Graph Theory and Networks in Biology

Oliver Mason and Mark Verwoerd

March 14, 2006

Abstract

In this paper, we present a survey of the use of graph theoretical techniques in Biology. In particular, we discuss recent work on identifying and modelling the structure of bio-molecular networks, as well as the application of centrality measures to interaction networks and research on the hierarchical structure of such networks and network motifs. Work on the link between structural network properties and dynamics is also described, with emphasis on synchronization and disease propagation.

1 Introduction and Motivation

The theory of complex networks plays an important role in a wide variety of disciplines, ranging from communications and power systems engineering to molecular and population biology [2, 13, 133, 51, 5, 3, 31, 6]. While the focus of this article is on biological applications of the theory of graphs and networks, there are also several other domains in which networks play a crucial role. For instance, the Internet and the World Wide Web (WWW) have grown at a remarkable rate, both in size and importance, in recent years, leading to a pressing need both for systematic methods of analysing such networks as well as a thorough understanding of their properties. Moreover, in sociology and ecology, increasing amounts of data on food-webs and the structure of human social networks are becoming available. Given the critical role that these networks play in many key questions relating to the environment and public health, it is hardly surprising that researchers in ecology and epidemiology have focussed attention on network analysis in recent years. In particular, the complex interplay between the structure of social networks and the spread of disease is a topic of critical importance. The threats to human health posed by new infectious diseases such as the SARS virus and the Asian bird flu [190, 7], coupled with modern travel patterns, underline the vital nature of this issue.

On a more theoretical level, several recent studies have indicated that networks from a broad range of application areas share common structural properties. Furthermore, a number of the properties observed in such real world networks are incompatible with those of the random graphs which had been traditionally employed as modelling tools for complex networks [2, 133]. The latter observation naturally poses the challenge of devising more accurate models for the topologies observed in biological and technological networks, while the former further motivates the development of analysis tools for complex networks. The common structural properties shared by diverse networks offers the hope that such tools may prove useful for applications in a wide variety of disciplines. Within the fields of Biology and Medicine, applications include the identification of drug targets, determining the role of proteins or genes of unknown function [96, 158], the design of effective containment strategies for infectious diseases [58], and the early diagnosis of neurological disorders through detecting abnormal patterns of neural synchronization in specific brain regions [162].

Recent advances in the development of high-throughput techniques in molecular biology have led to an unprecedented amount of data becoming available on key cellular networks in a variety of simple organisms [92, 43]. Broadly speaking, three classes of bio-molecular networks have attracted most

attention to date: metabolic networks of biochemical reactions between metabolic substrates; protein interaction networks consisting of the physical interactions between an organism's proteins; and the transcriptional regulatory networks which describe the regulatory interactions between different genes. At the time of writing, the central metabolic networks of numerous bacterial organisms have been mapped [152]. Also, large scale data sets are available on the structure of the protein interaction networks of *S. cerevisiae* [92, 178], *H. pylori* [151], *D. melanogaster* [67] and *C. elegans* [117, 43], and the transcriptional regulatory networks of *E. coli* and *S. cerevisiae* have been extensively studied [90, 169]. The large amount of data now available on these networks provides the network research community with both opportunities and challenges.

On the one hand, it is now possible to investigate the structural properties of networks in living cells, to identify their key properties and to hopefully shed light on how such properties may have evolved biologically. A major motivation for the study of biological networks is the need for tailored analysis methods which can extract meaningful biological information from the data becoming available through the efforts of experimentalists. This is all the more pertinent given that the network structures emerging from the results of high-throughput techniques are too complex to analyse in a non-systematic fashion. A knowledge of the topologies of biological networks, and of their impact on biological processes, is needed if we are to fully understand, and develop more sophisticated treatment strategies for, complex diseases such as cancer [184]. Also, recent work suggesting connections between abnormal neural synchronization and neurological disorders such as *Parkinson's disease* and *Schizophrenia* [162] provides strong motivation for studying how network structure influences the emergence of synchronization between interconnected dynamical systems.

The mathematical discipline which underpins the study of complex networks in Biology and elsewhere, and on which the techniques discussed throughout this article are based, is *graph theory* [47]. Alongside the potential benefits of applying graph theoretical methods in molecular biology, it should be emphasized that the complexity of the networks encountered in cellular biology and the mechanisms behind their emergence presents the network researcher with numerous challenges and difficulties. The inherent variability in biological data, the high likelihood of data inaccuracy [186] and the need to incorporate dynamics and network topology in the analysis of biological systems are just a sample of the obstacles to be overcome if we are to successfully understand the fundamental networks involved in the operation of living cells. Another important issue, which we shall discuss at various points throughout the article, is that the structure of biological and social networks is often inferred from sampled subnetworks. The precise impact of sampling on the results and techniques published in the recent past needs to be understood if these are to be reliably applied to real biological data.

Motivated by the considerations outlined above, a substantial literature dedicated to the analysis of biological networks has emerged in the last few years, and some significant progress has been made on identifying and interpreting the structure of such networks. Our primary goal in the present article is to provide as broad a survey as possible of the major advances made in this field in the recent past, highlighting what has been achieved as well as some of the most significant open issues that need to be addressed. The material discussed in the article can be divided naturally into two strands, and this is reflected in the organisation of the document. The first part of the article will primarily be concerned with the properties and analysis of cellular networks such as protein interaction networks and transcriptional regulatory networks. In the second part, we turn our attention to two important applications of Graph Theory in Biology: the phenomenon of synchronisation and its role in neurological disorders, and the interaction between network structure and epidemic dynamics.

In the interests of clarity, we shall now give a brief outline of the main topics covered throughout the rest of the paper. In Section 2, we shall fix the principal notations used throughout the paper, and briefly review the main mathematical and graph theoretical concepts that are required in the remainder of the article. As mentioned above, the body of the article is divided into two parts. The first part consists of Sections 3, 4 and 5 and the second part of Sections 6 and 7. At the end of each major section, a brief summary of the main points covered in that section is given.

In Section 3, we shall discuss recent findings on the structure of bio-molecular networks and discuss

several graph models, including *Scale-Free* graphs and *Duplication-Divergence* models, that have been proposed to account for the properties observed in real biological networks. Section 4 is concerned with the application of graph theoretical *measures of centrality or importance* to biological networks. In particular, we shall concentrate on the connection between the centrality of a gene or protein within an interaction network and its likelihood to be *essential* for the organism's survival. In Section 5, we shall consider the *hierarchical structure* of biological networks. In particular, we shall discuss *motifs in bio-molecular networks* and the identification of (typically larger) functional modules.

In the second part of the article, we shall discuss two major applications of Graph Theory to Biology. Section 6 is concerned with a number of issues and results related to the phenomenon of synchronization in networks of inter-connected dynamical systems and its relevance in various biological contexts. Particular attention will be given to suggested links between *patterns of synchrony* and *neurological disorders*. In Section 7, we shall discuss some recent work on the influence that the structure of a social network can have on the behaviour of various disease propagation models, and discuss the epidemiological significance of these findings. Finally, in Section 8 we shall present our concluding remarks and highlight some possible directions for future research.

2 Definitions and Mathematical Preliminaries

The basic mathematical concept used to model networks is a *graph*. In this section, we shall introduce the principal notations used throughout the paper, and recall some basic definitions and facts from graph theory. While the material of this section is mathematical in nature, we shall see in the remainder of the paper that all of the concepts recalled here arise in real biological networks. Furthermore, the notation and nomenclature introduced in this section will enable us to discuss the various biological networks encountered throughout the paper in a uniform and consistent manner.

Throughout, $\mathbb{R}$, $\mathbb{R}^n$ and $\mathbb{R}^{m\times n}$ denote the field of real numbers, the vector space of n-tuples of real numbers and the space of $m \times n$ matrices with entries in $\mathbb{R}$ respectively. A^T denotes the transpose of a matrix A in $\mathbb{R}^{m\times n}$ and $A \in \mathbb{R}^{n\times n}$ is said to be symmetric if $A = A^T$.

For finite sets S, T, $S \times T$ denotes the usual Cartesian product of S and T, while $|S|$ denotes the cardinality of S.

Directed and Undirected Graphs

The concept of a *graph* is fundamental to the material to be discussed in this paper. The graphs or networks which we shall encounter can be divided into two broad classes: *directed graphs* and *undirected graphs*, as illustrated in Figure 1.

Figure 1: An example of a directed graph (left) and an undirected graph (right), comprising two nodes and one edge.

Formally, a finite *directed graph*, G, consists of a set of *vertices* or *nodes*, $\mathcal{V}(G)$,

$$\mathcal{V}(G) = \{v_1, \ldots, v_n\},$$

together with an *edge* set, $\mathcal{E}(G) \subseteq \mathcal{V}(G) \times \mathcal{V}(G)$. Intuitively, each edge $(u, v) \in \mathcal{E}(G)$ can be thought of as connecting the starting node u to the terminal node v. For notational convenience, we shall often write uv for the edge (u, v). We shall say that the edge uv *starts* at u and *terminates* at v. For the

most part, we shall be dealing with graphs with finitely many vertices and for this reason, we shall often omit the adjective finite where this is clear from context.

In Biology, transcriptional regulatory networks and metabolic networks would usually be modelled as directed graphs. For instance, in a transcriptional regulatory network, nodes would represent genes with edges denoting the interactions between them. This would be a directed graph because, if gene A regulates gene B, then there is a natural direction associated with the edge between the corresponding nodes, starting at A and finishing at B. Directed graphs also arise in the study of neuronal networks, in which the nodes represent individual neurons and the edges represent synaptic connections between neurons.

An *undirected graph*, G, also consists of a vertex set, $\mathcal{V}(G)$, and an edge set $\mathcal{E}(G)$. However, there is no direction associated with the edges in this case. Hence, the elements of $\mathcal{E}(G)$ are simply two-element subsets of $\mathcal{V}(G)$, rather than ordered pairs as above. As with directed graphs, we shall use the notation uv (or vu as direction is unimportant) to denote the edge $\{u, v\}$ in an undirected graph. For two vertices, u, v of an undirected graph, uv is an edge if and only if vu is also an edge. We are not dealing with multi-graphs [47], so there can be at most one edge between any pair of vertices in an undirected graph. The number of vertices n in a directed or undirected graph is the *size* or *order* of the graph.

In recent years, much attention has been focussed on the protein-protein interaction networks of various simple organisms [92, 151]. These networks describe the direct physical interactions between the proteins in an organism's proteome and there is no direction associated with the interactions in such networks. Hence, PPI networks are typically modelled as undirected graphs, in which nodes represent proteins and edges represent interactions.

An edge, uv in a directed or undirected graph G is said to be *an edge at* the vertices u and v, and the two vertices are said to be *adjacent* to each other. In this case, we also say that u and v are *neighbours* . For an undirected graph, G and a vertex, $u \in \mathcal{V}(G)$, the set of all neighbours of u is denoted $\mathcal{N}(u)$ and given by

$$\mathcal{N}(u) = \{v \in \mathcal{V}(G) : uv \in \mathcal{E}(G)\}.$$

Node-degree and the Adjacency Matrix

For an undirected graph G, we shall write $\deg(u)$ for the degree of a node u in $\mathcal{V}(G)$. This is simply the total number of edges at u. For the graphs we shall consider, this is equal to the number of neighbours of u,

$$\deg(u) = |\mathcal{N}(u)|.$$

In a directed graph G, the *in-degree* , $\deg_{\text{in}}(u)$ (*out-degree* , $\deg_{\text{out}}(u)$) of a vertex u is given by the number of edges that terminate (start) at u.

Suppose that the vertices of a graph (directed or undirected) G are ordered as $v_1, \ldots, v_n$. Then the adjacency matrix, A, of G is given by

$$a_{ij} = \begin{cases} 1 & \text{if } v_i v_j \in \mathcal{E}(G) \\ 0 & \text{if } v_i v_j \notin \mathcal{E}(G) \end{cases} \tag{1}$$

Thus, the adjacency matrix of an undirected graph is symmetric while this need not be the case for a directed graph. Figure 2 illustrates this.

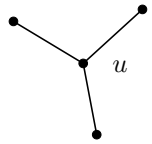
Paths, Path Length and Diameter

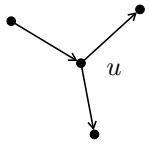
Let u, v be two vertices in a graph G. Then a sequence of vertices

$$u = v_1, v_2, \ldots, v_k = v,$$

such that for $i = 1, \ldots, k-1$:

$$A = \begin{bmatrix} 0 & 1 & 1 & 1 \\ 1 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 \end{bmatrix}$$





$$A = \begin{bmatrix} 0 & 1 & 1 & 0 \\ 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 \end{bmatrix}$$

Figure 2: The adjacency matrix of an undirected graph is symmetric; that of a directed graph generally is not. In this example, we have that $\deg(u) = 3$ for the undirected graph and $\deg_{\text{in}}(u) = 1$, $\deg_{\text{out}}(u) = 2$ for the directed graph.

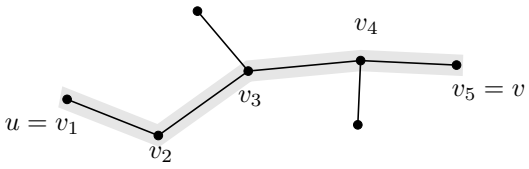


Figure 3: A path of length 4.

