Understanding signaling network evolution: A quantitative approach

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***Short Abstract* —To gain insight on signaling network evolution, we analyzed the conservation of human phosphorylation sites. While residues phosphorylated in human are highly conserved across multiple eukaryotic species, their flanking regions seem under positive selection, suggesting that kinetics of phosphorylation and interactions may be under pressure to change during evolution to fine-tune cell signaling. To detect parts of signaling network that could have undergone such changes, we are developing computational framework to incorporate published and in-house quantitative data for predicting binding affinity of ancestral signaling proteins with their ligands. I will present preliminary work on predicting binding affinity of PDZ domains.**

***Keywords* — Cell signaling, network evolution, protein phosphorylation, binding affinity, PDZ domains.**

I. Introduction

SIGNAL transduction involves assimilation of environmental and intra-cellular cues to elicit appropriate cellular behaviors. Underlying signal transduction is a network of tightly regulated biochemical reactions and protein-protein interactions. Perturbation to this signaling network by genetic changes can lead to dysfunctional cellular activities and diseases like cancer [1]. A prevailing biological question is how has signaling network evolved to its present state to encode complex and dynamic cellular behaviors. To gain insight to this question, we analyzed the conservation of human phosphorylation sites across multiple eukaryotic species, given that protein phosphorylation is one main regulatory mechanism in signal transduction [2].

II. Result

We focus on global conservation analysis of ~6,000 human phosphorylation sites collected in PhosphoSite and Phospho.ELM databases that were detected in low throughput experiments. These phospho-sites are found in ~ 2000 proteins. Orthologous sequences in 22 eukaryotic species spanning between human and yeast for each phospho-protein are inferred using *Inparanoid* program, followed by multiple sequence alignment with *Mafft* program.

Not unexpected, residues phosphorylated in human are overall more conserved than other residues. However, we found evidence of positive selection in sequence regions flanking phosphorylated tyrosines. As amino acids in the flanking regions are known to affect phosphorylation and interaction by kinases and phospho-binding protein domains respectively, we suspect kinetics of phosphorylation and interaction may have being under pressure to change during evolution to fine-tune cell signaling. We also suspect similar things may have occurred in interactions mediated by other peptide-binding protein domains in cell signaling [3].

Changes to phosphorylation and interaction kinetics in the context of signal regulatory circuits such as feedback loop can affect cellular behaviors. Hence, to better understand evolution of signaling network, it is essential to detect parts of the network that had undergone considerable changes in binding affinity to subsequently elucidate their cellular effects from a network context. A prerequisite is to predict binding affinity of ancestral signaling proteins with their ligands, which entail predictive models that take into account mutations in both proteins and ligands. Preliminary works using Support Vector Regression (SVR) on recently published PDZ domain binding affinity data [4] shows promising result. We are developing computational framework to incorporate published and in-house quantitative data to improve binding affinity prediction.

III. Conclusion

Evolutionary analysis on known protein phosphorylation sites had lead us to hypothesize that parts of signaling networks had undergone phosphorylation/interaction kinetics during evolution. We are now developing computational approaches to detect these parts to better understand evolution of biological functions.

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