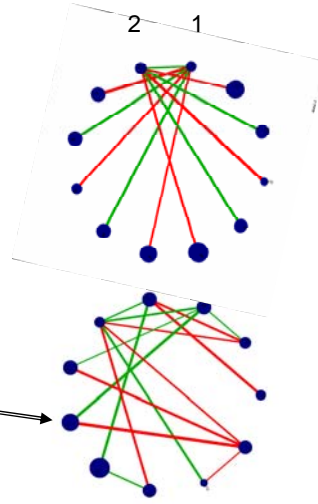
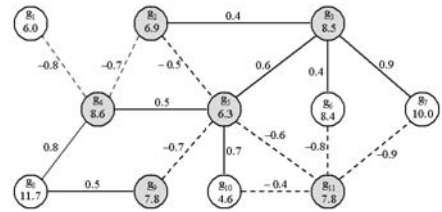
#### Examples

- Simulated (Luscombe et al. 04, Nature 431:308)

Exogenous



Endogenous



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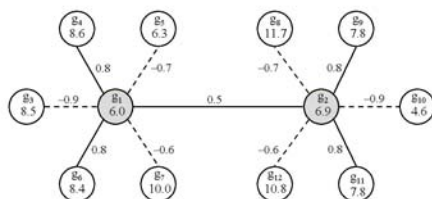


### Network Dynamics

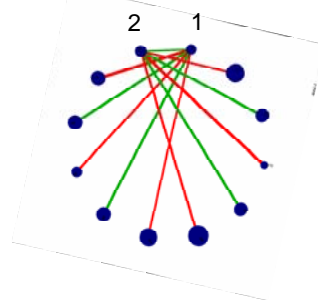
#### Examples

- Simulated (Luscombe et al. 04, Nature 431:308)

Exogenous



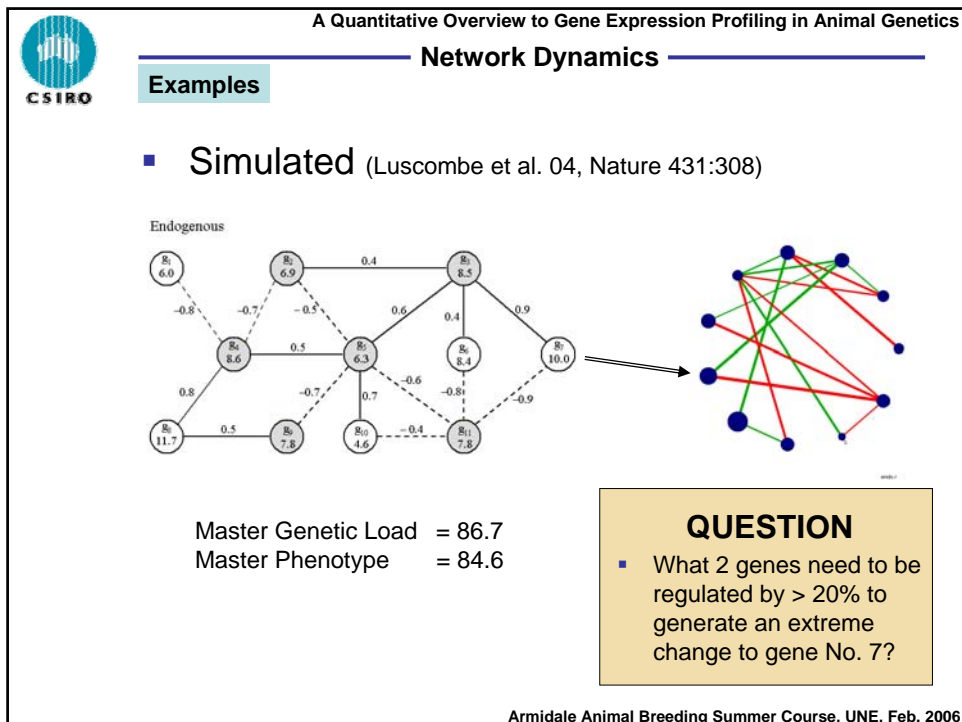
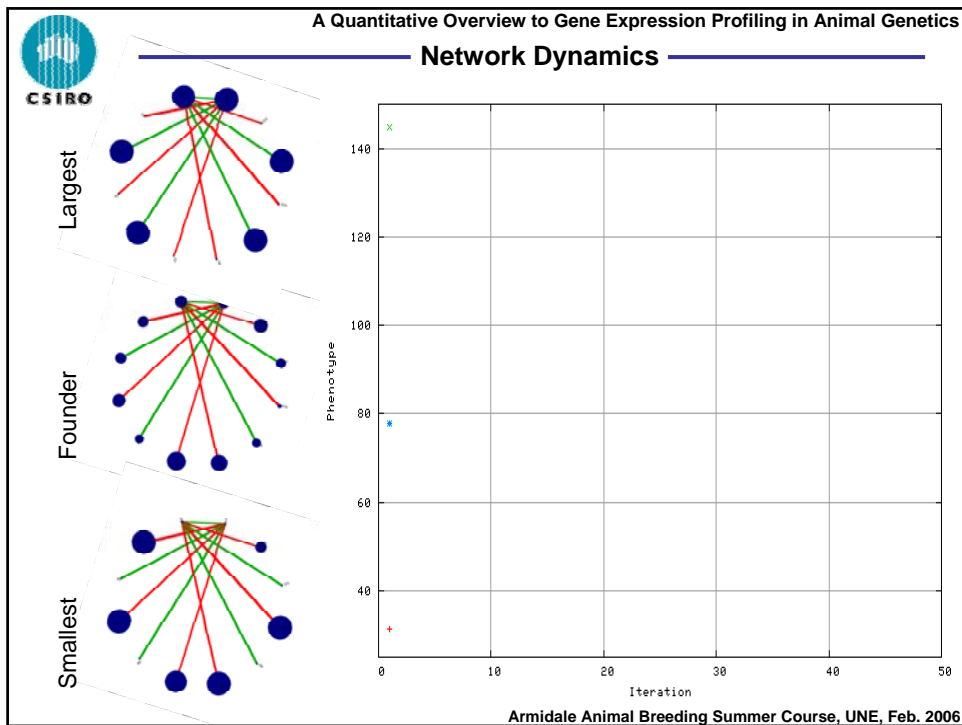
Master Genetic Load = 97.5  
Master Phenotype = 85.5



