World Journal of Gastroenterology

Online Submissions: http://www.wjgnet.com/1007-9327office
wjg@wjgnet.com
doi:10.3748/wjg.v16.i28.3491

World J Gastroenterol 2010 July 28; 16(28): 3491-3498
ISSN 1007-9327 (print)

REVIEW

Protein-protein interaction map is a key gateway into liver regeneration

Chao Xie, Jin Gao, Run-Zhi Zhu, Yun-Sheng Yuan, Hong-Lin He, Qiu-Shi Huang, Wei Han, Yan Yu

Chao Xie, Run-Zhi Zhu, Yun-Sheng Yuan, Hong-Lin He, Yan Yu, School of Agriculture and Biology, Shanghai Municipality Key Laboratory of Veterinary Biotechnology, Shanghai Jiao Tong University, Shanghai 200240, China
Chao Xie, Qiu-Shi Huang, Department of Food Science, Cornell University, Ithaca, NY 14853, United States
Jin Gao, Wei Han, Laboratory of Regeneromics, School of Pharmacy, Shanghai Jiao Tong University, Shanghai 200240, China
Author contributions: Xie C, Gao J, Zhu RZ, Yuan YS, He HL, Huang QS, Han W and Yu Y contributed equally to this review; Xie C wrote the paper; Gao J, Zhu RZ, Yuan YS and He HL contributed new ideas and some new data; Huang QS, Han W and Yu Y revised the paper; Xie C and Yu Y designed the review.
Supported by Chinese Human Liver Proteome Project, No. 2004BA711A19-08 and National 863 Project, No. 2007AA02Z100
Correspondence to: Yan Yu, PhD, School of Agriculture and Biology, Shanghai Municipality Key Laboratory of Veterinary Biotechnology, Shanghai Jiao Tong University, 800 Dongchuan Road, Shanghai 200240, China. yanyu@sjtu.edu.cn
Telephone: +86-21-34205833 Fax: +86-21-34205833
Received: February 2, 2010 Revised: April 6, 2010
Accepted: April 13, 2010
Published online: July 28, 2010

Abstract

Recent studies indicate that the process of liver regeneration involves multiple signaling pathways and a variety of genes, cytokines and growth factors. Protein-protein interactions (PPIs) play a role in nearly all events that take place within the cell and PPI maps should be helpful in further understanding the process of liver regeneration. In this review, we discuss recent progress in understanding the PPIs that occur during liver regeneration especially those in the transforming growth factor β signaling pathways. We believe the use of large-scale PPI maps for integrating the information already known about the liver regeneration is a useful approach in understanding liver regeneration from the standpoint of systems biology.

Key words: Liver regeneration; Protein-protein interaction; Protein-protein interaction maps; Transforming growth factor β

Peer reviewer: Isabel Fabregat, PhD, Associate Professor, Laboratori d'Oncologia Molecular, Institut d'Investigación Biomèdica de Bellvitge, Gran Via, Km 2,7, L'Hospitalet, 08907 Barcelona, Spain

Xie C, Gao J, Zhu RZ, Yuan YS, He HL, Huang QS, Han W, Yu Y. Protein-protein interaction map is a key gateway into liver regeneration. *World J Gastroenterol* 2010; 16(28): 3491-3498 Available from: URL: http://www.wjgnet.com/1007-9327/full/v16/i28/3491.htm DOI: http://dx.doi.org/10.3748/wjg.v16.i28.3491

INTRODUCTION

Liver has the capacity to regenerate by a process of compensatory growth following injury and various molecular and cellular pathways are involved in this process[1-4]. It is still difficult to understand precisely how the process of liver regeneration is regulated. Previous studies of liver regeneration have been made at the functional, cellular, molecular or gene level[5]. It is generally believed that most cellular processes are determined by protein-protein interactions (PPIs)[6,7] and, therefore, PPIs maps provide a valuable framework for a better understanding of the functional organization of the proteome during liver regeneration[2,8,9]. Various methods have been used to study the functions of specific proteins during liver regeneration. In this review, we describe the use of high-throughput experimental methods and algorithmic predictions to unravel the complex processes of liver regeneration through the use of PPI maps.