(i) $v_i v_{i+1} \in \mathcal{E}(G)$;

(ii) $v_i \neq v_j$ for $i \neq j$

is said to be a path of length $k-1$ from u to v. Figure 3 contains an example of a path of length 4.

The *geodesic distance*, or simply distance, $\delta(u, v)$, from u to v is the length of the shortest path from u to v in G. If no such path exists, then we set $\delta(u, v) = \infty$. If for every pair of vertices, $u, v \in \mathcal{V}(G)$, there is some path from u to v, then we say that G is *connected*. The average path length and diameter of a graph G are defined to be the average and maximum value of $\delta(u, v)$ taken over all pairs of distinct nodes, u, v in $\mathcal{V}(G)$ which are connected by at least one path.

Clustering Coefficient

Suppose u is a node of degree k in an undirected graph G and that there are e edges between the k neighbours of u in G. Then the clustering coefficient of u in G is given by

$$C_u = \frac{2e}{k(k-1)}. \tag{2}$$

Thus, C_u measures the ratio of the number of edges between the neighbours of u to the total possible number of such edges, which is $k(k-1)/2$. The average clustering coefficient of a graph G is defined in the obvious manner.

Statistical Notations

Throughout the paper, we shall often be interested in average values of various quantities where the average is taken over all of the nodes in a given network of graph. For some quantity, f, associated with a vertex, v, the notation $\langle f \rangle$ denotes the average value of f over all nodes in the graph.

3 Identification and Modelling of Bio-molecular Networks

As mentioned in Section 1, this review paper naturally splits into two parts. The first part consists of the current section and the following two sections and is primarily focussed on the structural properties of bio-molecular networks and on techniques that have been developed for their analysis.

Due to recent advances in high-throughput technologies for biological measurement, there is now more data available on bio-molecular networks than ever before. This has made it possible to study such networks on a scale which would have been impossible two decades ago. In fact, large-scale maps of protein interaction networks [197, 125, 186, 67, 151, 117], metabolic networks [97, 140] and transcriptional regulatory networks [114, 177] have been constructed for a number of simple organisms. Motivated by these developments, there has been a significant amount of work done on identifying and interpreting the key structural properties of these networks in recent years. In the current section, we shall give an overview of the main aspects of this work. In particular, we shall describe the principal graph theoretical properties of bio-molecular networks which have been observed in experimental data. We shall also discuss several mathematical models that have been proposed to account for the observed topological properties of these networks.

3.1 Structural Properties of Biological Networks

In this subsection, we shall concentrate on the following three aspects of network structure, which have received most attention in the last few years:

(i) Degree distributions;

(ii) Characteristic path lengths;

(iii) Modular structure and local clustering properties.

For each of these, we shall describe recently reported findings for protein interaction, metabolic and transcriptional regulatory networks in a variety of organisms.

Degree Distributions

Much of the recent research on the structure of bio-molecular and other real networks has focussed on determining the form of their degree distributions, $P(k), k = 0, 1, \ldots$, which measures the proportion of nodes in the network having degree k. Formally,

$$P(k) = \frac{n_k}{n},$$

where n_k is the number of nodes in the network of degree k and n is the size of the network. It was reported in [59, 12] that the degree distributions of the Internet and the WWW are described by a broad-tailed power law of the form[1],

$$P(k) \sim k^{-\gamma}, \qquad \gamma > 1 \tag{3}$$

Networks with degree distributions of this form are now commonly referred to as *scale-free networks*. This finding initially surprised the authors of these papers as they had expected to find that the degree distributions were Poisson or Gaussian. In particular, they has expected that the degrees of most nodes would be close to the mean degree, $\langle k \rangle$, of the network, and that $P(k)$ would decay exponentially as $|k - \langle k \rangle|$ increased. For such networks, the mean degree can be thought of as typical for the overall network. On the other hand, the node-degrees in networks with broad-tailed distributions

[1] In fact, the form $P(k) \sim (k + k_0)^{-\gamma} e^{-k/k_c}$ with offset k_0 and an exponential cutoff k_c is more usually fitted to real network data.

vary substantially from their mean value, and $\langle k \rangle$ cannot be thought of as a typical value for the network in this case.

Following on from the above findings on the WWW and the Internet, several authors have investigated the form of the degree distributions, $P(k)$, for various biological networks. Recently, several papers have been published that claim that interaction networks in a variety of organisms are also scale-free. For instance, in [97], the degree distributions of the central metabolic networks of 43 different organisms were investigated using data from the WIT database [140]. The results of this paper indicate that, for all 43 networks studied, the distributions of in-degree, $P_{in}(k)$, and out-degree, $P_{out}(k)$, have tails of the form (3), with $2 < \gamma < 3$.

Similar studies on the degree distributions of protein interaction networks in various organisms have also been carried out. In [200], the protein interaction network of *S. cerevisiae* was analysed using data from four different sources. As is often the case with data of this nature, there was little overlap between the interactions identified in the different sets of data. However, in all four cases, the degree distribution appeared to be broad-tailed and to be best described by some form of modified power law. Similar findings have also been reported for the protein interaction networks of *E. coli, D. melanogaster, C. elegans* and *H. pylori* in the recent paper [70]. Note however that for transcriptional regulatory networks, while the outgoing degree distribution again appears to follow a power law, the incoming degree distribution is better approximated by an exponential rule of the form $P_{in}(k) \sim e^{-\beta k}$ [13, 74, 60].

At this point, it is important to record some remarks on the observations of scale-free topologies in biological interaction networks. First of all, the broad-tailed degree distributions observed in these networks is not consistent with the traditional random graph models which have been used to describe complex networks [21, 2]. In these models, node-degrees are closely clustered around the mean degree, $\langle k \rangle$, and the probability of a node having degree k decreases exponentially with $|k - \langle k \rangle|$. However, in scale-free networks, while most nodes have relatively low degree, there are significant numbers of nodes with unusually high degree - far higher than the mean degree of the network. Such nodes are now usually referred to as *hubs.* It has been noted [4] that the scale-free structure has implications for the robustness and vulnerability of networks to failure and attack. Specifically, while removing most of the nodes in a scale-free network will have little effect on the network's connectivity, the targeted removal of hub nodes can disconnect the network relatively easily. This has led to the suggestion that genes or proteins which are involved in a large number of interactions, corresponding to hub nodes, may be more important for an organism's survival than those of low degree. The connection between network topology and the biological importance of genes and proteins has been extensively studied recently and we shall describe this strand of research in detail in Section 4.

A second important point is that all of the analysis described above has been carried out on *sampled subnetworks* rather than on a complete network. For instance, the protein interaction networks which have been studied usually contain only a fraction of the complete set of proteins of an organism. Moreover, the interactions included in these networks are far from complete. Thus, the conclusion being drawn are based on a subnetwork containing only a sample of the nodes and edges of the complete network. While some studies have indicated that the statistical properties of interaction networks may be robust with respect to variations from one data set to another, the impact of sampling and inaccurate/incomplete information on the identified degree distributions is an important issue which is not yet fully understood. For instance, in [176] it was shown using a model of protein interaction networks that an approximate power law distribution can be observed in a sampled sub-network while the degree distribution of the overall network is quite different. Further evidence of the need for caution in drawing conclusions about the overall structure of biological networks based on samples has been provided in [38, 175], where results on the sampling properties of various types of network models were presented. For instance, in [38], a sampling regime based on the construction of *spanning trees* [47] was studied. Here, starting from a source vertex v_0, a tree T is constructed by first adding the neighbours of v_0 to T and then selecting one of these, and repeating the process. In this paper, approximate arguments were presented to show that such a sampling regime can lead to a subnetwork

with degree distribution of the form $P(k) \sim 1/k$ even when the complete network has a Poisson degree distribution.

Diameter and Characteristic Path Length

Several recent studies have revealed that the average path lengths and diameters of bio-molecular networks are "small" in comparison to network size. Specifically, if the size of a network is n, the average path length and diameter are of the same order of magnitude as $\log(n)$ or even smaller. This property has been previously noted for a variety of other technological and social networks [2], and is often referred to as the *small world property* [192]. This phenomenon has now been observed in metabolic, genetic and protein interaction networks. For instance, in [189, 97], the average path lengths of metabolic networks were studied. The networks analysed in these papers had average path lengths between 3 and 5 while the network sizes varied from 200-500. Similar findings have been reported for genetic networks in [177], where a network of approximately 1000 genes and 4000 interactions was found to have a characteristic path length of 3.3, and for protein interaction networks in [187, 201, 200].

In a sense, the average path length in a network is an indicator of how readily "information" can be transmitted through it. Thus, the small world property observed in biological networks suggests that such networks are efficient in the transfer of biological information: only a small number of intermediate reactions are necessary for any one protein/gene/metabolite to influence the characteristics or behaviour of another.

Clustering and Modularity

The final aspect of network structure which we shall discuss here is concerned with how densely clustered the edges in a network are. In a highly clustered network, the neighbours of a given node are very likely to be themselves linked by an edge. Typically, the first step in studying the clustering and modular properties of a network is to calculate its average clustering coefficient, C, and the related function, $C(k)$, which gives the average clustering coefficient of nodes of degree k in the network. As we shall see below, the form of this function can give insights into the global network structure.

In [152], the average clustering coefficient was calculated for the metabolic networks of 43 organisms and, in each case, compared to the clustering coefficient of a random network with the same underlying degree distribution. In fact, the comparison was with the Barabasi-Albert (BA) model of scale-free networks which we shall discuss in the next subsection. In each case, the clustering coefficient of the metabolic network was at least an order of magnitude higher than that of the corresponding BA network. Moreover, the function $C(k)$ appeared to take the form $C(k) \sim k^{-1}$. Thus, as the degree of a node increases, its clustering coefficient decreases. This suggests that the neighborhoods of low-degree nodes are densely clustered while those of hub nodes are quite sparsely connected. In order to account for this, the authors of [152] suggested a hierarchical modular structure for metabolic networks in which:

(i) Individual modules are comprised of densely clustered nodes of relatively low degree;

(ii) Different modules are linked by hub nodes of high degree.

Similar results for the clustering coefficient and the form of the function $C(k)$ have been reported in [70] for the protein interaction networks of *S. cerevisiae, H. pylori, E. coli* and *C. elegans*, indicating that these undirected networks may also have a modular structure, in which hub nodes act as links or bridges between different modules within the networks. Further evidence for the intermediary role of hub nodes was provided in [122] where correlations between the degrees of neighbouring nodes in the protein interaction network and the transcriptional regulatory network of *S. cerevisiae* were investigated. The authors of this paper found clear evidence of such correlation; in fact, for both networks, nodes of high degree are significantly more likely to connect to nodes of low degree than to other "hubs". This property of a network is referred to as *disassortativity.* For more discussion on

this topic, see [40]. Finally, we note that a high degree of local clustering has also been observed in the transcriptional regulatory network of *S. cerevisiae* in [177].

3.2 Mathematical Models for Interaction Networks

Given the empirically observed properties of interaction networks discussed above, it is natural to ask whether these can be explained by means of mathematical models based on plausible biological assumptions. Furthermore:

(i) Reliable models for the evolution of interaction networks may deepen our understanding of the biological processes behind their evolution.

(ii) Such models could be used to assess the reliability of experimental results on network structure and to assist in experimental design. For instance, the strategy for optimally identifying protein-protein interaction (PPI) network structure described in [112] relies on the statistical abundance of nodes of high degree in scale-free networks which we shall discuss in more detail below. Furthermore, this strategy was suggested as a means of determining the PPI network in humans. Note also the work described in [71] on assessing the reliability of network data and predicting the existence of links in a PPI network which have not yet been determined. The methods in this paper were based on properties of the *small-world* network model of Watts and Strogatz introduced in [192] to described social and neurological networks.

To date, several different mathematical models of complex networks have been proposed in the literature. A number of these were not developed with specifically biological networks in mind, but rather to account for some of the topological features observed in real networks in Biology and elsewhere. On the other hand, in the recent past several models for protein interaction and genetic networks have been proposed based on biological assumptions. In this subsection, we shall describe the main models that have been used to model biological networks, and some theoretical results on the structure of these models.

Classical Models and Scale-free Graphs

In the 1950's, Paul Erdös and Alfred Renyi introduced their now classical notion of a *random graph* to model non-regular complex networks. The basic idea of the Erdös-Renyi (ER) random graph model is the following. Let a set of n nodes, $\{v_1, \ldots, v_n\}$, and a real number p with $0 \leq p \leq 1$ be given. Then for each pair of nodes, v_i, v_j, an edge is placed between v_i and v_j with probability p. Effectively, this defines a probability space where the individual elements are particular graphs on $\{v_1, \ldots, v_n\}$ and the probability of a given graph with m edges occurring is $p^m(1-p)^{n-m}$. For background on the mathematical theory of ER graphs, consult [21, 47].

The theory of random graphs has been a highly active field of mathematics for fifty years and many deep theorems about the properties of ER graphs have been established. For example, it has been proven for these networks that the characteristic path length is proportional to the logarithm of the network size, and that the average clustering coefficient is inversely proportional to network size. Perhaps the most relevant fact about the ER model in our context is the relatively straightforward result that the degree distribution is binomial. Thus, the degree distribution of a large ER network can be approximated by a Poisson distribution. The tails of such distributions are typically narrow, meaning that, for ER graphs, the node degrees tend to be tightly clustered around the mean degree $\langle k \rangle$.