#### QUESTION

- What global changes are required to generate an extreme phenotype?

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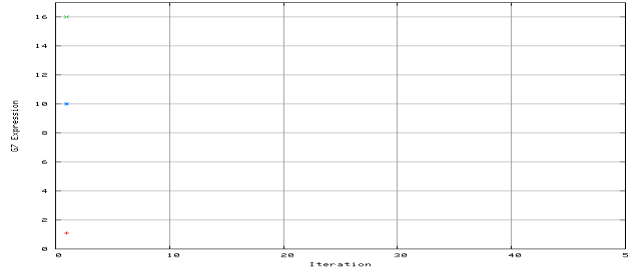
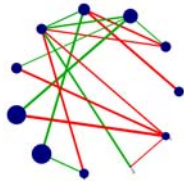




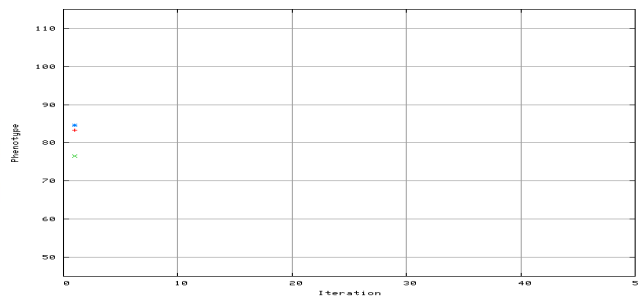
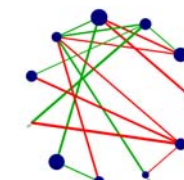
Network Dynamics

Examples

Largest



Smallest



Network Dynamics

Examples

ENDOGENOUS NETWORK

- What 2 genes need to be regulated to by > 20% to generate an extreme change to gene No. 7?

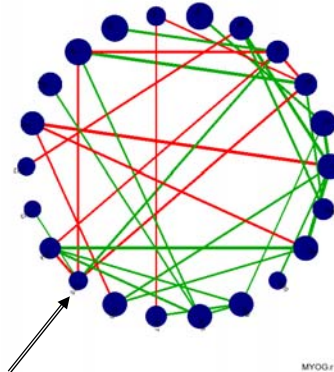
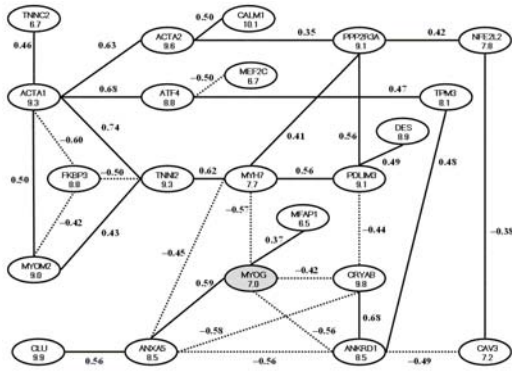
	G1	G2	G3	G4	G5	G6	G7	G8	G9	G10	G11
G1		24	27	18	29	21		23	19	39	21
G2	37		13	16	19	16		21	19	31	22
G3	30	31		22	17	18		12	17	38	24
G4	22	18	10		26	19		15	20	34	20
G5	33	26	21	25		22		17	30	28	19
G6	15	17	11	4	23			14	19	30	21
G7											
G8	7	9	3	3	6	2			14	32	15
G9	31	32	12	11	38	14		3		32	12
G10	65	51	29	45	65	31		15	54		35
G11	30	20	11	4	16	11		6	14	39	