LIVER REGENERATION

Source of liver cells and genes analysis in liver regeneration processes

Liver regeneration can be seen as a timely sequence of

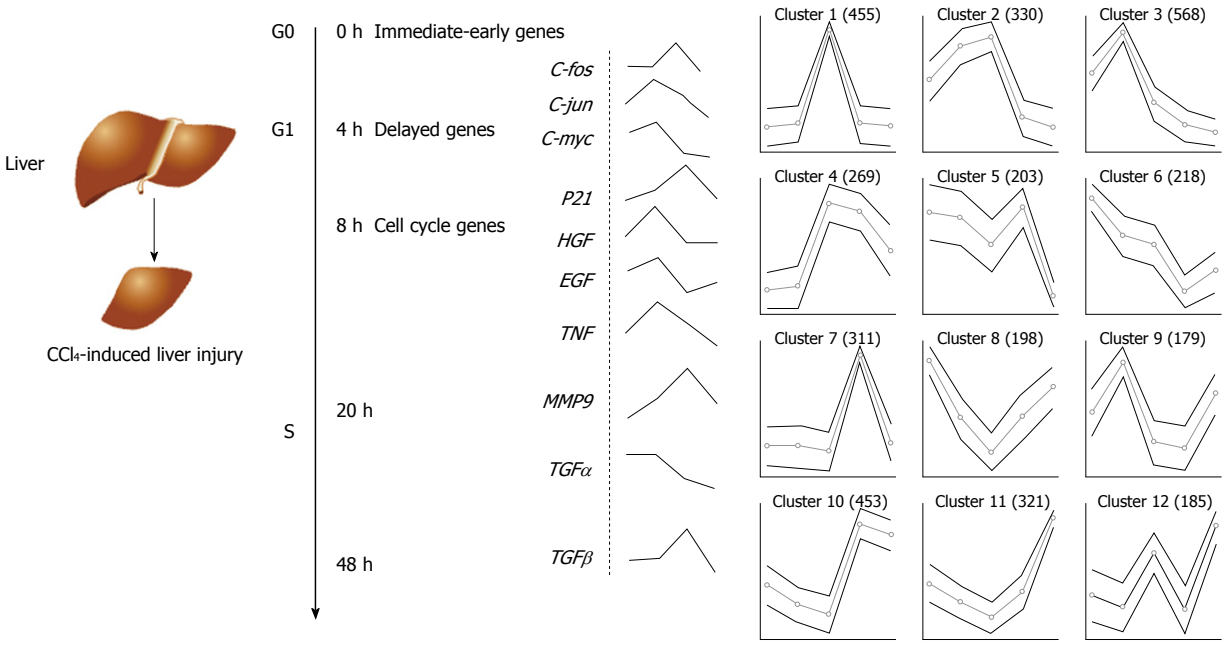

Figure 1 Hepatocyte cell cycle during liver regeneration and clustering algorithm of gene expressions in gene chip during mouse liver regeneration following CCl_4-induced liver injury. Some important genes are activated in the regenerating liver after partial hepatectomy or hepatocellular injury induced by CCl_4. Liver regeneration can be divided into four phases: G0: Corresponds to approximately the first 4 h; G1: Quiescent cells enter the cell cycle during production of hepatocyte growth factor (HGF), epidermal growth factor (EGF), tumor necrosis factor (TNF), *etc.*; S: Chromosomal DNA is replicated and peak DNA production occurs at approximately 24 h; then, after 48 h or more, the process of regeneration is terminated. The curves following the gene products and the dashed line represent the changes in gene expressions measured in our lab using Gene Chip® mouse 430 2.0 during liver regeneration following CCl_4-induced liver injury. Changes on the level of gene expression were log2-transformed using signal value at 0 h time-point as baseline. Genes with their expression levels varying at least 2-fold between any two time-points were subjected to hierarchical clustering analysis. TGF: Transforming growth factor; MMP9: Matrix metalloproteinase 9.

Table 1 Liver cell types and their functions during liver regeneration

Cell types	Functions
Hepatocytes	Organized in single cell plates; perform metabolic and detoxification function; can secrete HGF, IL-6, proteases and protease inhibitors
Sinusoidal endothelial cells	Involved in endocytosis and metabolism of molecules; can produce TGFβ, HGF, IL-6 and nitric oxide
Biliary epithelial cells	Can promote fibrogenesis by attraction of hepatic stellate cells and can secrete cytokines such as MCP-1 and IL-6
Kupffer cells	Major producers of cytokines such as TNF and IL-6
Hepatic stellate cells	Store vitamin A and secrete laminins, collagens and growth factor: HGF, EGF, TGFβ and cytokines IL-6; also produce MMPs
Oval cells	Can differentiate to biliary and hepatocytes lineage

HGF: Hepatocyte growth factor; IL-6: Interleukin-6; EGF: Epidermal growth factor; TGF: Transforming growth factor; MCP: Monocyte chemotactic protein; MMP: Matrix metalloproteinase; TNF: Tumor necrosis factor.

morphological events; resulting in the reconstitution of the lost liver mass following surgical resection or carbon tetrachloride (CCl_4) induced injury. Depending on the nature of the regenerative processes, several sources of liver cells are involved[4,10,11] (Table 1).

After partial hepatectomy (PH) or toxic forms of liver injury, some immediate early genes are expressed simultaneously in the liver. C-fos, c-jun and c-myc are up-regulated immediately and they activate hepatic non-parenchymal cells. Then tumor necrosis factor α, epidermal growth factor, hepatocyte growth factor and transforming growth factor α (TGFα) are released to provide the cooperative signals for the hepatocytes cell cycle to move from G0 through G1 to S phase, leading to DNA synthesis, and hepatocyte proliferation. TGFβ, which controls hepatocyte DNA synthesis, is blocked during the proliferative phase, but is again restored at the end of the process of regeneration. TGFβ is believed to be a key factor in returning hepatocytes to the quiescent state and ending liver regeneration[1,3,4,12].

Our laboratory has used a mouse model of CCl_4-induced liver injury to investigate changes of gene expression during the process of liver regeneration (Figure 1). Changes in the expression of 3642 genes were detected during the process of liver regeneration by microarray analysis[13] (Figure 1). Genes whose expression levels varied at least 2-fold at any time-point were subjected to self-organizing analysis, using Cluster 3.0 (Stanford University). Their specific functions of each cluster were analyzed and will be described elsewhere (manuscript in preparation).

Termination response during liver regeneration

Many research studies have focused on the regulation of the initiation and proliferation phases of liver regeneration. Although the molecular mechanisms for termination of liver regeneration are still not completely understood, TGFβ and activins, which belong to the TGFβ superfam-

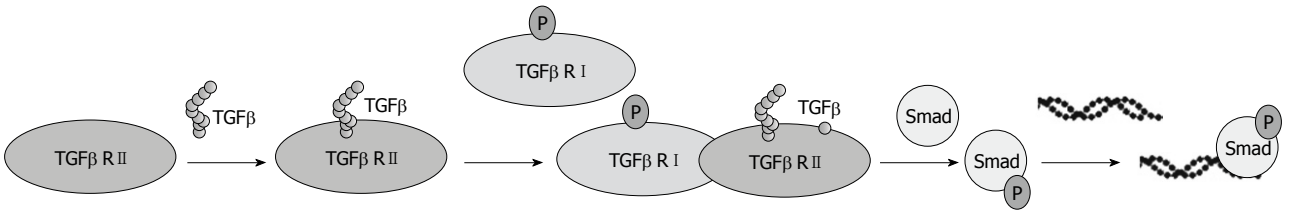

Figure 2 The classical and basic view of transforming growth factor β signaling pathway during liver regeneration. Generally speaking, transforming growth factor β (TGFβ) superfamily ligands bind to a type Ⅱ receptor (RⅡ), which recruits and phosphorylates a RⅠ. The RⅠ then phosphorylates receptor-regulated Smads. Then complexes accumulate in the nucleus where they act as transcription factors and participate in the regulation of target gene expression.

ily, appear to be important[14]. TGFβ and activin A may bind to their high-affinity cell surface type Ⅱ receptor (TGFβRⅡ) and ActRⅡ or ActRⅡb, respectively and TGFβ inhibits G1 to S phase transition in hepatocytes. TGFβ levels rise rapidly after CCl_4-induced injury and, therefore, TGFβ is known to have growth inhibitory effects on liver regeneration. So, at its simplest, the basic TGFβ signals through its Type Ⅰ and Type Ⅱ receptors (TGFβRⅠ and TGFβRⅡ) cause phosphorylation of Smad proteins and activate the Smads complex that controls transcription (Figure 2). Dierssen *et al*[15] demonstrated that gp130-dependent Stat3 activation and concomitant suppressor of cytokine signaling 3 (Socs3) is involved in timing of DNA synthesis during liver regeneration. Also, Riehle *et al*[16] showed that Socs3 modulates several signaling pathways and involved in physiological proliferative processes and protects hepatocyte proliferation in liver regeneration. Therefore, there is more to be understood about the TGFβ signaling pathway and its effects in termination of regeneration, but this still should underscore the complexity of other related pathways and their contribution to the process of termination of liver regeneration[17]. Further studies need to be conducted on understanding the mechanism of liver regeneration.

