

Contents

1	Introduction	1
1.1	Mouse embryonic stem cells: control of the pluripotent state	4
1.2	Downstream differentiation events triggered by the pluripotency network	7
2	Global regulation of expression in early differentiation of mESCs	15
2.1	The X:A ratio is fine tuned by two opposing processes	15
2.1.1	Introduction	15
2.1.2	The transcriptome time series captures the signature of X inactivation	19
2.1.3	No X chromosome upregulation in undifferentiated cells . . .	22
2.1.4	Ancestral X-linked genes are upregulated during differentiation	25
2.1.5	Discussion and outlook	31
2.2	Progress of differentiation is timed to compensation of X dosage . .	35
2.2.1	Introduction	35
2.2.2	Female mESC are delayed in differentiation	36
2.2.3	Methylation is linked to differentiation delay	40
2.2.4	X dosage and methylation are connected via MAPK signaling	43
2.2.5	Discussion and outlook	49
3	A mechanism for coordinated expression of adjacent genes	53
3.1	Introduction	53
3.2	Topological domains and co-expression modules	55
3.3	Both the Tsix and Xist module are essential for proper Xist expression	59
3.4	Association of co-expression and domains holds genome-wide	61
3.5	Expression fluctuations of neighboring loci are reduced in domains .	67
3.6	Proteins encoded in the same domain preferentially interact	69
3.7	Discussion and outlook	74

4 Uncovering regulation of individual genes by transcription factors in mESCs 77

4.1 Introduction 77

4.2 Statistical measures of association used for network prediction . . . 83

4.3 Gold standards for determining direct TF-gene interactions 86

4.4 How the transcriptome data was obtained 89

4.5 Benchmark of network predictions: pruning determines success . . . 91

4.6 Predicted topologies of the TF-TF network differ strongly 94

4.7 Discussion and outlook 100

5 Conclusions 103

6 Materials and Methods 129

6.1 Array Analysis for the three cell lines XO,XY and XX 129

6.2 Chapter 2 129

6.3 Chapter 3 135

6.4 Chapter 4 137

7 Supplementary Figures 143