### Network Dynamics

#### Real Example

- MYOG (Reverter et al. 05, Bioinformatics 21:1112  
Blais et al. 05, Genes & Development 19:553)



Master Genetic Load = 186.9  
Master Phenotype = 279.9

MYOG ...to be knocked out



### Network Dynamics

#### Real Example

- What negligible and extreme changes can be expected after knocking out MYOG?

```

areverte@bioserver:~/Digital_Org/MYOG
[MYOG]$ cat Table.out
Conn  Master  Closest  Small (127)  Large (673)
-----
ACTA1  6    0.3    9.1  3.78    1.5  0.43    15.5  0.45
ACTA2  3    9.7    8.9  3.78    2.1  0.85    15.0  0.80
ANKRD1 5    8.5    8.6  3.80    14.9 0.85    2.3  0.91
ANKYAL5 5    8.5    8.7  3.92    14.8 0.86    2.3  0.92
ATF4   3    8.9    8.7  3.93    4.0  2.34    13.3  1.92
CALH1  1    10.1   9.1  3.75    8.4  3.85    10.9  3.37
CAV3   2    7.2    8.8  3.95    15.2 0.62    1.9  0.70
CLU    1    9.9    8.7  3.86    6.8  3.64    11.6  3.03
CRYAB  4    9.9    8.9  3.88    13.4 1.94    4.0  2.24
DES    1    8.9    8.8  3.89    8.5  4.01    10.5  3.54
FKBP3  3    8.8    8.7  3.68    15.4 0.50    1.6  0.51
MEF2C  1    6.7    8.8  3.80    14.9 0.75    2.2  0.91
MFAP1  1    6.5    8.8  3.73    11.0 3.20    7.8  3.78
MYH7   5    7.9    9.0  3.73    6.7  3.51    12.0  2.87
MYOG   5    7.0    1.0  0.00    1.0  0.00    1.0  0.00
MYOM2  2    9.0    8.7  3.79    7.7  3.89    10.7  3.47
NFE2L2 2    7.8    8.8  3.76    13.9 1.33    3.4  1.70
PDLIM3 4    9.1    9.0  3.78    1.8  0.68    15.3  0.58
PPP2R3A 4    9.1    9.0  3.66    2.0  0.83    15.1  0.70
TNMC2  1    6.7    8.9  3.87    9.2  3.76    10.0  3.65
TNMI2  5    9.3    8.9  3.73    2.3  0.93    14.7  1.00
TPM3   2    8.1    8.8  3.84    2.7  1.19    14.4  1.24
  
```

Biological Inconsistency

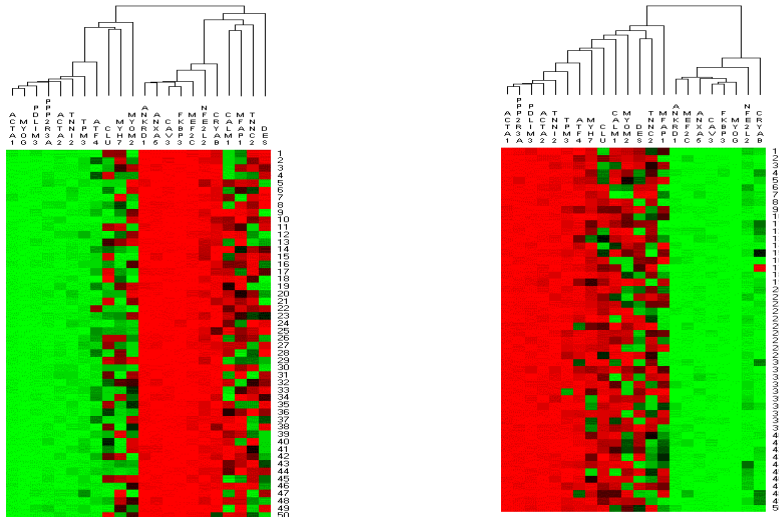
Biological Challenge



Network Dynamics

Real Example

- What negligible and extreme changes can be expected after knocking out MYOG?



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Network Dynamics

Digital Organisms

Final Remarks

- Note similarities with Genetic Algorithms.
- Potentially naïve computation of Phenotype
- ...to be improved with WGS studies:

$$P = \sum [(L_R \otimes L_G)^b] + E$$

- Further improvements from population growth rate.
- Aggravating vs Buffering effect of connections.
- Initial results warrant further research

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