PPIs AND CORRESPONDING MAPS

PPIs refer to the association of protein molecules and PPI maps/networks summarize large amounts of PPI data, both from experiments and prediction.

There are a multitude of methods to detect PPI including biochemical, physical/biophysical and theoretical methods. The yeast two-hybrid (Y2H) technology is one of the most reliable and feasible experimental biochemical methods to detect PPI since it was established in 1989[18]. The improvement of Y2H techniques can provide high-throughput methods which are widely used in proteome studies of PPIs. Co-immunoprecipitation (CoIP) is also considered to be the gold standard assay for PPIs, especially when it is performed with endogenous proteins. Pull-down assays are a common variation and immunoprecipitation and immunoelectrophoresis are used identically, although this approach is more amenable for an initial screening of interacting proteins[19]. Fluorescence resonance energy transfer is a common biophysical technique used for observing the interactions of two different proteins[20]. There are many ways to detect PPI and each of the approaches has its own strengths and weaknesses, therefore, it is wise to determine the specific method after weighing the advantages and disadvantages.

Visualization of PPI is a popular application and already, the map of PPI among *Saccharomyces cerevisiae*[21], *Drosophila melanogaster*[22], *Caenorhabditis elegans*[6], constructed through a series of experiments and algorithmic predictions, has proved its usefulness for analyzing complex gene regulation and cellular behavior[23]. Similar efforts for PPI maps for human are ongoing and the first human PPI map was constructed from more than 3000 PPIs[24] (Table 2). Recently, the size of PPI maps in different organisms were estimated (with the PPI map for humans being nearly 650 000 PPIs) and the size is strongly believed to have correlation with the organism's apparent biological complexity[25].

Although the PPI maps are still far from complete, whether its deciphered from DNA parts are molecules, cells, or living organisms, a PPI map provides an insight into the systems biology. Our current challenge is to know how large-scale PPI maps possible functions and to understand how cells operate in an integrated manner to carry out phenotypic functions, besides increasing the coverage and accuracy of the existing and novel PPI data sets[39,40].

PEELING THE PPI MAPS DURING LIVER REGENERATION

During liver regeneration, cytokine, growth factors and metabolic pathways were both active after PH or CCl_4-induced liver injury, and the pathways interacted with each other[2]; although, there is indeed a flow of information *via* interactions between DNA, RNA and proteins on which this review will mainly focus. More research about PPI maps is on the level of proteome (Table 2), and these PPI maps will reveal the connectivity of the proteome. If we construct a PPI map during liver regeneration, it will reflect the particular cellular or unique signaling pathway status. As to the PPI map analysis, the so called small-word and scale-free behavior are considered, which indicated that in the maps only few nodes (stand for proteins) are highly connected with others (hub protein) and most of the nodes are connected with only a few nodes (low degree)[41]. In this processes, to capture the changes in protein connectivity and find the key signaling pathways, especially those that interact, is most attractive. For the mechanisms of liver regeneration to be completely understood, a multitude of PPI maps must be coordinated[37].

Table 2 Some protein-protein interaction maps established in recent years

Researcher	Yr	Level	Model organisms	Method	PPI numbers
Bartel *et al*[26]	1996	Proteome	*T7 phage*	Y2H	25
Uetz *et al*[21]	2000	Proteome	*Saccharomyces cerevisiae*	Y2H	1389
Ito *et al*[27]	2000	Proteome	*Saccharomyces cerevisiae*	Y2H	183
Walhout *et al*[28]	2000	Proteome	Caenorhabditis elegans	Y2H	148
Ito *et al*[29]	2001	Proteome	*Saccharomyces cerevisiae*	Y2H	4549
Ho *et al*[30]	2002	Proteome	*Saccharomyces cerevisiae*	MS	367
Giot *et al*[22]	2003	Proteome	Drosophila melanogaster	Y2H	20240
Colland *et al*[31]	2004	Pathway	Caenorhabditis elegans	Y2H	755
Stanyon *et al*[32]	2004	Cell cycle	Drosophila melanogaster	Y2H	20000
Li *et al*[6]	2004	Proteome	Caenorhabditis elegans	Y2H	3955
Lehner *et al*[33]	2004	mRNA	Human	Y2H	247
Formstecher *et al*[34]	2005	Proteome	Drosophila melanogaster	Y2H	2300
Stelzl *et al*[24]	2005	Proteome	Human	Y2H	3083
Rual *et al*[8]	2005	Proteome	Human	Y2H	2529
Ewing *et al*[35]	2007	Proteome	Human	MS	24540
Parrish *et al*[36]	2007	Proteome	*Camp. jejuni.*	Y2H	11687
Gao *et al*[37]	2008	Liver regeneration	Human	Y2H	64
Chen *et al*[38]	2008	Protein degradation	Human	Y2H	114

Y2H: Yeast two-hybrid; MS: Mass spectrometry; PPI: Protein-protein interaction.

PPI maps and TGFβ signaling pathway

Understanding the processes and mechanisms of liver regeneration involves recognizing components in liver regeneration system, the dynamic change of these components and their interactions[42].

In this review, it is pointed out that PPI maps (or PPI data) are closely correlated with TGFβ regulated Smad signaling pathways during liver regeneration. Colland *et al*[31] have used Y2H technique identified 755 interactions, mainly in a focused analysis of TGFβ signaling pathways and have constructed the PPI maps. They used this method to analyse LMO4, HYPA, KIAA1196 and LAPTm5 proteins, which are additional proteins involved in regulation of TGFβ signaling pathways. Also, they present an integrated approach for the identification of new factors implicated in TGFβ signaling pathway involved in several human pathologies and in the termination of liver regeneration. From this point of view, we can apply this strategy to study liver regeneration.

This review focuses on PPI maps and liver regeneration, and pays attention to the TGFβ signaling pathway. The PPI maps were constructed containing proteins related to the TGFβ signaling pathway and some of these proteins may have potential functions on the termination of liver regeneration (Figure 3). It can be easily used to find the key proteins in this process and additional experiments should be done to validate this hypothesis. Regardless, it is confirmed that the PPI map is an effective tool to study liver regeneration.