This last fact contrasts with the findings reported in the previous subsection that the degree distributions of many biological networks appear to follow a broad-tailed power law. The same observation has also been made for several man-made networks including the WWW and the Internet. This behaviour is inconsistent with the classical ER model of random graphs and led Barabasi, Albert and co-workers to devise a new model for the dynamics of network evolution. This model is based on

the two fundamental mechanisms of *growth* and *preferential attachment*, and has been the subject of intensive research in the last few years. It is usually referred to as the Barabasi-Albert (BA) model.

The core idea of Barabasi and Albert was to consider a network as an evolving entity and to model the dynamics of network growth. The simple BA model is now well known and is usually described in the following manner [2]. Given a positive integer, m and an initial network, G_0, the network evolves according to the following rules (note that this is a discrete-time process):

(i) *Growth:* At each time j, a new node of degree m is added to the network;

(ii) *Preferential Attachment:* For each node u in the existing network, the probability that the new node connects to it is proportional to the degree of u. Formally, writing G_j for the network at time j and $P(u, j)$ for the probability that the new node added at time k is linked to u in G_{j-1}:

$$P(u, j) = \frac{\deg(u)}{\sum_{v \in \mathcal{V}(G_{j-1})} \deg(v)}. \tag{4}$$

Using computer simulations and approximate arguments based on "mean field theory" it has been argued that the above scheme generates a network whose degree distribution asymptotically approaches the power law $P(k) \sim k^{-\gamma}$ with $\gamma = 3$ [2]. A number of variations on the basic BA model have also been proposed that have power law degree distributions with values of the degree exponent other than three. See for instance, the models for evolving networks described in [50, 106] which give rise to power law degree distributions with exponents in the range $2 < \gamma < +\infty$.

Some Issues in the Use of Scale-free Models

While the degree distributions of BA and related scale-free models appear to fit the experimental data on bio-molecular networks more accurately than classical ER networks, there are several issues related to their use that should be noted. In [25], it was pointed out that the commonly used definition of BA graphs is ambiguous. For instance, the question of how to initiate the process of network evolution is not explicitly dealt with in the original papers; how do we connect the new node to the existing nodes with probability proportional to their degrees if all such nodes have degree zero to begin with? This issue can be circumvented by beginning with a network which has no isolated nodes. However, this immediately raises the difficult question of how the choice of initial network influences the properties of the growing network. These issues have been discussed in detail in [25, 22] where more mathematically rigorous formulations of the preferential attachment mechanism for network growth have been presented. A number of formal results concerning degree distributions, network diameter, robustness to node removal and other network properties have also been presented in [23, 24].

There has been a remarkable level of interest in the scale-free family of random graphs in recent years and numerous papers claiming that biological interaction networks follow a power law and belong to this class have been published. However, it is important to note that a number of reservations about the use of the BA and related models in Biology have been raised recently [62].

(i) Firstly, the BA model is not based on specific biological considerations. Rather, it is a mathematical model for the dynamical growth of networks that replicates the degree distributions, and some other properties, observed in studies of the WWW and other networks. In particular, it should be kept in mind that the degree distribution is just one property of a network and that networks with the same degree distribution can differ substantially in other important structural aspects [185].

(ii) Many of the results on BA and related networks have only been empirically established through simulation, and a fully rigorous understanding of their properties is still lacking. A number of authors have started to address this issue in the recent past but this work is still in an early stage. Also, as noted above, the definition of BA graphs frequently given in the literature is ambiguous [25].

(iii) Most significantly, from a practical point of view, the observations of scale-free and power law behaviour in biological networks are based on partial and inaccurate data. The techniques used to identify protein interactions and transcriptional regulation are prone to high levels of false positive and false negative errors [186]. Moreover, the networks being studied typically only contain a fraction of the genes or proteins in an organism. Thus, we are in effect drawing conclusions about the topology of an entire network based on a *sample* of its nodes, and a noisy sample at that. In order to do this reliably, a thorough understanding of the effect of sampling on network statistics, such as distributions of node degrees and clustering coefficients, is required. Some authors have recently started to address this issue and the following two results, presented in [38, 175], are extremely relevant in the present context:

(a) Subnetworks sampled from a scale free network are not in general scale free;

(b) It is possible for a sampled subnetwork of a network with Poisson degree distribution (which is certainly not scale-free) to appear to be scale-free.

Further results of a similar nature have recently been reported in [76]. Here, the sampling process in the large-scale yeast-2-hybrid (Y2H) experiments which have generated many of the existing protein-interaction maps for yeast was simulated on four different types of network models. The degree distributions of the models considered were normal, exponential, scale-free and truncated normal respectively. Based on the findings reported in this paper, the authors argued that, given the coverage of the yeast interactome currently available, none of the four models considered could be definitively ruled out as a model of the complete yeast interaction network! These facts cast doubt on the hypothesis that the complete PPI networks of living organisms are in fact scale free. At the very least, it demonstrates the need to be careful about the effects of sampling and data noise when we attempt to draw conclusions about the structure of biological and other real world networks.

Before moving on to discuss a number of more biologically motivated models for interaction networks, we note the recent paper [148] in which *geometric random graphs* [145] were suggested as an alternative model for protein interaction networks. This suggestion was based on comparing the frequency of small subgraphs in real networks to their frequency in various network models, including geometric graphs. However, as with BA models, there is no clear biological motivation for choosing geometric graphs to model protein interaction networks and, furthermore, the comparisons presented in [148] are based on a very small number of sample random networks. On the other hand, the authors of this paper make the important point that the accuracy of network models is crucial if we are to use these to assess the reliability of experimental data or in the design of experiments for determining network structure.

Duplication and Divergence Models

Many of the recent models for network evolution are founded on some variation of the basic mechanisms of growth and preferential attachment. However, there are other, more biologically motivated models which have been developed specifically for protein interaction and genetic regulatory networks. As with the models discussed above, these are usually based on two fundamental processes: *duplication* and *divergence*. The hypothesis underpinning these so-called Duplication-Divergence (DD) models is that gene and protein networks evolve through the occasional copying of individual genes/proteins, followed by subsequent mutations. Over a long period of time, these processes combine to produce networks consisting of genes and proteins, some of which, while distinct, will have closely related properties due to common ancestry.

To illustrate the main idea behind DD models, we shall give a brief description of the model for protein interaction networks suggested in [181]. Given some initial network G_0, the network is updated at each time t according to the rules:

(i) *Duplication:* A node v is chosen from the network G_{t-1} at random and a new node v' - a duplicate of v - is added to the network and connected to all of the neighbours of v;

(ii) *Divergence:* For each neighbour, w, of v', the edge $v'w$ is removed with probability q.

As pointed out in [181], the above scheme effectively introduces a preferential attachment mechanism into the network and generates a power law degree distribution. A number of basic properties of the model and its suitability to model the PPI network of S. cerevisae are discussed in this paper also. The same basic model has also been studied more analytically in [37]. In this paper, it was shown that if $q < 1/2$, then the degree distribution of the DD network is given by a power law whose exponent γ satisfies $\gamma < 2$. The authors of this paper also considered some closely related models for the growth of gene networks in the earlier paper [18]. Here it was pointed out that duplication alone will not give rise to a power law degree distribution.

The model described in [181] allowed for self-interacting proteins, where the copy v' can also form a link to the original v with some non-zero probability. However, there are several assumptions associated with the basic scheme described above whose biological validity is questionable.

(i) The new node, v', can only form links to neighbours of the original node v - this restricts the types of mutations allowed for duplicate genes;

(ii) A node can only undergo mutation or divergence at the instant when it is added to the network - this ignores the possibility of genes continuing to mutate long after the duplication event;

(iii) Nodes and edges can only be added to the network and not removed - this clearly places a significant restriction on the types of mutation and evolution possible.

Several extensions of the basic DD model have been proposed to relax some of the assumptions outlined above. For instance, point (i) above has been addressed in [170], while a model that allows for edge additions and removals at a much faster rate than gene duplications has been described and analysed in [16]. Yet another growth model (based on a preferential attachment mechanism) which allows edges to be added and deleted between nodes in the existing network, and for new nodes to be added to the network has been presented in [188]. Finally, the issue in point (iii) has been addressed in the recent paper [36] by a growth-deletion model that allows for the addition and removal of both edges and nodes.

While DD models of network growth are based on more plausible biological assumptions than the BA-type models, several of the caveats expressed above for BA networks still apply. In particular, the question of how reliably we can infer a network's structure from studying a sample of its nodes is critical, as is the impact of noisy data on identifying network structure. However, these points should not be seen as a criticism of the models themselves. Our aim is rather to highlight important issues that need to be taken into account in assessing how accurately such models reflect the biological reality. Of course, more reliable data is required for this to be possible. Finally, we should note that the theory of DD networks is still in a very early stage and many of their key mathematical properties are only partially understood.

3.3 Summarizing Comments

(i) Significant progress has been made recently on constructing maps of bio-molecular networks in simple organisms. Using the available data, the structural properties of protein-protein interaction, transcriptional regulatory and metabolic networks have been studied and preliminary results have been reported. The networks studied appear to have scale-free degree distributions, short characteristic path lengths and high clustering coefficients. The observed properties are not in agreement with those of traditional random graph models for complex networks.

(ii) Several new mathematical models for the growth of random networks have been proposed in the recent past. These include a number of variations on the basic BA scale-free model, and the more biologically inspired *Duplication-Divergence* models for gene and protein networks. The

mathematical theory of these models is only beginning to be developed and offers many exciting and challenging opportunities for future biologically motivated research.

(iii) Both in the identification of network properties, and in the development of mathematical models, the issues of inaccurate data and sampling are of paramount importance. Recent results on the sampling properties of networks with power law and Poisson degree distributions highlight the need for caution when drawing conclusions on global network properties from an analysis of a sampled subnetwork.

4 Measures of Centrality and Importance in Biological Networks

The problem of identifying the most important nodes in a large complex network is of fundamental importance in a number of application areas, including Communications, Sociology and Management. To date, several measures have been devised for ranking the nodes in a complex network and quantifying their relative importance. Many of these originated in the Sociology and Operations Research literature, where they are commonly known as *centrality measures* [191]. More recently, driven by the phenomenal growth of the World Wide Web, schemes such as the PageRank algorithm on which GOOGLE is based, have been developed for identifying the most relevant web-pages to a specific user query.

As described in the previous section, there is now a large body of data available on bio-molecular networks, and there has been considerable interest in studying the structure of these networks and relating it to biological properties in the recent past. In particular, several researchers have applied centrality measures to identify structurally important genes or proteins in interaction networks and investigated the biological significance of the genes or proteins identified in this way. Particular attention has been given to the relationship between centrality and essentiality, where a gene or protein is said to be essential for an organism if the organism cannot survive without it. The use of centrality measures to predict essentiality based on network topology has potentially significant applications to drug target identification [184, 96].

In this section, we shall describe several measures of network importance or centrality that have been applied to protein interaction and transcriptional regulatory networks in the recent past. We shall place particular emphasis on the efforts to assess the biological significance of the most central genes or proteins within these networks.

4.1 Classical Centrality Measures

In this subsection, we shall discuss four classical concepts of centrality which have recently been applied to biological interaction networks:

(i) Degree centrality;

(ii) Closeness centrality;

(iii) Betweenness centrality;

(iv) Eigenvector centrality.

Degree Centrality

Degree centrality is the most basic of the centrality measures to be discussed here. The idea behind using degree centrality as a measure of importance in network is the following:

An important node is involved in a large number of interactions.

Formally, for an undirected graph G, the degree centrality of a node $u \in \mathcal{V}(G)$ is given by

$$C_d(u) = \deg(u). \tag{5}$$

For directed networks, there are two notions of degree centrality: one based on in-degree and the other on out-degree. These are defined in the obvious manner. Degree centrality and the other measures discussed here are often normalised to lie in the interval $[0, 1]$.

As discussed in the previous section, a number of recent studies have indicated that bio-molecular networks have broad-tailed degree distributions, meaning that while most nodes in such networks have a relatively low degree, there are significant numbers of so-called hub nodes. The removal of these hub nodes has a far greater impact on the topology and connectedness of the network than the removal of nodes of low degree [4]. This naturally leads to the hypothesis that hub nodes in protein interaction networks and genetic regulatory networks may represent essential genes and proteins. In [95], the connection between degree centrality and essentiality was investigated for the protein-protein interaction network in *S. cerevisiae*. The analysis was carried out on a network consisting of 1870 nodes connected by 2240 edges, which was constructed by combining the results of earlier research presented in [178, 197]. In this network, 21% of those proteins that are involved in fewer than 5 interactions, $C_d(u) \leq 5$, were essential while, in contrast, 62% of proteins involved in more than 15 interactions, $C_d(v) \geq 15$, were essential.

More recently, similar findings were reported in [201]. Again, the authors considered a network of protein interactions in yeast, this time consisting of 23294 interactions between 4743 proteins. The average degree of an essential protein in this network was 18.7, while the average degree of a non-essential protein was only 7.4. Moreover, defining a hub to be a node in the first quartile of nodes ranked according to degree, the authors of [201] found that over 40% of hubs were essential while only 20% of all nodes in the network are essential.

The above observations have led some authors to propose that, in protein interaction networks, node degree and essentiality may be related [201, 95]. However, the precise nature of this relationship is far from straightforward. For instance, using a network constructed from data published in [92, 178], the author of [194] has claimed that there is little difference between the distributions of node degrees for essential and non-essential proteins in the interaction network of yeast. However, in this network, the degrees of essential proteins are still typically higher than those of non-essential proteins.

