# Ref1.2.4.1: tissue specificity

Tissue specificity of the TFs were obtained from Ravasi et al, where the expression levels of ~1200 human transcription factors in 34 different tissues were quantified using qRT-PCR. For each TF, a tissue specificity score (TSPS) was calculated as the relative entropy that quantify the extent to which the observed TF expression pattern departs from the null distribution of uniform expression across all tissues. Higher TSPS of indicates high tissue specific expression of a TF.

To make the definition clear, we have

1. put more sentences in Supptext (P6)
2. a short description in main text? (P11)

Ref1.1.4: method detail ref63 (TIP)

Ref1.2.2

Ref1.2.5

TIP (Target gene identification with a probabilistic model) is a method to identify target genes of a TF based on ChIP-seq data. The method takes into account the distance of TF binding signals from the transcription start site of genes and the characteristic binding profile of the TF. The method has been published in Cheng et al (Bioinformatics, 2011).

1. update the citation
2. 1-2 sentence description in supp-text?

Ref1.4.6: siRNA validation

We apologize that we did not explain this issue clearly. The number of targets shown in FigS.1D is the number of targets of a TF in all cell lines in which the ChIP-seq data is available. However, the siRNA experiment was only performed in K562, so the selection of TF was based on their target number in K562 only. FigS.1E shows the TFs for which at least 150 target genes were identified by both the peak-based and TIP methods. Note CTCF also meet the criteria, but it was excluded for its specialty. To clarify this,

(1) We update FigS. 1D by adding two new columns providing the number of K562 specific targets by peak-based and TIP methods.

(2) update caption of FigS. 1E

Ref1.5.0.1: binding expression correlation

We apologize that the correlations in Table S3D were mislabeled. It should be (T:0.622 M:0.626 B:0.56). Thus, it is valid to claim that TFs at the top and middle levels exhibit a greater degree of correlation between binding and expression. But we do notice the weak significance of correlation with h. In fact, we find the correlation between h and expression is mainly caused by enrichment of general TFs at the top level. If general TFs are excluded, correlation between h and expression is not significant any more. Thus,

1. correct Table S3D
2. mention the results of excluding general TFs somewhere

Ref1.5.04: more tightly regulated

This is a nice suggestion. To address this, we calculated the expression variation of TFs (log10 expression) by using the RT-PCR data in 34 tissues from Ravasi et al. We find that there is no significant different between TFs in different levels in terms of their expression variations.

It would be more interesting if we know the internal expression noises of each TF, e.g. the variation of expression levels of a TF in a population of single cells. This can more accurately represent the regulatory tightness for a TF. Unfortunately, such type of data is still not available.

1. Prepare a new Table.

Ref1.5.08: null model of Figure 4B

The null model assumes that the interaction between TFs is independent of their levels. That is, the interaction is randomly distributed in the hierarchical network. Thus, the probability of interaction for any TF-pair is calculated as the number of observed interactions being divided by the total possible TF-TF pairs. We described this in caption of Figure S6 “TF-TF interaction”.

Ref1.5.09: Is the enrichment of MM, TT and TM interactions a result of the hierarchical structure of the network?

The hierarchical structure is determined based on the TF-TF regulatory relationships from ChIP-seq data. We don’t think that the enrichment of MM, TT and TM interactions is a result of the hierarchical structure of the network for the following reasons.

1. physical interaction of TFs is from PPI. It is not a result of the hierarchical structure of the network.
2. Physical co-association of TFs is based on the significance of their co-binding with all targets. It is also not a result of hierarchical structure of the network.
3. The co-cooperativity of TFs is based on comparison of expression of shared and unique target genes. Again, this is not impacted by hierarchical structure of the network.

Ref1.5.10: clarify source network

As also described in Ref1.5.09, the physical co-association and the co-cooperativity are both determined based on all targets of TFs (TF-gene network).

1. Clarify this somewhere in main text or suppl

Ref3.m5: adding \* in Figure 4B

 We think this is a nice suggestion. We have revised the figure by adding \* atop of significant bars.