A PPI maps acting during liver cell proliferation

Gao *et al*[37] constructed a PPI map of transcription factors acting during liver regeneration which contains 32 regulatory proteins. Among them, 27 transcription factor genes that might have roles in the control of liver regeneration and five other genes that encode signal transducers might modulate transcription. After using a matrix mating Y2H technique, a PPI map in which all the components are related with liver cell proliferation was constructed (Figure 4) and some of the interactions were validated by α-glutathione S-transferase pull-down and CoIP assays. From this PPI map, Gao *et al*[37] pointed out that ATF3, a member of the mammalian activation transcription factor/cAMP responsive element-binding protein family of transcription factors, interacts with FHL2 which may be an important interaction during liver regeneration, especially for liver cell proliferation. When it comes to the termination response during liver regeneration, FHL2 and ATF3 may form a complex which abolishes its function on DNA synthesis and might terminate the liver regeneration. Also, it is possible that FHL2 may interact with Stat3 to inhibit its function in activating downstream gene expression that is necessary to terminate the liver regeneration. Nearly all the interactions in this map are growth repressors during liver regeneration and this is one of the ways in which the termination of hepatocyte proliferation and liver regeneration is regulated. Although this is one hypothesis for termination of liver regeneration, there is still growing evidence which shows that it is feasible to understand liver regeneration.

This PPI map is only a small-scale map and already can make sense of termination of liver regeneration. It would be no exaggeration to say that if large-scale, more complicated PPI maps are constructed it will greatly help us to know more about mechanism of liver regeneration.

FUTURE PROSPECTS

During the last decade, several efforts had been made to demonstrate the mechanism of liver regeneration. But unfortunately, an important gap is the lack of understanding of liver regeneration and therefore, some difficulties appear in liver cancer treatments or liver transplantation and drug development, which remain to be

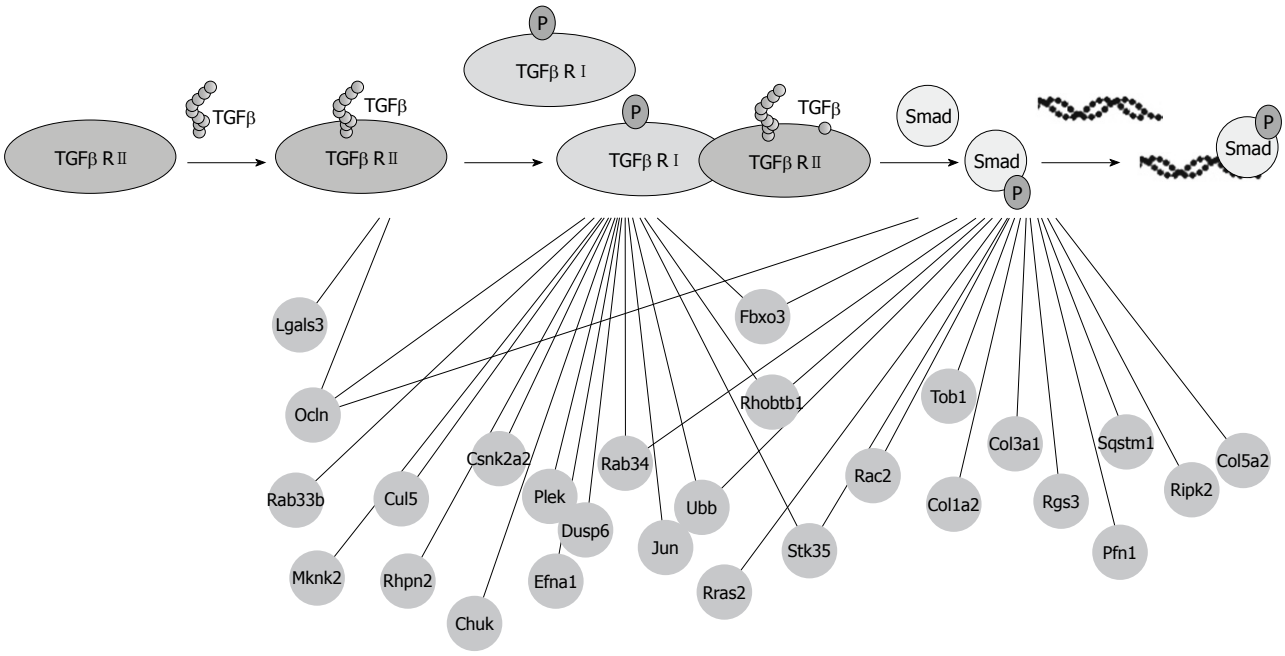

Figure 3 A protein-protein interaction comprising the transforming growth factor β signaling pathway. This figure just lists the protein-protein interactions which correlated with transforming growth factor β type Ⅰ receptor (TGFβ R Ⅰ), TGFβ R Ⅱ and Smads and all the proteins that directly interact with these three proteins which indicate that additional partners are not represented in this figure.