In [75] the connection between the degree of a protein and the rate at which it evolves was investigated. The authors reasoned that if highly connected proteins are more important to an organism's survival, they should be subject to more stringent evolutionary constraints and should evolve at a slower rate than non-essential proteins. However, the authors of [75] found no evidence of a significant correlation between the number of interactions in which a protein is involved and its evolutionary rate. Once again, this indicates that while node degree gives some indication of a protein's likelihood to be essential, the precise relationship between essentiality and node degree is not a simple one.

Closeness Centrality Measures

We shall now discuss *closeness centrality measures* which are defined in terms of the geodesic distance, $\delta(u, v)$ between nodes in a graph or network. The basic idea behind this category of measures is the following:

An important node is typically "close" to, and can communicate quickly with, the other nodes in the network.

In the recent paper [196], three closeness measures, which arise in the context of resource allocation problems, were applied to metabolic and protein interaction networks. The specific measures considered in this paper were *excentricity, status, and centroid value.*

The excentricity, $C_e(u)$, of a node u in a graph G is given by

$$C_e(u) = \max_{v \in \mathcal{V}(G)} \delta(u, v), \tag{6}$$

and the *centre* of G is then the set

$$\mathcal{C}(G) = \{v \in \mathcal{V}(G) : C_e(v) = \min_{w \in \mathcal{V}(G)} C_e(w)\}. \tag{7}$$

Thus, the nodes in $\mathcal{C}(G)$ are those that minimise the maximum distance to any other node of G.

The *status*, $C_s(u)$, of a node v is given by

$$C_s(u) = \sum_{v \in \mathcal{V}(G)} \delta(u, v), \tag{8}$$

and the *median* of G is then the set

$$\mathcal{M}(G) = \{v \in \mathcal{V}(G) : C_s(v) = \min_{w \in \mathcal{V}(G)} C_s(w)\}. \tag{9}$$

The nodes in $\mathcal{M}(G)$ minimise the *average* distance to other nodes in the network.

The final measure considered in [196] is the *centroid* value which is closely related to the status defined above. In fact, these two measures give rise to identical rankings of the nodes in a graph and, for this reason, we shall not formally define centroid value here.

A number of points about the results presented in [196] are worth noting. First of all, on both ER graphs and the BA model of scale-free graphs, all three measures were found to be strongly correlated with node-degree. The measures were then applied to the central metabolic network of *E. coli* and the centre, $\mathcal{C}(G)$, and the median, $\mathcal{M}(G)$, of this network were calculated. The authors reasoned that central nodes represent "cross-roads" or "bottlenecks" in a network and should correspond to key elements of the organism's metabolism. In support of this assertion, the centre, $\mathcal{C}(G)$, contained several of the most important known substrates, including ATP, ADP, AMP and NADP. On the other hand, in the protein interaction network of *S. cerevisiae*, no discernible difference between the excentricity distribution of essential and non-essential proteins was observed. In the same paper, centrality measures were also applied to networks of protein domains where two domains are connected by an edge if they co-occur in the same protein. The nodes with the highest centrality scores in these networks corresponded to domains involved in signal transduction and cell-cell contacts.

Betweenness Centrality Measures

In [64], the concept of *betweenness centrality* was introduced as a means of quantifying an individual's influence within a social network. The idea behind this centrality measure is the following:

An important node will lie on a high proportion of paths between other nodes in the network.

Formally, for distinct nodes, $u, v, w \in \mathcal{V}(G)$, let σ_{uv} be the total number of geodesic paths between u and v and $\sigma_{uv}(w)$ be the number of geodesic paths from u to v that pass through w. Also, for $w \in \mathcal{V}(G)$, let $V(u)$ denote the set of all ordered pairs, (u, v) in $\mathcal{V}(G) \times \mathcal{V}(G)$ such that u, v, w are all distinct. Then, the betweenness centrality of w, $C_b(w)$, is given by

$$C_b(w) = \sum_{(u,v) \in V(w)} \frac{\sigma_{uv}(w)}{\sigma_{uv}}. \tag{10}$$

Recently, in [99] the measure C_b was applied to the yeast protein interaction network and the mean value of C_b for the essential proteins in the network was approximately 80% higher than for non-essential proteins. In fact, the results in this paper indicate that the performance of C_b as an indicator

of essentiality is comparable to that of node degree. In this paper, it was also noted that there were significant numbers of proteins with high betweenness centrality scores but low node degree. The authors pointed out that this was not consistent with the scale-free BA model or with the more biologically motivated DD models proposed in [170, 181]. Furthermore, there was considerable variation in the value of $C_b(u)$ for proteins u with the same degree. This naturally raises the following question: if two proteins, u, v have the same degree k but $C_b(u) > C_b(v)$, is u more likely to be essential than v? However, no clear evidence to support this hypothesis was found in the data considered in [99].

In the present context, it is worth noting the work in [136] where a definition of betweennness centrality based on random paths between nodes, rather than on geodesic paths was considered. This centrality measure was motivated by the fact that, in real networks, information does not always flow along the shortest available path between two points. To the best of the authors' knowledge, this new concept of betweenness centrality has yet to be applied to bio-molecular networks in a systematic way.

Eigenvector Centrality Measures

As with many of the measures considered in this section, eigenvector centrality measures appear to have first arisen in the analysis of social networks, and several variations on the basic concept described here have been proposed [26, 27, 191, 28]. This family of measures is a little more complicated than those considered previously and eigenvector centrality measures are usually defined as the limits of some iterative process. The core idea behind these measures is the following.

An important node is connected to important neighbours.

In much of the original work presented in the sociology literature, the eigenvector centrality scores of a network's nodes were determined from the entries of the principal eigenvector of the network's adjacency matrix. Formally, if A is the adjacency matrix of a network G with $\mathcal{V}(G) = \{v_1, \ldots, v_n\}$, and

$$\rho(A) = \max_{\lambda \in \sigma(A)} |\lambda|$$

is the spectral radius of A, then the eigenvector centrality score, $C_{ev}(v_i)$ of the node v_i is given by the i^{th} co-ordinate, x_i, of a suitably normalised eigenvector x satisfying

$$Ax = \rho(A)x.$$

In the recent paper [57], the connection between various centrality measures, including eigenvector centrality, and essentiality within the protein interaction network of yeast was investigated. In this paper, the performance of eigenvector centrality was comparable to that of degree centrality and it appeared to perform better than either betweenness centrality or closeness centrality measures. A number of other centrality measures which we shall mention later in this section were also studied. Before concluding our discussion of the classical centrality measures and their possible application to the identification of essential genes or proteins, it is worth noting the following points about eigenvector centrality.

(i) In order for the definition above to uniquely specify a ranking of the nodes in a network it is necessary that the eigenvalue $\rho(A)$ has geometric multiplicity one. For general networks, this need not be the case. However, if the network is connected then it follows from the Perron-Frobenius Theorem for irreducible non-negative matrices [17, 86] that this will be the case.

(ii) Similar ideas to those used in the definition of eigenvector centrality have recently been applied to develop the Page-Rank algorithm on which the GOOGLE search engine relies [32, 111]. The HITS algorithm for the ranking of web pages, proposed by Kleinberg [105], also relies on similar reasoning.

Other Centrality Measures

Finally for this subsection, we briefly note several less standard centrality measures which have been developed in the last decade or so, with potential applications in the analysis of biological networks. For instance, in [57] the notion of *subgraph centrality* was introduced and the relationship between the subgraph centrality of a protein in the yeast interaction network and its likelihood to be essential was investigated. Loosely speaking, the subgraph centrality of a node measures the number of subgraphs of the overall network in which the node participates, with more weight being given to small subgraphs. Formally, if A is the adjacency matrix of a network with vertex set, $\mathcal{V}(G) = \{v_1, \ldots, v_n\}$, and we write $\mu_k(i)$ for the (i, i) entry of A^k, then the subgraph centrality of node v_i, $C_{sg}(v_i)$ is defined by

$$C_{sg}(v_i) = \sum_{k=0}^{\infty} \frac{\mu_k(i)}{k!}. \tag{11}$$

The findings presented in [57, 56] indicate that C_{sg} performs as well as node degree in predicting essentiality.

Other concepts of centrality that have been proposed include *flow betweenness centrality* [65], *information centrality* [172]. For completeness, we also note the recent measure introduced in [113] which ranks nodes according to the effect their removal has on the efficiency of a network in propagating information and the centrality measure based on game theoretic concepts defined in [72]. We shall not discuss these in detail here however as little work on their biological relevance has been done to date.

4.2 Alternative Approaches to Predicting Essentiality

We shall now briefly discuss some other methodologies for predicting gene or protein essentiality that have been proposed in the last few years.

Functional Classes and Essentiality

In the Yeast Protein Database (YPD) [43] various functional classes are defined to which the proteins in yeast can be assigned. Using the functional classification of proteins in the Yeast Protein Database (YPD) [43], the authors of [96] studied the relationship between the functions of a protein in the interaction network of yeast and its likelihood to be essential. They found that the probability of essentiality varied significantly between the 43 different functional classes considered. For instance, in one class containing proteins that are required for DNA splicing, the percentage of essential proteins was as high as 60% while only 4.9% of the proteins in the class responsible for small molecule transport were essential. This suggests that to predict essentiality, the functional classification of proteins should be taken into account. However, the fact that many proteins are as yet unclassified is a significant impediment to such an approach.

In the same paper, the nodes within each of the 43 functional classes were ranked according to their degree and, within each class, the degree of a protein was found to be a good indicator of its likelihood to be essential. Genes were also ranked using the standard deviation of their expression levels across a large number of different yeast derivatives: each derivative corresponding to one gene deletion. Some connection between the variability in the mRNA expression of a gene and its likelihood to be essential was observed. Specifically, genes whose expression levels varied little were more likely to be essential. It is hypothesised in [96] that this may be due to robustness mechanisms that maintain the expression levels of essential genes close to a constant level, while those of less important genes are subject to less stringent constraints, and hence can be more variable.

Damage in Metabolic and Protein Networks

The concept of *damage* was recently defined for metabolic networks in [115] and then later for protein interaction networks in [161]. In the first of these papers, metabolic networks were modelled as directed bi-partite graphs [47]. Such a graph has two distinct sets of nodes: one contains the metabolites while the nodes of the other set represent the reactions catalysed by the enzymes of the metabolism. Each

such enzyme, v, is assigned a score $dg(v)$, its *damage* , which characterises the topological effect of deleting v from the network. Essentially, $dg(v)$ is the number of metabolites that would no longer be produced if the enzyme v and all the reactions catalysed by it were removed from the network. The following findings about the relationship of this concept to essentiality were reported in [115].

(i) For each value of the damage, $D > 0$, let f_D be the fraction of enzymes, v with $dg(v) = D$ which are essential. An F-test indicated that there was a statistically significant correlation between D and f_D.

(ii) The set of enzymes v for which $dg(v) \geq 5$ contains 9% of all enzymes and 50% of the essential enzymes.

Based on their findings, the authors of [115] suggested that enzymes with high damage are potential drug targets. However, it should be noted that there exist several essential enzymes, v, for which $dg(v)$ is quite low and that, conversely, there are also non-essential enzymes with high damage scores.

More recently, in [161] an analogous concept for protein interaction networks was defined and applied to the yeast protein interaction network. The results of this paper indicate that any correlation between damage and essentiality is very weak. On the other hand, the authors of this paper found that if the set of nodes disconnected from the network by the removal of a protein v contains an essential protein, then there is a high probability of v itself being essential. Finally, we note another measure of importance in biological networks which was recently described in [149]. This measure was based on the notion of *bottle-necks* within networks and its relationship to essentiality was investigated in this paper.

4.3 Final Thoughts on Essentiality

Finally, we shall discuss a number of issues with the various approaches to predicting essentiality that have been described throughout this section.

(i) **Marginal Essentiality**

While our discussion has focussed on essentiality, a gene or protein may be important to an organism without necessarily being essential. For instance, some sets of non-essential genes are *synthetically lethal,* meaning that the simultaneous removal of the genes in the set kills the organism while individual deletions are non-fatal. In the paper [201], the less restrictive concept of *marginal essentiality* and its relationship to various topological measures was studied in the protein interaction network of *S. cerevisiae*. Here, proteins were classified into five groups based on their marginal essentiality: those with the lowest marginal essentiality scores being assigned to group 1, and those with the highest assigned to group 5. The authors of [201] found that the average degree and clustering coefficient of the nodes in a group increases monotonically with the group number. For instance, the average degree of those proteins assigned to Group 1 is about half of that of the proteins in Group 4. Moreover, defining a hub node to be one in the first quartile of nodes ranked according to degree, they found that less than 10% of the proteins in Group 1 are hubs while more than 35% of those in Group 5 are hubs. The percentage again increased monotonically with the group number.

(ii) **Fitness Effect and Evolutionary Rate**

In [63] it was reported that the degree of a protein in the interaction network of yeast was positively correlated with the *fitness effect* of deleting the gene that encodes the protein. Here, fitness effect measures the reduction in the growth rate of the organism when the gene is deleted. This investigation was motivated by the question of whether the importance of a gene or protein for an organism correlates with the rate at which it evolves. For more information, and varying opinions on this topic, consult [75, 98, 198, 141, 80, 79].

(iii) **Sensitivity to Data Errors**

The issue of sensitivity to data inaccuracy is of critical importance for all of the techniques described here. It was noted in [161] that the measure damage discussed above is quite sensitive to false negative errors, in which a real interaction between two nodes in a network has not been identified due to experimental error. Clearly, such sensitivity to data noise has serious implications for the practical use of any of the methods described here. In particular, it is important to have a thorough understanding of the effect of missing or inaccurate data on the performance of centrality measures or other approaches to predicting essentiality. While there has been some research into this fundamental issue recently [42, 202, 29, 163], more intensive quantitative and theoretical studies are needed before we can reliably apply the techniques discussed here to the problem of essentiality prediction. This issue is all the more important given that much of the data available on bio-molecular networks contains large numbers of false positive and false negative results [40, 186].

(iv) **Essentiality and Modules**

Finally for this section, we note the work of [46] on determining the essentiality and cellular function of modules within the yeast PPI network. The results of this paper indicate that the essentiality (or non-essentiality) and functionality of an overall complex is largely determined by a core set of proteins within the complex. Moreover, the essentiality of individual proteins appears to depend on the importance of the modules in which they lie. This suggests that it may be more appropriate to address the question of essentiality at the level of modules rather than individual proteins or genes and motivates the problem of extending centrality measures to deal with groups of nodes.

4.4 Summarizing Comments

(i) In this section, we have discussed several measures of the importance, or centrality, of the nodes in complex networks, including degree centrality, betweenness centrality, closeness centrality and eigenvector centrality. We have described the findings of several recent studies which have applied these measures to datasets on protein-protein interaction and transcriptional regulatory networks.

(ii) Most of the studies discussed in the text indicate a link between the centrality score of a gene or protein and its likelihood to be essential for survival.

(iii) There appears to be no compelling evidence at the current time that the more complex centrality measures described here perform any better as indicators of essentiality than simple degree centrality.

(iv) As with the identification of network structure discussed in Section 3, the impact of inaccurate and incomplete data on the performance of centrality measures as indicators of essentiality is of critical importance and needs to be more fully investigated.

5 Motifs and Functional Modules in Biological Networks

The analysis methods discussed in the previous section were concerned with identifying individually important nodes within a network. However, several recent studies have revealed that bio-molecular networks are often modular in nature, with groups of individual nodes collaborating to carry out some specific biological function. This has led researchers to investigate more closely the hierarchical structure of real interaction networks, and to provide biological explanations for how the observed structure of such networks has emerged.

Recently, a loose hierarchical structure for bio-molecular networks has been proposed in [10, 13]. The lowest level in this hierarchy consists of individual nodes, which are then organised into so-called *network motifs*. Motifs are small subgraphs that occur significantly more often in a network than would be expected by chance. These are in turn grouped into larger modules of functionally related nodes before finally, the modules are themselves connected to form the overall network. In this section, we shall discuss recent work on identifying motifs within specific biological networks, and the efforts of a number of researchers to use motifs to classify networks into distinct families. We shall also consider the question of why motifs occur so frequently in real networks. Towards the end of the section, we shall consider the problem of identifying communities of functionally related nodes in bio-molecular networks and discuss a number of algorithms that have been proposed for this purpose.

5.1 Identification of Network Motifs

The concept of a network motif and a basic scheme for motif detection were described in the paper [126]. Specifically, given a directed network G, the motifs in G of size k are identified as follows:

(i) For each possible subgraph, S of size k, of G count the number of occurrences, N_S, of S in G.

(ii) Next randomly generate a large number of networks such that in each random network:

  (a) Each node has the same in-degree and out-degree as in the real network G;

  (b) Every subgraph of size $k-1$ occurs with the same frequency as in the real network G. Two schemes for generating the random networks are described in [126] and its supporting material.

(iii) A subgraph, S, is then said to be a motif of G if it satisfies the following three conditions:

  (a) The probability of S occurring in a random network more often than N_S times is less than some prescribed value P (in [126] P is taken to be .01);

  (b) There are at least four distinct occurrences of S in the network G;

  (c) The actual number of occurrences of S in G is significantly larger than the average number of occurrences of S in the randomly generated networks, denoted $\langle N_S^{rand} \rangle$; formally, $N_S - \langle N_S^{rand} \rangle > 0.1 \langle N_S^{rand} \rangle$.

Comments

(i) The scheme described above can, and has been [195], easily adapted to detect motifs in undirected networks such as protein interaction networks.

(ii) The identification of motifs within large complex networks is computationally intensive and, to the best of the authors' knowledge, standard methods are only feasible for motifs containing less than 7 or 8 nodes.

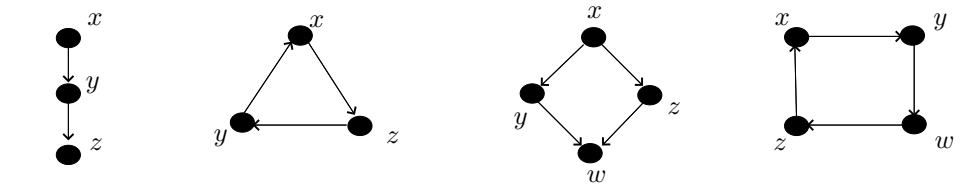
(iii) In [205] a systematic method of defining network measures or "scalars" which are related to subgraphs and can be used to detect motifs was introduced. The techniques of this paper address some of the issues with standard motif detection algorithms but the precise relationship between "scalars" and subgraphs is not straightforward.

Using the scheme described above, small motifs have been identified in a number of real biological networks. In particular, the transcriptional regulatory networks of *E. coli* and *S. cerevisiae* have been found to have one three-node motif and one four-node motif. These are the so-called *feed-forward* motif and *bi-fan* motif, shown in Figure 4 below.

Feed-forward loop Bi-Fan

Figure 4: Feed-forward and Bi-Fan motifs of transcriptional networks

The feed-forward and bi-fan patterns are also motifs of the neuronal network of the nematode *C. elegans*. This network has an additional four-node motif known as the bi-parallel motif. Other common motifs which have been detected in food webs, electronic circuits and the World-Wide-Web include the *three-chain* , *three and four-node feedback loops* and the *fully-connected triad* shown below. Note that the network motifs of the transcriptional network of yeast have also been investigated in the paper [114], where the motifs identified have also been related to specific information processing tasks.



Three-chain Three-node feedback Bi-parallel Four-node feedback

Figure 5: Common motifs in real networks

Before proceeding, a number of facts about the findings reported in [126] are worth noting. The feed-forward loop and bi-fan motifs have been found in transcriptional regulatory networks and neuronal networks, both of which involve some form of information processing. Also, the motifs found in the food-webs studied are distinct from those found in transcriptional regulatory networks and the WWW, while electronic circuits with distinct functions tend to have different sets of motifs. These observations have led some authors to suggest that there is a connection between a network's motifs and its function, and hence, that complex networks may be classified into distinct functional families based on their typical motifs. For instance, given that information processing is fundamental to both neuronal and transcriptional networks, it is reasonable to suggest that feed-forward loops and bi-fans occur often in such networks because of their suitability for information processing tasks. On the other hand, there is no overlap between the motifs observed in transcriptional networks and those of the functionally unrelated food-web networks.

Finally for this subsection, we note that the transcriptional network of *E. coli* has been investigated in more detail in [169] and several additional motifs have been identified: *single input modules (SIMs)* ; *dense overlapping regulons* and *negative autoregulatory units*.

5.2 Dynamical Properties of Motifs

In the recent past, there have been several theoretical and experimental studies carried out on the dynamical properties of specific motifs and on clarifying the relationship between motifs and functionality. For example, in [169], it was demonstrated that the feed-forward loop motif can provide a mechanism of filtering out transient or fluctuating input signals. This motif structure also responds to persistent activation with a slight delay and shuts down rapidly once the activating signal is removed. Circuits of this type are said to act as *sign-sensitive delays*. The authors of [169] presented a simple mathematical model to describe the action of the feed-forward loop in transcriptional regulatory circuits and then studied the behaviour of this model under the assumption that the circuit was *coherent* in the following sense. Each regulatory interaction is assigned a positive or negative sign depending on whether it is excitatory or inhibitory. The circuit is coherent if the indirect and direct paths have the same sign, and incoherent otherwise. Under the assumption that all regulations are excitatory, it was shown numerically in [169] that the FFL motif does indeed act as a sign-sensitive delay element.

A more complete mathematical analysis of the kinetic behaviour of the FFL motif was presented in the paper [120], where the response times of all of the different possible configurations of the FFL were studied. Note however that coherent configurations seem to occur far more frequently than incoherent configurations in real systems such as the transcriptional network of *E. coli* [121]. Also, in [77], a more detailed model of the coherent FFL circuit was described and analysed. Here, the robustness of the model's behaviour with respect to variations in parameter values and external perturbations was investigated. For instance, the sign-sensitive delay action was found to be quite sensitive to variations in the model's parameters and, while the circuit is quite robust with respect to the size of external perturbations, the duration of the perturbation in comparison to the internal time-scales of the circuit appears to be critical.

In addition to the theoretical investigations described above, the kinetics of the coherent FFL motif have been studied experimentally in [121]. Specifically, the authors of this paper analysed the *l-arabinose* utilization circuit in *E. coli* and confirmed that, in this case, the coherent FFL circuit functions as a sign-sensitive delay element that filters out transient activation signals from a fluctuating environment.

Before finishing our discussion of this topic, we should note a number of other theoretical and experimental investigations of the dynamical properties of network motifs. The negative autoregulatory circuit consisting of a transcription factor that down-regulates its own transcription was studied in [157], where the response times of a simple transcriptional unit (without autoregulation) and a negative autoregulatory circuit were compared. Here, it was shown theoretically that the response-time of the autoregulatory circuit is shorter than that of the simple transcriptional circuit, with the same steady state. In fact, for very strong auto-repression, the response-time of the auto-regulatory circuit is only one fifth of that of simple transcription. It has also been demonstrated experimentally in the same paper that while a transcriptional circuit without autoregulation has a response-time of approximately one cell-cycle, the response-time for a circuit with negative auto-regulation is about one-fifth of a cell cycle. Finally, we also note the recent work on the kinetics of the single-input module (SIM) motif in *E. coli* [169] and the p53-Mdm2 feedback loop [110].

5.3 Evolutionary Conservation, Extensions and Final Thoughts on Motifs

Motifs and Evolutionary Conservation

The work discussed in the last subsection was concerned with investigating the dynamical properties and biological function of a number of common motifs. The biological significance of motifs has been considered from a slightly different point of view in [195] where the extent to which motifs in the protein interaction network of yeast are evolutionarily conserved was studied. Specifically, 678 proteins in the

yeast PPI network were identified which have orthologs [2] in each of five higher organisms, and for each 2, 3, 4 and 5 node motif, the percentage of motifs which were completely conserved across all of the 5 higher organisms was determined. A sub-graph is completely conserved if all of the proteins in it have orthologs in each of the higher organisms. For the yeast PPI network, motifs which have a higher number of nodes and are more densely interconnected also have a higher rate of conservation. For instance, the completely connected five-node motif has the highest rate of conservation of all motifs with between 2 and 5 nodes.

To validate these findings, the same number of orthologs was positioned randomly on the network and the percentages of completely conserved motifs were again calculated. In this case, the rates of conservation were considerably lower, and moreover, the rate of conservation decreased with increasing motif size, in contrast to what was observed for the real orthologs. In particular, for the completely connected five-node motif, the natural rate of conservation was found to be 47.24% while the random conservation rate was as low as .02%. Furthermore, larger, more tightly connected and conserved motifs were found to be more functionally homogeneous. In fact, for a significant number of these, all of the proteins in the complex belonged to at least one common functional class.

Note also that in [182] a correlation between the natural rate of conservation of motifs in the yeast PPI network and the suitability of the motif structure for synchronization of interconnected Kuramoto oscillators was reported. We shall have more to say about the question of synchronization later in the article.

Extensions of the Motif Concept

In [128], the significance profile (SP) was proposed as a means of classifying networks. Given a network, G, for each possible subgraph, S, the number of occurrences of S in a real network G is calculated and compared to the average number of occurrences of S in an ensemble of random networks with the same degree profile as G. The Z-score for each such subgraph is then calculated as

$$Z_S = \frac{N_S - \langle N_S^{rand} \rangle}{std(N_S^{rand})} \tag{12}$$

where N_S, $\langle N_S^{rand} \rangle$ and $std(N_S^{rand})$ denote the number of occurrences of S in G, and the mean and standard deviation of the number of occurrences of S in the ensemble of random networks respectively. The vector of Z-scores for subgraphs of a fixed size is then normalized to give the *significance profile* vector.

$$SP_S = \frac{Z_S}{(\sum_S Z_S^2)^{1/2}}. \tag{13}$$

Significance profiles for subgraphs of sizes three and four are calculated in [128] for a number of real biological networks. While this method has been proposed as a means of identifying different classes of complex networks, it should be noted that some networks with similar SP vectors for three-node subgraphs have distinct four-node SPs. As mentioned in [128], this means that higher order SPs are needed if this technique is to be used effectively to classify networks. Also it is not clear at the moment how to determine the maximal subgraph size required to correctly distinguish network classes using this technique.