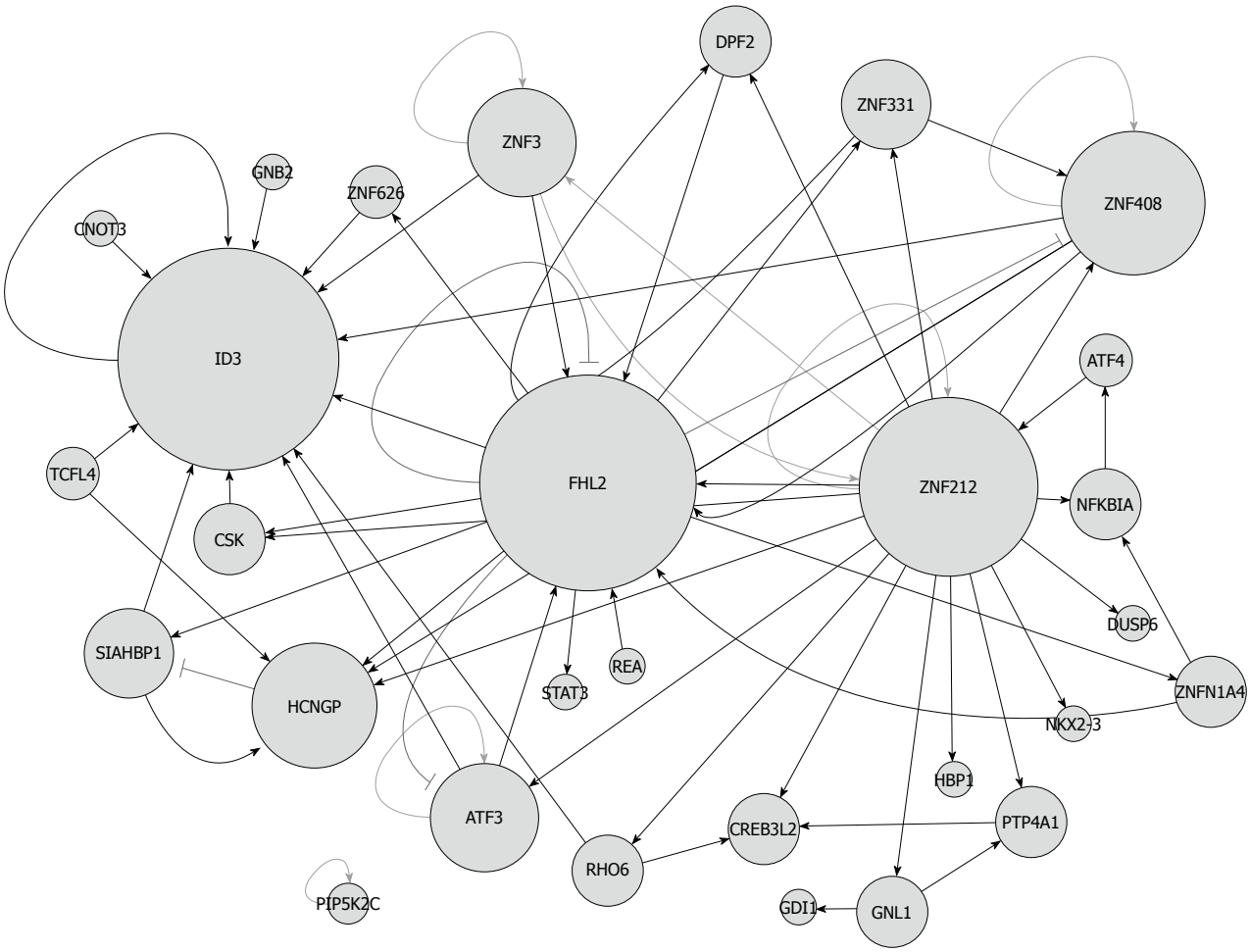

Figure 4 Protein-protein interaction maps comprising the transforming growth factor β signaling pathway of transcription factors associated with liver cell proliferation. The protein-protein interaction maps consist of different stages of liver regeneration (0 d, 0.5 d, 1.5 d, 4.5 d and 7 d after CCl_4-induced liver injury).

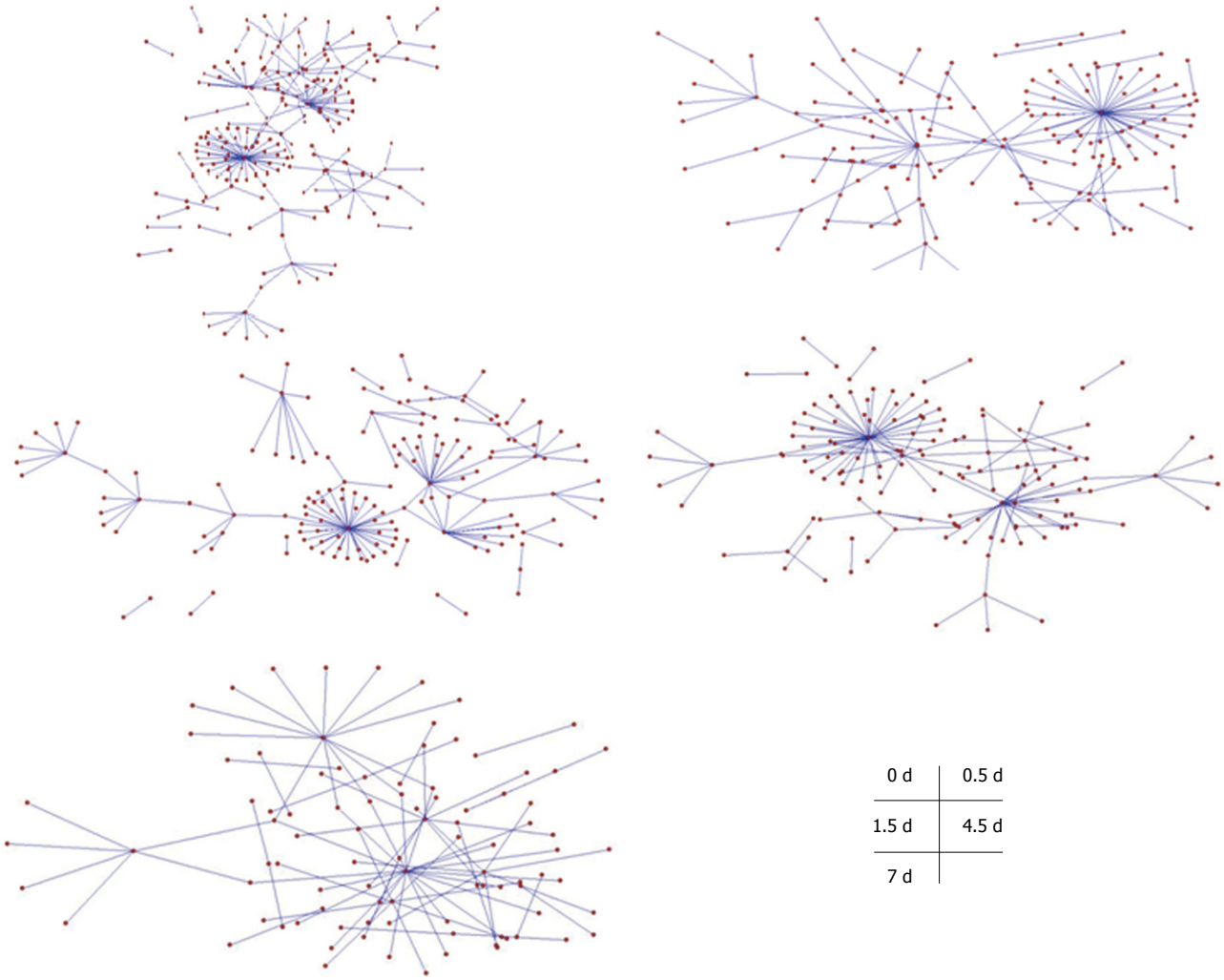

Figure 5 A protein-protein interaction map of transcription factors associated with liver cell proliferation. Node stands for protein and the edge means the two proteins are interactive; the node size symbolizes the degree of this protein (the more highly connected with others, the bigger node); the edge target arrow shape and color: delta and blue, interactions are only validated by yeast two-hybrid (Y2H); diamond and green means validated by Y2H and α-glutathione S-transferase pull-down; T and black means validated by Y2H and co-immunoprecipitation.

solved. Constructing PPI maps is a powerful step toward addressing these challenges. The most important profiles shall be discussed.