Another possible extension of the motif concept was recently suggested in [103]. Here, so-called *topological generalizations* of subgraphs and motifs were introduced based on duplicating certain nodes within the subgraph. Several significant motif generalizations within the transcriptional regulatory networks of *E. coli* and *S. cerevisiae* were identified and possible functions for the observed generalizations were also proposed and investigated on simple mathematical models of transcriptional regulation and neuronal networks. While most of our discussion has focussed on transcriptional networks or protein interaction networks in isolation, this distinction is somewhat artificial, and ultimately the methods described here will need to be extended to more integrated cellular networks. In this context, the work

[2] Orthologs are genes with a common ancestor.

of [199] on identifying motifs within a more complete cellular network, which takes into account both transcriptional interactions and direct protein-protein interactions, and the study of motifs within an integrated network involving five different interaction types in [203] should be noted.

Some Final Thoughts on Motifs

Before drawing our discussion of network motifs to a close, we note a number of motivations for the study of network motifs as well as some caveats that should be kept in mind.

(i) Studying the motifs of a complex biological network can provide useful insights into the both the structure and function of the network. For instance, once we have identified a network's motifs, analysis such as that described above on the dynamical properties of the FFL motif can help us to determine the key functional roles of the network. Motifs can also be used to help develop more complete models for the evolution of bio-molecular networks than those discussed in Section 3.

(ii) As mentioned above, motifs and extensions such as the significance profile could be used to identify distinct categories of complex networks. However, as noted in [128], networks with the same motif profile for three-node subgraphs can have different four-node or higher order motifs and this casts some doubt on how effective these methods are likely to prove as a means of classifying networks. Moreover, the identification of higher order motifs is likely to be very costly from a computational point of view.

(iii) A knowledge of the motifs of a network is a necessary step in unravelling its hierarchical structure and can help in identifying modules which can then be used to simplify the network's analysis.

(iv) The precise biological significance of the various network motifs which we have discussed is still unclear and it should be noted that while motifs are *statistically* significant subgraphs, there may be other subgraphs within a network, occurring in smaller numbers, that are biologically important. This issue has been debated in [9, 127], and in [41] two biological reasons for the emergence of motifs have been considered: gene duplication and convergent evolution. The findings described in [41] indicate that the motifs in the transcriptional regulatory networks of *S. cerevisiae* and *E. coli* have not emerged due to gene duplication, which the authors argue, provides evidence for claims that network motifs have emerged as the result of some mechanism of natural selection. Further evidence that motifs have emerged as a result of some biological optimization was recently presented in [93]. Here, the motif patterns of *geometric networks,* where links are formed based on the spatial proximity of nodes, were studied analytically. The results of this paper show that simple geometric constraints alone are not sufficient to account for the motifs observed in biological and social networks. The authors of this paper argued that this indicates that additional, possibly biological, factors have played a role in determining the emergence of motifs in such networks.

5.4 Community Structure and Functional Modules in Biological Networks

In the introductory remarks to this section, we outlined a loose hierarchical structure for biological networks, in which the next level above that of motifs was that of modules or communities of functionally related nodes. Recent research has indicated that functionally coherent families of genes and proteins can be determined from the topology of interaction networks [200]. Hence, the development of reliable methods for identifying such *functional modules* would have significant implications for the problem of assigning functions to unannotated proteins or genes. This issue is of considerable importance given that the biological function of many of the genes and proteins within even simple organisms such as yeast are still unknown. In this subsection, we shall discuss a number of algorithms and techniques that have been developed for the identification of modules and community structure within complex networks. To begin with, we shall review those methods that have been developed specifically to detect functional families and hierarchical structure in bio-molecular networks. We shall

also discuss some techniques that have been proposed specifically for the problem of protein function assignment.

Network Hierarchy and Motif Clusters

The authors of [48] studied how the FFL and bi-fan motifs in the *E. coli* transcriptional regulatory network are integrated into the overall network structure. The findings of this paper suggest a hierarchical organization of this network, where motifs are first aggregated into larger *motif clusters,* with each cluster primarily consisting of the same motif type. These clusters are then further combined into so-called *super-clusters* which form the core of the overall network. For instance, all but one of the identified feed-forward loops (FFLs) in the network were contained in six FFL clusters, and similarly, all but one of the bi-fan motifs were contained in two bi-fan clusters. Moreover, these motif clusters combined to form one large super-cluster containing all but one feed-forward loop and one bi-fan motif.

Another approach to investigating the hierarchical and modular structure of the transcriptional network of *E. coli* was described in [119]. Here, five different regulatory levels were identified, such that each node is either self-regulatory or else can only regulate nodes at lower levels. Based on this hierarchical decomposition of the network, a scheme for identifying modules of functionally related genes was described which appears to work quite well in identifying sets of genes with similar functionality. The authors of this paper also found that many of the FFL and bi-fan motifs in this network contained genes responsible for regulating modules with diverse function. They argue that this fact is not in agreement with the view that motifs themselves form the basic building blocks of functional modules, as, for instance, it shows that the same feed-forward loop can be involved in the regulation of numerous different modules.

Graph Theoretical Approaches to Identifying Functional Modules

A graph clustering algorithm for identifying families of related nodes in networks was described in [55], where the problem of how to cluster proteins in large databases into families based on sequence similarity was considered. The first step in this algorithm was to assign sequence similarity scores to each pair of proteins using an algorithm such as BLAST. A weighted graph was then constructed, whose nodes are proteins and where the weight of an edge between two nodes is the similarity score calculated in the previous step. The TRIBE-MCL algorithm for detecting communities of related nodes within this graph was then described. This technique is based on *Markov chain clustering* , and identifies communities through iterating two different mathematical operations of *inflation* and *expansion* . The core concept behind this method is that families of related nodes are densely interconnected and hence there should be more "long" paths between pairs of nodes belonging to the same family than between pairs of nodes belonging to distinct families. Subsequently, in [146] this algorithm was used to identify functionally related families in the protein interaction network of *S. cerevisiae*. In fact, the algorithm was applied to the *line-graph* $L(G)$, where the nodes of $L(G)$ are the edges of G and two nodes in $L(G)$ are connected if the corresponding edges in G are incident on a common node in G. Three separate schemes of protein function classification were then used to validate the modules identified with this algorithm, and the coherence of functional assignment within these modules was significantly higher than that obtained for random networks obtained by shuffling protein identifiers between modules. This together with further analysis indicated that the identified modules did represent functional families within the network.

Further approaches to the determination of functional modules within biological networks have been described in [149, 166]. The technique in [149] relies on searching for *highly connected subgraphs (HCS)* where a HCS of a graph G is a subgraph S for which at least half of the nodes of S must be removed in order to disconnect it. On the other hand, in [166, 165] a procedure is described which identifies modules of related genes in the transcriptional regulatory network of yeast as well as the regulators of each such module. Other approaches to determining functional modules within transcriptional networks have been described in [11, 90]. The techniques described in these papers are not based on a graph theoretical analysis of network topology however; in fact, they rely on analysing gene expression

data across different experimental conditions and determining sets of genes which are regulated by common transcription factors.

Predicting Protein or Gene Function from Network Structure

Several direct approaches to assigning functions to unannotated proteins have also been proposed recently. The simplest of these is the so-called *majority rule* which works in the following way [164, 131]. Given a classification scheme with an associated set of functions,

$$\mathcal{F} = \{f_s : 1 \leq s \leq M\},$$

an interaction network, G, and an unannotated protein i in G, each function, $f_s \in \mathcal{F}$, is assigned a score which is simply the number of times f_s occurs among the annotated neighbours of i. The functions with the highest scores are then identified as the most likely functions for the protein i. A simple extension of this concept which takes into account nodes other than the immediate neighbours of the unannotated protein was presented in [81]. It should be noted that this approach has the major drawback of relying entirely on the functions of previously annotated proteins, while it can often happen that none of the neighbours of a protein of unknown function have been annotated.

Two more sophisticated approaches to protein function prediction that avoid the above mentioned difficulty were described in [180, 102]. Essentially, these algorithms assign functions to the proteins in an interaction network so as to minimize the number of pairs of interacting proteins with different functional assignments. A key aspect of these approaches is that the optimal global assignment of protein function is not unique. In practice, a number of different optimal solutions are determined, and the frequency with which a given function f_s is assigned to a protein i is interpreted as the probability of the protein having that function.

The work presented in two other recent papers is also worth noting in the present context. Firstly, in [131], the *functional flow* algorithm was described. The core idea of this method is to consider annotated proteins within the network as reservoirs or sources of flow for the functions assigned to them. Each such function then "flows" through the network according to a specified set of rules and the amount of each function at a node when the iterations finish is used to determine the most likely functions for that node. On the other hand, the technique described in [158] is based on the hypothesis that pairs of proteins with a high number of common interaction partners are more likely to share common functions. Formally, for a pair of proteins i, j, of degrees n_1, n_2 respectively, with m common interaction partners, the probability $p(i, j, m)$ of them having m common partners if links were distributed randomly is calculated. This method was applied to the protein interaction network of *S. cerevisiae* and, of the 100 pairs of proteins with the lowest value of $p(i, j, m)$, over 95% of them consisted of proteins with similar function. The authors also described how to use these basic ideas to identify modules within an interaction network and validated the method on the yeast interaction data. A related probabilistic approach to using interaction network topology to predict protein function has also been presented in [116].

In the recent paper [167], the PRISM algorithm for identifying modules of functionally related genes based on analysing *epistatic* interactions was presented. [3] The core idea behind this algorithm is that genes belonging to one functional module should interact with genes in another module in a similar fashion. Using this algorithm, it was possible to group genes with similar functional annotation into the same module even in the absence of a direct interaction between them. Finally, we note that in [33], a technique for identifying *quasi-cliques* in protein interaction networks based on the eigenvectors of the network's adjacency matrix was described and applied to the yeast interaction network. Most of the quasi-cliques identified in this way were found to have homogeneous functional annotation in the MIPS database suggesting that this technique could be useful in assigning function to unannotated proteins.

Module Identification in General Networks

[3] Epistatic networks describe the interactions through which different genetic mutations either aggravate or buffer each other's effects on an organism.

The problem of identifying communities and cohesive modules is also of importance in a number of other application domains including the analysis of social networks, communication networks, and power networks. Before concluding this section, in the interest of completeness, we briefly note some techniques that have been developed recently for this general problem which could also be used to identify functional modules within bio-molecular networks; see [134] for an overview of recent and traditional approaches to the problem of identifying community structure in networks. In [68, 137, 150, 61] the notions of betweenness and information centrality were first extended to apply to edges rather than nodes. Then, based on the observation that edges connecting distinct communities will typically have higher betweenness and information centrality scores than edges within communities, divisive algorithms for identifying communities were described. The basic principle of these algorithms has been adapted to determine the hierarchical and modular structure of metabolic networks in [82]. More recently, algorithms based on edge-betweenness have been applied to datasets on protein interaction networks in yeast and humans in [52]. Here, the ability of these clustering algorithms to identify families of functionally related proteins was studied. The robustness of these approaches to false positive errors in the datasets was also investigated.

In contrast to the divisive approaches discussed above, the techniques proposed in [135, 39] work in an agglomerative rather than divisive fashion. Here, a function that measures the *modularity* of a proposed division of a network into communities was defined and algorithms to optimize this function appropriately were presented. A different, information-theoretic measure of modularity which applies directly to a network rather than a specific partition of the network has recently been proposed in [204]. Algorithms for splitting a network into modules were also described in the same paper and their effectiveness was tested on real and synthetic network data, with promising results. Note also the approaches based on analysis of the spectrum of the Laplacian matrix of the network described in [35, 49].

5.5 Summarizing Comments

(i) In many real biological, and technological, networks, certain small subgraphs occur far more frequently than would be expected for randomly wired networks with the same degree distribution. Such subgraphs are known as motifs.

(ii) Experimental observations have indicated that networks with similar function tend to have similar sets of motifs. For instance, feed-forward loops are very common in both neuronal and transcriptional regulatory networks; both of which are involved in the processing of biological information. This has led researchers to consider a network's motifs as being characteristic of the network in some sense.

(iii) While the precise biological significance of motifs is still not completely understood, several recent studies on the dynamical properties of simple motifs have provided some insights into this question. In particular, the dynamics of the FFL motif and the auto-regulatory motif in transcriptional networks have been studied and linked to biological function.

(iv) Graph theoretical techniques have been used to determine the role of proteins or genes whose function is currently unknown. Several such techniques have been described in the text. General algorithms for identifying communities in complex networks can also be applied to protein interaction networks to identify modules of functionally homogeneous proteins.

6 Synchronization

So far, our discussion of complex biological networks has largely focussed on their *structural* properties. We have discussed some of the key topological parameters for bio-molecular networks, as well as numerical techniques for identifying the most important nodes within such networks and for elucidating

their hierarchical structure. As a next step, we shall consider some aspects of network *function* and network *dynamics*, with particular emphasis on the relationship between dynamic behaviour and network topology. Specifically, in this section, we shall review the results of recent work on the connection between synchronizability—that is, fitness for synchronization— and network properties such as characteristic motifs, average node-degree, betweenness centrality and degree distribution.

The outline of the section is as follows. We start off with a general introduction to synchronization phenomena. We then consider some aspects of mathematical modelling, focusing on the Kuramoto model of coupled oscillators, and discuss some common measures of synchrony. Following this, we review results on synchronizability. Lastly, we discuss the role of synchronization in the brain with particular emphasis on the connection between abnormal synchrony and neurological disorders. We close with some summarizing comments.

6.1 Biological Oscillators and Synchrony

Consider a graph wherein each node represents a dynamical process and each edge an interaction between two processes. This simple construct makes for an elegant description of many biological systems. Consider, for example, the group of pacemaker cells that make up the Sinoatrial Node [69] in the heart. For this system we can construct a graph, such that each node represents a pacemaker cell and each edge an interaction between two such cells. The topology of this graph determines, to a large degree, the system's overall behaviour. Indeed, abnormalities in the way these cells are wired up can have a more significant impact on the functioning of the Node than defects in individual cells. To appreciate how important these interactions are, consider that in isolation, each pacemaker cell oscillates at its own distinct frequency; yet when put together, these same cells coordinate their action in such a way as to generate a single impulse exactly once during each cardiac cycle [104]. This is an instance of a phenomenon known as synchronization, or more precisely, frequency synchronization. Roughly speaking, synchronization is the process through which the output of a system aligns itself with that of another system or group of systems. A special case of this is the synchronization of oscillators, to which we shall confine ourselves in this survey.

Oscillators can lock to both internal and external stimuli. The locking to external stimuli is generally called *entrainment*. Examples of entrainment are commonplace in human physiology. Many of the fundamental rhythms in our body, for instance, are entrained by the light-dark cycle [69]. More specifically, neural circuits have been found to support entrainment, particularly in the gamma frequency range (50-100 Hz) [139].

Synchronization is a population effect in the sense that it emerges in complex systems comprising a large number of identical or nearly identical components. In the natural world, synchronization manifests itself across many different levels of organization, from groups of organisms (the synchronous flashing of fireflies [34]) down to groups of cells (the pacemaker cells in the example of the Sinoatrial Node). Less well known, perhaps, is its implication in discussions on the binding problem, one of the central problems in the philosophy of mind. Specifically, in this context, synchronization has been put forward as a mechanism to explain how information, distributed across the brain, might be integrated to make coherent perception possible [54]. Given the variety of applications, the importance of understanding the principles of synchronization is clear. One way to gain such understanding is to try to reproduce this phenomenon *in silico*, using a simple mathematical model of coupled oscillators. In the next subsection we review some aspects of the Kuramoto model, which has been the principal model used for the study of synchronization phenomena over the past thirty years.

6.2 A Model of Synchronization

Over the years, a great deal of interest has been expressed in the physics and mathematics of synchronization. One of the first to present a detailed mathematical treatment of the subject was Arthur

Winfree. His 1967 paper [193] laid the basis for the work of Kuramoto and others, who helped develop it into a mature mathematical theory with applications in different fields [174].

In this review we shall focus on a model of synchronization, introduced and popularized by Kuramoto [107, 108, 1]:

$$\dot{\theta}_i = \omega_i + \frac{K}{N} \sum_{j=1}^{N} \sin(\theta_i - \theta_j) \tag{14}$$

Here θ_i and ω_i respectively denote the phase and intrinsic frequency of oscillator i; K is the coupling strength, and N is the number of oscillators. This setting assumes undirected all-to-all coupling, meaning that the underlying graph is complete [47]

Kuramoto's model describes a mechanism of self-organization in a population of coupled oscillators. The model is thought to reflect some aspects of coordinated behaviour in natural systems. Studies indicate that the emergence of synchronization in the model is robust with respect to variations in the interconnection structure, albeit that the transition dynamics generally depend upon the details of the underlying topology. We shall discuss this dependence in more detail shortly.

A qualitative description of the behaviour of the system (14) is as follows (see [173]). When the interactions are weak, i.e. K is small, the system is in an incoherent state, in which the distribution of the phases $\{\theta_i\}$ is roughly uniform. In this state, each oscillator tends to oscillate at its own intrinsic frequency, ω_i. When the level of interaction is gradually increased, clusters of oscillators will emerge, oscillating at a common frequency and (sometimes) phase. When the coupling is still further increased, more and more oscillators will join in, leading eventually to a state of full synchronization in which all oscillators are oscillating as one. Note that, strictly speaking, full synchronization is only possible when all the oscillators are identical, i.e. when $\omega_i = \omega_j$ for all i, j. The transition from a completely incoherent to a completely coherent state is typically steep, and associated with it is some critical coupling strength K_c, which marks the start of this transition.

The analysis of the Kuramoto model has a long and rich history, and while a full understanding of its dynamics is still lacking, a number of important results have been obtained in recent years. Among them a proof of the instability of the unsynchronized state for large coupling strengths, and formulae for the critical coupling, and the steady state coherence. Most of these results are only strictly valid in the thermodynamic limit when $N \to \infty$, though some results are available for large but finite populations [94]. For an extensive review of results related to the Kuramoto model and its applications, the reader may consult [1].

The Kuramoto model has found application in many areas, including neuroscience, physics, engineering and biology [147, 1]. This wide applicability appears to be both a strength and a weakness in the sense that, as the model captures the essence of synchronization, it necessarily lacks the specificity to fully describe any one phenomenon in particular. We shall come back to this when we discuss the role of synchronization in the brain.

6.3 Types and Measures of Synchrony

In a system of coupled oscillators such as (14), the emergence of synchronization is easy to detect and quantify. In experiment, this is not quite as easy. The fundamental problem is to extract from the complex time series that are your data information about phase and frequency. This is a non-trivial problem as the underlying processes are typically non-stationary and, in a strict sense, non-periodic.

Before trying to detect synchronization proper, there are a few other things one can do. For instance, to test for statistical dependence between two time series, one could compute the spectral covariance or *coherence* [100]. In [156, 168] this technique was used to quantify task-specific interactions in the brain. In recent years, it has been suggested that this measure would lack the sensitivity required to

detect subtler forms of synchrony, such as phase synchrony, as it would not separate out effects of amplitude and phase.

Other measures of synchrony include phase coherence [171, 109], entropy, and mutual information [87, 91]. These latter measures are particularly popular among experimentalists, who seek to establish, for instance, whether or not a particular phase relationship exists between a given set of experimental variables. The application of these measures is limited by the fact that, in a typical experiment, phase information is not directly accessible, but needs to be extracted from the recorded time series using specialized algorithms. This is a nontrivial problem as the time series (e.g. EEG recordings) are generally non-periodic, and hence standard notions of phase do not apply. Fortunately, there exist alternative notions of phase that do generalize to non-periodic signals. Based on these notions, computational techniques have been developed that are capable of extracting phase information from arbitrary time series [147]. These techniques have been successfully applied to the analysis of brain data [109, 87, 171], revealing interesting patterns of synchrony.

Another factor that might complicate the application of these measures in practice, is the lacks of statistics. If prior information about the data were available one could use that to specify what degree of coherence should be considered statistically significant. But in an experimental setting, such information is typically not available. One way to overcome this problem is to use schemes which generate ensembles of surrogate data that are in some sense statistically similar to the original time series [87]. An early example of an application of this approach can be found in [109].

For the system of coupled oscillators (14), the standard measure of synchrony is the order parameter, typically but not exclusively defined as [124, 84, 173]:

$$r(t) \quad = \quad \left| \frac{1}{N} \sum_{j=1}^{N} e^{i\theta_j(t)} \right|, \tag{15}$$

where, as before, N denotes the number of oscillators in the network, and $\theta_j(t)$ the instantaneous phase of oscillator j. Geometrically, the value of the order parameter indicates how well a given set of unit vectors are aligned with respect to one another (with 1 indicating perfect alignment). A slightly more general definition is adopted in [154], incorporating the adjacency matrix to account for the network's local structure. Much the same measure is used again in [89].

6.4 Synchronizability

Recent studies have indicated that particular network properties, such as the average clustering coefficient and betweenness centrality, among others, have a major impact on the dynamics of a network [124, 89, 84, 73]. Here we shall review those results that relate specifically to synchronization.

Kuramoto Oscillators on Random Graphs

The study of systems of coupled oscillators has recently been extended from dealing exclusively with networks with all-to-all coupling to include networks with local connectivity, such as lattices and, indeed, random networks (particularly scale-free and small-world networks). It has become clear that these complex networks differ from their regular (random) counterparts in many ways, and not least in terms of their dynamic properties. Great interest has been expressed in the question as to what extent the topology of a network determines the behaviour of the same; or more in particular for a system of coupled oscillators: to what extent the topology impacts the transition behaviour.

As regards the latter question, in [154] the transition behaviour of an appropriately defined order parameter was approximated to good accuracy in large networks of almost arbitrary structure. In particular, the following expression for the critical coupling strength was derived:

$$k_c \quad = \quad \frac{k_0}{\lambda}. \tag{16}$$

Here k_0 is a constant, depending on the distribution of the oscillators' intrinsic frequencies, and λ is the spectral radius of the network's adjacency matrix. Note that this estimate requires full knowledge of the adjacency matrix. A less restrictive estimate was obtained by introducing the additional assumption that the components of the eigenvector associated with the spectral radius are proportional to the vector of node degrees. The expression thus obtained reads

$$k_c = k_0 \frac{\langle k \rangle}{\langle k^2 \rangle}, \tag{17}$$

which coincides with the result reported in [89]. A detailed account of the validity of the various assumptions involved can be found in the paper [154]. In the above expression (17), $\langle k \rangle$ and $\langle k^2 \rangle$ denote the first and second moments of the node degree distribution, respectively. As pointed out in [89], for scale-free networks with a power law coefficient between 2 and 3, the second moment grows without bound as the number of nodes tends to infinity. This would suggest that, in such networks, there is no critical coupling in the thermodynamic limit; or indeed no threshold for coherent oscillations. This has been demonstrated not to be the case for finite networks [182, 89]. Indeed, in [89] it is reported that there exists a clear dependence between the critical coupling strength and the network size. We draw attention to the fact that related observations have been reported in the literature on disease propagation. Particularly, the absence of an epidemic threshold has been established as a characteristic feature of disease spread models on (infinite) scale-free networks. Finite-size effects have also been discussed in this context [123]. The similarity between the physics of coupled oscillators and models of disease spread has been discussed previously in [89]. We shall have more to say about this connection in the next section.

Factors that Promote Synchronization

Let us consider what structural properties of a network promote synchronization. A recent study [124] suggests that one factor might be the amount of *clustering*. Indeed, the study indicates that networks (Poisson or scale-free) that share the same number of nodes, the same number of edges and the same degree distribution, but have a different average clustering coefficient, may have very different synchronization properties. In particular, it was found that increasing the clustering coefficient of a Poisson network leads to a more gradual transition from incoherence to coherence. For scale-free networks, the effect was more ambiguous in that increased clustering appeared to promote the onset of synchronization at low coupling strengths, suppressing the same at high coupling strengths. For moderate coupling strengths the network would seem to split into several dynamic clusters oscillating at different frequencies. The authors proposed that scale-free networks with high clustering undergo two separate transitions: a first transition to a partially synchronized state, corresponding to the formation of clusters oscillating at distinct frequencies; followed by a second transition to full synchronization when the clusters are tuned to a common frequency.

Other factors, reported in [84], in a study of Watts-Strogatz small-world networks, include large maximum degree, short characteristic path length, heterogeneity of the degree distribution and a low value for the average betweenness centrality. Among these factors, betweenness centrality was found to account for the strongest correlations. Some of these findings have been shown not to hold for other types of networks. Notably, for scale-free networks, it appears that homogeneity in the degree distribution, rather than heterogeneity would promote synchronization [138]. This would contradict the popular belief that because the average path length in heterogeneous networks tends to be smaller than, for example, in lattices, communication between oscillators would be more efficient, which would amount to better synchronizability. Using the ratio between the smallest (nonzero) and the largest eigenvalue of the Laplacian as a measure of synchrony (which was also the measure used in [84]), the authors demonstrated the opposite, namely that heterogeneity in a scale-free network tends to inhibit rather than promote its ability to synchronize.

In [182], it was demonstrated numerically that (finite-size) scale-free networks of Kuramoto oscillators exhibit a phase transition at a coupling strength that is inversely proportional to the average node degree. In the same study, the authors also investigated the 'fitness for synchronization' of particular

network motifs, defining fitness as the (normalized) coupling strength at which the probability that a motif synchronizes first exceeds one half. The results suggested that motifs with high interconnectedness are more prone to synchronize. Interestingly, this ability to synchronize was found to be correlated with the motif's natural conservation rate in the yeast protein interaction network (see Section 5.3).

In a study involving d-dimensional lattices of coupled oscillators [85], it was investigated what the minimal dimension d^* of a lattice should be in order for the oscillators to synchronize in the limit of strong coupling. Based on extensive simulations the authors conclude that $d^* = 3$ for frequency synchronization, and $d^* = 5$ for phase synchronization.

For networks of Erdös-Rényi type, the authors of [73] derived a lower bound on the critical average degree, that is the smallest average degree for which synchronization is possible. Moreover, if p is the probability that an edge is placed between a given pair of nodes in an ER network, it was shown that networks with different values of p share the same critical coupling strength, which is that of the globally coupled network.

In small-world networks, the onset of phase and frequency synchronization appears to depend strongly on the rewiring probability when this probability is small, and no synchronization whatsoever is observed when this probability is identically zero [83]. Interestingly, the synchronization behaviour appears to be roughly the same for larger values of the rewiring probability, suggesting some form of saturation to set in.

6.5 Synchronization in the brain

Having described various theoretical aspects of synchronization, we shall close this section with a discussion on the proposed role of synchronization in the brain.