Integration of PPI data druing liver regeneration

Different high-throughput PPI data are difficult to cover even if in the same species, also, it has a higher rate of false positives than that of small-scale data[44]. However, it may be useful to increase the capacity of false positive identification in order to find out the real PPI from the noise data. The first step is to collect the correct and reliable PPI data and find several criteria which can be used to evaluate the PPI data sets. By integrating small-scale and large-scale PPI data, PPI databases have emerged and it is generally believed that a PPI database is a symbol of the level of PPIs. Although a series of PPI databases such as BIND (http://bind.ca), MIPS (http://mips.gsf.de) and DIP (http://dip.doe-mbi.ucla.edu) are popular and helpful, there are still no PPI databases for liver cells, liver regeneration or specific liver diseases, such as liver cancer. PPI data should be collected, evaluated, retrieved and systemically stored (including the detailed information for PPIs).

With liver regeneration PPI data which is integrated into the databases and is made readily accessible through the internet, researchers will be able to quickly locate the PPIs for their proteins of interest during liver regeneration, also, they will get the interpretation of PPIs in detail, which means any type of available information on proteins or protein domains can be verified. Meanwhile, we should also explore a tool that allows easy navigation in this complex of PPI databases especially for liver regeneration.

Already, the Y2H system has increasingly been applied in high-throughput applications intended to map genome-scale PPI for liver regeneration and is definitely believed to be an effective way to construct large-scale PPI maps during liver regeneration. Also, the low coverage and experimental bias call for development of computational methods to predict PPIs[45], and mining of existing inter-

action data to infer additional interactions is also a trend to enlarge the PPI database. Nowadays, lots of PPI data are obtained from different organisms and we can get PPI from interacting proteins to exhibit similar phylogenetic trees[46]. As to PPIs during liver regeneration, much more computational methods and algorithms must be fixed in order to predict PPIs during liver regeneration, also it may from signaling pathways level or proteome level.

Static and dynamic architecture of PPI maps during liver regeneration

Static PPI maps during liver regeneration, especially the PPI maps on signaling pathways, will help us to understand the different phases and different gene changes of liver regeneration. In order to understand the mechanism of liver regeneration, the PPI map furthered the understanding of the architecture of cellular machinery and revealed fundamental properties[47]. The large-scale and static PPI maps show us some information and they are quite important to understand the spatiotemporal existence of PPIs. Obviously, PPI maps are dynamic and not static, but unfortunately nearly all the PPI maps are static and do not consider the PPI strength and spatiotemporal existence let alone the types of PPI maps and do not reflect the actual situation in liver cells. Therefore, dynamic PPI maps are of more importance for cell signaling and dictate timing and intensity of map outputs. Our lab has also tried to construct dynamic PPI maps during mouse liver regeneration.

We picked up 5 major time points (0 d, 0.5 d, 1.5 d, 4.5 d and 7 d) during the mouse liver regeneration process after CCl_4-induced liver injury, in which all the proteins are transcription factors associated with TGFβ signaling pathway and constructed 5 PPI maps (Figure 5). It is easy to identify the PPIs change in the whole process of liver regeneration including numbers and the protein category. The numbers of PPIs increased at the beginning and then decreased. A detailed analysis of these PPI maps are in progress, which is a mark of the beginning of dynamic PPI maps construction.

Though no large-scale data sets are yet available on liver PPI map dynamics, the time dimension can be added by projecting time series of liver regeneration mRNA expression data onto transcription factors, allowing one method to interpret dynamic PPI maps. Researchers also need to consider addressing where and when interactions take place in different phases of liver regeneration and how they regulate the process. It will be a great help for us to know about liver regeneration if we know the full range of PPIs, from static to dynamic, and this is a useful method for studying liver regeneration over the next few years which will hopefully improve our ability to understand the PPI maps during liver regeneration.

Liver regeneration remains a fascinating project and the exact cellular and molecular mechanisms are still a mystery to us. In order to understand this phenomenon, many methods must integrate and we are just beginning to appreciate the relationship between PPIs and liver regeneration. We strongly believe that PPI maps from systems biology is a key gateway for deciphering liver regeneration. Certainly, like the sequencing of the human genome, the construction of a PPI map during liver regeneration will represent a major step along the path towards understanding the mechanisms of liver regeneration.

REFERENCES

1 **Schibler U**. Circadian rhythms. Liver regeneration clocks on. *Science* 2003; **302**: 234-235
2 **Fausto N**, Campbell JS, Riehle KJ. Liver regeneration. *Hepatology* 2006; **43**: S45-S53
3 **Michalopoulos GK**. Liver regeneration. *J Cell Physiol* 2007; **213**: 286-300
4 **Taub R**. Liver regeneration: from myth to mechanism. *Nat Rev Mol Cell Biol* 2004; **5**: 836-847
5 **Pahlavan PS**, Feldmann RE Jr, Zavos C, Kountouras J. Prometheus' challenge: molecular, cellular and systemic aspects of liver regeneration. *J Surg Res* 2006; **134**: 238-251
6 **Li S**, Armstrong CM, Bertin N, Ge H, Milstein S, Boxem M, Vidalain PO, Han JD, Chesneau A, Hao T, Goldberg DS, Li N, Martinez M, Rual JF, Lamesch P, Xu L, Tewari M, Wong SL, Zhang LV, Berriz GF, Jacotot L, Vaglio P, Reboul J, Hirozane-Kishikawa T, Li Q, Gabel HW, Elewa A, Baumgartner B, Rose DJ, Yu H, Bosak S, Sequerra R, Fraser A, Mango SE, Saxton WM, Strome S, Van Den Heuvel S, Piano F, Vandenhaute J, Sardet C, Gerstein M, Doucette-Stamm L, Gunsalus KC, Harper JW, Cusick ME, Roth FP, Hill DE, Vidal M. A map of the interactome network of the metazoan C. elegans. *Science* 2004; **303**: 540-543
7 **Gavin AC**, Aloy P, Grandi P, Krause R, Boesche M, Marzioch M, Rau C, Jensen LJ, Bastuck S, Dümpelfeld B, Edelmann A, Heurtier MA, Hoffman V, Hoefert C, Klein K, Hudak M, Michon AM, Schelder M, Schirle M, Remor M, Rudi T, Hooper S, Bauer A, Bouwmeester T, Casari G, Drewes G, Neubauer G, Rick JM, Kuster B, Bork P, Russell RB, Superti-Furga G. Proteome survey reveals modularity of the yeast cell machinery. *Nature* 2006; **440**: 631-636
8 **Rual JF**, Venkatesan K, Hao T, Hirozane-Kishikawa T, Dricot A, Li N, Berriz GF, Gibbons FD, Dreze M, Ayivi-Guedehoussou N, Klitgord N, Simon C, Boxem M, Milstein S, Rosenberg J, Goldberg DS, Zhang LV, Wong SL, Franklin G, Li S, Albala JS, Lim J, Fraughton C, Llamosas E, Cevik S, Bex C, Lamesch P, Sikorski RS, Vandenhaute J, Zoghbi HY, Smolyar A, Bosak S, Sequerra R, Doucette-Stamm L, Cusick ME, Hill DE, Roth FP, Vidal M. Towards a proteome-scale map of the human protein-protein interaction network. *Nature* 2005; **437**: 1173-1178
9 **Bader S**, Kühner S, Gavin AC. Interaction networks for systems biology. *FEBS Lett* 2008; **582**: 1220-1224
10 **Fausto N**. Liver regeneration. *J Hepatol* 2000; **32**: 19-31
11 **Mohammed FF**, Khokha R. Thinking outside the cell: proteases regulate hepatocyte division. *Trends Cell Biol* 2005; **15**: 555-563
12 **Khan AZ**, Mudan SS. Liver regeneration: mechanisms, mysteries and more. *ANZ J Surg* 2007; **77**: 9-14
13 **Yuan Y**, Wu X, Ou Q, Gao J, Tennant BC, Han W, Yu Y. Differential expression of the genes involved in amino acids and nitrogen metabolisms during liver regeneration of mice. *Hepatol Res* 2009; **39**: 301-312
14 **Zimmermann A**. Regulation of liver regeneration. *Nephrol Dial Transplant* 2004; **19** Suppl 4: iv6-iv10
15 **Dierssen U**, Beraza N, Lutz HH, Liedtke C, Ernst M, Wasmuth HE, Trautwein C. Molecular dissection of gp130-dependent pathways in hepatocytes during liver regeneration. *J Biol Chem* 2008; **283**: 9886-9895
16 **Riehle KJ**, Campbell JS, McMahan RS, Johnson MM, Beyer RP, Bammler TK, Fausto N. Regulation of liver regeneration and hepatocarcinogenesis by suppressor of cytokine signaling 3. *J Exp Med* 2008; **205**: 91-103

17 **Michalopoulos GK**. Liver regeneration after partial hepatectomy: critical analysis of mechanistic dilemmas. *Am J Pathol* 2010; **176**: 2-13
18 **Fields S**, Song O. A novel genetic system to detect protein-protein interactions. *Nature* 1989; **340**: 245-246
19 **Brymora A**, Valova VA, Robinson PJ. Protein-protein interactions identified by pull-down experiments and mass spectrometry. *Curr Protoc Cell Biol* 2004; **Chapter 17**: Unit 17.5
20 **Trugnan G**, Fontanges P, Delautier D, Ait-Slimane T. [FRAP, FLIP, FRET, BRET, FLIM, PRIM...new techniques for a colourful life] *Med Sci* (Paris) 2004; **20**: 1027-1034
21 **Uetz P**, Giot L, Cagney G, Mansfield TA, Judson RS, Knight JR, Lockshon D, Narayan V, Srinivasan M, Pochart P, Qureshi-Emili A, Li Y, Godwin B, Conover D, Kalbfleisch T, Vijayadamodar G, Yang M, Johnston M, Fields S, Rothberg JM. A comprehensive analysis of protein-protein interactions in Saccharomyces cerevisiae. *Nature* 2000; **403**: 623-627
22 **Giot L**, Bader JS, Brouwer C, Chaudhuri A, Kuang B, Li Y, Hao YL, Ooi CE, Godwin B, Vitols E, Vijayadamodar G, Pochart P, Machineni H, Welsh M, Kong Y, Zerhusen B, Malcolm R, Varrone Z, Collis A, Minto M, Burgess S, McDaniel L, Stimpson E, Spriggs F, Williams J, Neurath K, Ioime N, Agee M, Voss E, Furtak K, Renzulli R, Aanensen N, Carrolla S, Bickelhaupt E, Lazovatsky Y, DaSilva A, Zhong J, Stanyon CA, Finley RL Jr, White KP, Braverman M, Jarvie T, Gold S, Leach M, Knight J, Shimkets RA, McKenna MP, Chant J, Rothberg JM. A protein interaction map of Drosophila melanogaster. *Science* 2003; **302**: 1727-1736
23 **Guido NJ**, Wang X, Adalsteinsson D, McMillen D, Hasty J, Cantor CR, Elston TC, Collins JJ. A bottom-up approach to gene regulation. *Nature* 2006; **439**: 856-860
24 **Stelzl U**, Worm U, Lalowski M, Haenig C, Brembeck FH, Goehler H, Stroedicke M, Zenkner M, Schoenherr A, Koeppen S, Timm J, Mintzlaff S, Abraham C, Bock N, Kietzmann S, Goedde A, Toksöz E, Droege A, Krobitsch S, Korn B, Birchmeier W, Lehrach H, Wanker EE. A human protein-protein interaction network: a resource for annotating the proteome. *Cell* 2005; **122**: 957-968
25 **Stumpf MP**, Thorne T, de Silva E, Stewart R, An HJ, Lappe M, Wiuf C. Estimating the size of the human interactome. *Proc Natl Acad Sci USA* 2008; **105**: 6959-6964
26 **Bartel PL**, Roecklein JA, SenGupta D, Fields S. A protein linkage map of Escherichia coli bacteriophage T7. *Nat Genet* 1996; **12**: 72-77
27 **Ito T**, Tashiro K, Muta S, Ozawa R, Chiba T, Nishizawa M, Yamamoto K, Kuhara S, Sakaki Y. Toward a protein-protein interaction map of the budding yeast: A comprehensive system to examine two-hybrid interactions in all possible combinations between the yeast proteins. *Proc Natl Acad Sci USA* 2000; **97**: 1143-1147
28 **Walhout AJ**, Sordella R, Lu X, Hartley JL, Temple GF, Brasch MA, Thierry-Mieg N, Vidal M. Protein interaction mapping in C. elegans using proteins involved in vulval development. *Science* 2000; **287**: 116-122
29 **Ito T**, Chiba T, Ozawa R, Yoshida M, Hattori M, Sakaki Y. A comprehensive two-hybrid analysis to explore the yeast protein interactome. *Proc Natl Acad Sci USA* 2001; **98**: 4569-4574
30 **Ho Y**, Gruhler A, Heilbut A, Bader GD, Moore L, Adams SL, Millar A, Taylor P, Bennett K, Boutilier K, Yang L, Wolting C, Donaldson I, Schandorff S, Shewnarane J, Vo M, Taggart J, Goudreault M, Muskat B, Alfarano C, Dewar D, Lin Z, Michalickova K, Willems AR, Sassi H, Nielsen PA, Rasmussen KJ, Andersen JR, Johansen LE, Hansen LH, Jespersen H, Podtelejnikov A, Nielsen E, Crawford J, Poulsen V, Sørensen BD, Matthiesen J, Hendrickson RC, Gleeson F, Pawson T, Moran MF, Durocher D, Mann M, Hogue CW, Figeys D, Tyers M. Systematic identification of protein complexes in Saccharomyces cerevisiae by mass spectrometry. *Nature* 2002; **415**: 180-183
31 **Colland F**, Jacq X, Trouplin V, Mougin C, Groizeleau C, Hamburger A, Meil A, Wojcik J, Legrain P, Gauthier JM. Functional proteomics mapping of a human signaling pathway. *Genome Res* 2004; **14**: 1324-1332
32 **Stanyon CA**, Liu G, Mangiola BA, Patel N, Giot L, Kuang B, Zhang H, Zhong J, Finley RL Jr. A Drosophila protein-interaction map centered on cell-cycle regulators. *Genome Biol* 2004; **5**: R96
33 **Lehner B**, Fraser AG. A first-draft human protein-interaction map. *Genome Biol* 2004; **5**: R63
34 **Formstecher E**, Aresta S, Collura V, Hamburger A, Meil A, Trehin A, Reverdy C, Betin V, Maire S, Brun C, Jacq B, Arpin M, Bellaiche Y, Bellusci S, Benaroch P, Bornens M, Chanet R, Chavrier P, Delattre O, Doye V, Fehon R, Faye G, Galli T, Girault JA, Goud B, de Gunzburg J, Johannes L, Junier MP, Mirouse V, Mukherjee A, Papadopoulo D, Perez F, Plessis A, Rossé C, Saule S, Stoppa-Lyonnet D, Vincent A, White M, Legrain P, Wojcik J, Camonis J, Daviet L. Protein interaction mapping: a Drosophila case study. *Genome Res* 2005; **15**: 376-384
35 **Ewing RM**, Chu P, Elisma F, Li H, Taylor P, Climie S, McBroom-Cerajewski L, Robinson MD, O'Connor L, Li M, Taylor R, Dharsee M, Ho Y, Heilbut A, Moore L, Zhang S, Ornatsky O, Bukhman YV, Ethier M, Sheng Y, Vasilescu J, Abu-Farha M, Lambert JP, Duewel HS, Stewart II, Kuehl B, Hogue K, Colwill K, Gladwish K, Muskat B, Kinach R, Adams SL, Moran MF, Morin GB, Topaloglou T, Figeys D. Large-scale mapping of human protein-protein interactions by mass spectrometry. *Mol Syst Biol* 2007; **3**: 89
36 **Parrish JR**, Yu J, Liu G, Hines JA, Chan JE, Mangiola BA, Zhang H, Pacifico S, Fotouhi F, DiRita VJ, Ideker T, Andrews P, Finley RL Jr. A proteome-wide protein interaction map for Campylobacter jejuni. *Genome Biol* 2007; **8**: R130
37 **Gao J**, Li WX, Feng SQ, Yuan YS, Wan DF, Han W, Yu Y. A protein-protein interaction network of transcription factors acting during liver cell proliferation. *Genomics* 2008; **91**: 347-355
38 **Chen C**, Huang C, Chen S, Liang J, Lin W, Ke G, Zhang H, Wang B, Huang J, Han Z, Ma L, Huo K, Yang X, Yang P, He F, Tao T. Subunit-subunit interactions in the human 26S proteasome. *Proteomics* 2008; **8**: 508-520
39 **Shoemaker BA**, Panchenko AR. Deciphering protein-protein interactions. Part I. Experimental techniques and databases. *PLoS Comput Biol* 2007; **3**: e42
40 **Xia Y**, Yu H, Jansen R, Seringhaus M, Baxter S, Greenbaum D, Zhao H, Gerstein M. Analyzing cellular biochemistry in terms of molecular networks. *Annu Rev Biochem* 2004; **73**: 1051-1087
41 **Uetz P**, Dong YA, Zeretzke C, Atzler C, Baiker A, Berger B, Rajagopala SV, Roupelieva M, Rose D, Fossum E, Haas J. Herpesviral protein networks and their interaction with the human proteome. *Science* 2006; **311**: 239-242
42 **Kitano H**. Systems biology: a brief overview. *Science* 2002; **295**: 1662-1664
43 **Allen NP**, Huang L, Burlingame A, Rexach M. Proteomic analysis of nucleoporin interacting proteins. *J Biol Chem* 2001; **276**: 29268-29274
44 **von Mering C**, Krause R, Snel B, Cornell M, Oliver SG, Fields S, Bork P. Comparative assessment of large-scale data sets of protein-protein interactions. *Nature* 2002; **417**: 399-403
45 **Shoemaker BA**, Panchenko AR. Deciphering protein-protein interactions. Part II. Computational methods to predict protein and domain interaction partners. *PLoS Comput Biol* 2007; **3**: e43
46 **Goh CS**, Cohen FE. Co-evolutionary analysis reveals insights into protein-protein interactions. *J Mol Biol* 2002; **324**: 177-192
47 **Stumpf MP**, Kelly WP, Thorne T, Wiuf C. Evolution at the system level: the natural history of protein interaction networks. *Trends Ecol Evol* 2007; **22**: 366-373

S- Editor Tian L **L- Editor** O'Neill M **E- Editor** Zheng XM