Synchronization and the Problem of Integration of Information

Synchronization has been put forward by some as the mechanism that would make possible the integration of distributed neural activity in our brain [179]. Others have argued against this. Here we shall focus on the supporting evidence. What is neural integration and what part does synchronization have to play in it? Recent studies suggest that during processing of visual and auditory stimuli, activities of functionally specific brain regions are temporally aligned so as to produce a unified cognitive moment. This would imply that an inability to synchronize, due to abnormalities in the neural circuit for instance, could have severe behavioral implications [139, 171]. An understanding of the mechanics of this phenomenon may thus hold the key to devising new treatments for neurological disorders.

It has been known for a long time that groups of neurons *within a single sensory modality* such as the visual cortex, selectively synchronize their activities, supposedly to integrate the particular features for which they encode. However, the fact that this same kind of integration would take place *across different sensory modalities* was discovered only recently. In a study reported by Roelfsema *et al.* [156], five cats were conditioned to press and release a lever in response to particular visual stimuli. Electrodes were implanted at different locations in the motor and visual cortices to monitor the electrical activity during execution of the task. Coupling between these brain areas was investigated using cross-correlation analysis on pairs of LFP (Local Field Potential) traces. Tighter coupling was observed when the animals were engaged in the specific visuomotor task than when engaged in feeding or at rest. Based on these and other findings, Varela *et al.* have suggested that "large-scale synchrony is the underlying basis for active attentive behaviour". [179, 155].

In a more recent study [168], it was investigated how the interactions between selected areas in the hippocampus and amygdala in fear-conditioned mice compare against those in controls. The response of the fear-conditioned group indicated a selective synchronization in the theta frequency range (4-7 Hz) upon presentation of the conditioned stimulus, which was not found in the control group. No

significant synchronization was observed in either group during presentation of the unconditioned stimulus. It was argued that these results are indicative of a functional relationship between theta rhythm synchronization and the retrieval and expression of fear.

Abnormal Neural Synchrony and Schizophrenia

Assuming synchronization is the mechanism that underlies neural integration, it seems reasonable to suppose that disruptions in neural synchrony would impact one's behaviour. Interestingly, an impaired ability to integrate information has long been identified as one of the symptoms of Schizophrenia, which makes this disorder particularly relevant in this context. Schizophrenia, a complex and debilitating disease, is generally defined in terms of its symptoms, which may be divided into (a) positive symptoms, which include delusions, hallucinations, and incoherent thoughts; and (b) negative symptoms, which include social withdrawal, poor motivation, and apathy [160, 101]. In recent years, it has been proposed that these cognitive and affective impairments may be related to a defect in the mechanism believed to be responsible for the integration of distributed neural activity, that is, to gamma band synchronization [171, 139, 84].

A recent report supports this [171]: when a set of Gestalt images were presented to a group of patients diagnosed with Schizophrenia (SZ) and a group of Normal Control (NC) subjects, a significant difference in neural orchestration between the two was observed. A phase-locking response, persistent among individuals from the NC group, but absent in the SZ group, was hypothesized to reflect a feature-binding mechanism in the visual cortex which would explain the more efficient task performance by healthy individuals.

Further evidence for abnormal neural synchrony in Schizophrenia was reported in [91]. In this study, two groups, patients and controls, were presented with a set of images depicting six basic human emotions, which they were to recognize. The response of each individual was measured using whole head MEG (Magnetoencephalogram). Local activity was averaged over a so called region of interest (ROI) and a coherence score was computed as the mutual information (MI) [44] between ROIs. The MI analysis revealed a very organized pattern of linkages for normal subjects, as opposed to the overall disturbed linkages for Schizophrenia patients. At some level, these results agree with the outcome of another study [84], which involved first-degree relatives of patients with Schizophrenia. Gamma-band synchronization was found to be reduced in first-degree relatives with Schizophrenia Spectrum Personality Problems.

A Theory of Neural Synchronization?

It has been established beyond doubt that the processing of particular audiovisual stimuli coincides with the temporal synchronization of neural activities in functionally specialized brain regions. In addition there is some evidence that patients with Schizophrenia or related neurological disorders are more likely to display abnormal patterns of synchrony than controls. Meanwhile, the mechanics of this synchronization and its supposed role in the integration of information remain poorly understood. Most experimental studies resort to elementary statistical techniques to conclude with confidence that some form of synchronization takes place. Beyond that, there appears to be a shortage of quantitative models; models that do not just extract information from the data, but indeed attempt to explain the data. With no disrespect for the seminal importance of Kuramoto's work, and that of others' who have contributed to the theory of coupled oscillators, it appears that we are still far removed from effectively applying this theory in the context of the neural synchronization problem. Fortunately, there is reason to believe that this gap is closing fast, considering on the one hand the rate at which measurement techniques are being refined, and, on the other hand, some of the pioneering work that is being done on the theoretical front.

6.6 Summarizing Comments

In this section we discussed some aspects of synchronization, using the Kuramoto model of coupled oscillators as a starting point. We reviewed recent results on the relation between network structure and synchronizability.

(i) the onset of synchronization in complex networks is determined by a few key factors that include: the average clustering coefficient, the second moment of the degree distribution, the maximum degree, the characteristic path length and the average betweenness centrality. These factors may impact different networks in different ways.

(ii) For scale-free network of infinite size and with a power law exponent between 2 and 3, the value for the critical coupling is zero. For finite-size scale-free networks, the critical coupling is nonzero. We pointed out parallel results in the disease propagation literature.

(iii) We discussed the role of synchronization in the brain and argued that, ongoing efforts notwithstanding, there is a lot of work to be done in the way of tuning the abstract mathematical models of coupled oscillators to the experimentalist's needs.

7 Network Structure and Disease Propagation

The final major topic that we shall consider here is the impact of network structure on disease propagation models. Given that several of the novel network properties considered in the recent past have been observed in social networks and in networks of human sexual contacts [118], it is natural to ask what effect these properties have on the spread of disease through such networks. Given the emergence of new virulent diseases such as the SARS virus and the Asian bird flu, the importance of understanding the interaction between network structure and the dynamics of disease propagation cannot be over-emphasised. The current section is organised as follows. First, we shall discuss recent numerical and theoretical work on the effect of different degree distributions on the behaviour of classical epidemic models, with particular emphasis on the effect of power-law distributions on the so-called *epidemic threshold*. We shall then discuss extensions of this basic line of research which have attempted to take into account finite-size effects correlations between the degrees of connected nodes. Finally, we shall discuss a number of other issues pertaining to disease spread on networks, including the containment of epidemics on different network topologies and the evolution of different disease strains.

7.1 Scale-free Networks and Epidemic Thresholds

The mathematical theory of epidemics has been the subject of intensive research for some time now and several different models for disease spread have been developed. A detailed discussion of the properties of all of these models is well beyond the scope of the current document, and the interested reader should consult [8, 78]. Here, we shall confine our discussion to results concerned with two basic models of disease spread: the *Susceptible-Infected-Susceptible* or *SIS* model and the *Susceptible-Infected-Removed* or *SIR* model. Much of the recent work on disease propagation through networks has focussed on these two core models.

In the SIS model, a population is divided into two groups: the first (S) consists of susceptible individuals, who are not infected but can contract the disease from members of the second group (I) of infected individuals. After a period of time, an infected person recovers and then becomes susceptible again. Hence no immunity is conferred by contracting the disease and the recovered infective can become infected again at a later time. In contrast, in the SIR model, a recovered infective is regarded as being immune to the disease and cannot subsequently become infected again. Hence, the population

is divided into three groups in such models: susceptibles (S), infectives (I) and removed or recovered (R).

There are two fundamental parameters associated with any SIS or SIR model: the probability λ of an infective passing on the disease to a susceptible with whom they are in contact during the period in which they are infective, and the rate ν at which an infective recovers. In basic models of population epidemiology, it is assumed that the population is homogeneously mixed. This essentially amounts to assuming that each individual, or node, in the population has the same number of contacts. Under the assumptions of homogeneous mixing and a fixed population size, the standard equations for the SIR model are given by [130, 30]

$$\begin{aligned} \frac{dS}{dt} &= -\lambda SI \\ \frac{dI}{dt} &= \lambda SI - \nu I \\ \frac{dR}{dt} &= \nu I. \end{aligned} \tag{18}$$

Here, the variables $S(t), I(t), R(t)$ represent the total number of individuals in the susceptible, infected and recovered classes respectively at time t. From a network point of view, we can consider the population as a graph, G, in which each individual is represented by a node and each edge represents a contact or connection between individuals, through which the disease can spread. In a homogeneously mixed population, each node v in G has the same degree, which would be equal to the mean degree, $\langle k \rangle$, of the network. This assumption is only reasonable for networks whose degree distributions are narrow, meaning that the coefficient of variation, $C_V = \sqrt{\langle k^2 \rangle / \langle k \rangle^2 - 1}$, is very small.

Under the assumption of homogeneous mixing, the quantity $\iota_0 = \langle k \rangle \lambda / \nu$, represents the average number of secondary infections that would result from the introduction of a single infected individual into an entirely susceptible population. In this case, the introduction of an infective into the population will result in an epidemic if the basic reproductive number $R_0 = \iota_0$ is greater than one, while if $R_0 < 1$, the disease will die out. Thus, defining $\lambda_c = \nu / \langle k \rangle$, an epidemic occurs if the spreading rate, λ satisfies $\lambda > \lambda_c$ while the disease dies out if $\lambda < \lambda_c$. The constant λ_c is usually referred to as the epidemic threshold.

While the assumption of homogeneous mixing might be reasonable for the classical ER random graph models, it is entirely inappropriate for BA and other scale-free networks with broad-tailed degree distributions. In [123], the dynamics of the SIR model on heterogeneous networks of this type were studied. It was pointed out that for such networks, the basic reproductive number R_0 is given by the formula

$$R_0 = \rho_0 (1 + C_V^2). \tag{19}$$

Now, in the limit as network size tends to infinity, for a scale-free network with degree distribution of the form $P(k) \sim k^{-\gamma}$ with $2 < \gamma < 3$, the coefficient of variation C_V of its node-degrees is infinite (more precisely, the second moment $\langle k^2 \rangle$ diverges as the network size, n, tends to infinity, while $\langle k \rangle$ remains finite). Thus, for any non-zero spreading rate λ, the introduction of an infective into the population can result in an epidemic. Similar findings were also reported in [142] for the SIS model. These results lead to the somewhat surprising conclusion that for scale-free networks, the epidemic threshold is effectively zero. This also follows from the following formula for the epidemic threshold for scale-free networks with degree distribution $P(k) \sim k^{-3}$, which was presented in [144] (as well as a number of other sources).

$$\lambda_c = \frac{\langle k \rangle}{\langle k^2 \rangle} \tag{20}$$

Note that this same formula has appeared above in the context of coherent synchronization on random networks (17).

The authors of [123] also derived approximate expressions for the fraction of nodes, I, in a scale-free network that are ever infected for an SIR model of disease spread. (I is usually referred to as the final epidemic size.) First of all, for scale-free networks with power-law exponent $\gamma = 3$, they demonstrated that, essentially, I decreases with decreasing λ as $Ce^{-(A/\lambda)}$ for constants A, C. Using approximate, mean-field arguments and simulations, a similar result was derived in [142] for the steady-state prevalence of an SIS epidemic[4]. The dependence of I on λ for networks with $2 < \gamma < 3$ was also calculated in [123] and it was established that in this case, I decays with decreasing λ according to a power law of the form $C(\lambda)^{1/(3-\gamma)}$. In the same paper, it was also shown that, for networks with $\gamma = 3$, the number of infected nodes of low-degree is small, while many (essentially all) nodes of high-degree are infected. These findings are in agreement with those described in [14], which indicate that disease spreads in a hierarchical cascade from hub nodes to nodes with intermediate degree to nodes with low degree. These observations clearly have significant implications for the development of containment strategies. Specifically, they suggest that an effective containment strategy would first and foremost target the hubs of a network. Similar recommendations have been made in [45].

Before we proceed, it should be noted that the results discussed in the previous paragraph are based on a number of assumptions.

(i) The results described above were derived for the limiting case of an infinite network or population, and rely on a continuous approximation of the node-degree variable k. It has been noted in [123] that when finite size effects are taken into account the epidemic threshold does not vanish but in fact takes a positive value.

(ii) These results apply to networks in which there is no correlation between the degrees of connected nodes.

(iii) Finally, as with biological interaction networks, the inferred scale-free nature of social and sexual networks typically relies on sampled network data. Hence, in order to reliably apply the results discussed here, it is vital to understand the effect of sampling on the identification of a network's topology.

Later in this section, we shall describe the results of a number of authors who have attempted to address some of these limitations.

7.2 Impact of Finite Size and Local Structure on Disease Spread

The above results on the properties of SIS and SIR models on scale-free networks were derived for the limiting case of networks of infinite size. Of course, real networks of social and sexual contacts are finite and, for this reason, a number of authors have studied the dynamics of disease spread on scale-free networks with finitely many nodes. In [143], the epidemic threshold, λ_c, and the steady-state prevalence, ρ, for the SIS model on finite scale-free networks were investigated. It was found that λ_c is non-vanishing in this case, and formulae approximating the dependence of λ_c and ρ on the network size, n, were also derived. Note that while the epidemic threshold is non-vanishing for the finite scale-free networks studied in [143], it is considerably smaller than for a corresponding homogeneous network with the same average degree. In fact for scale-free networks of size larger than 1000, the threshold is at least one order of magnitude smaller than in the homogeneous case. These findings are largely in agreement with the remarks on finite-size effects for SIR models made towards the end of the paper [123]. Note also the findings reported in [88] where the behaviour of the SIS model on two different types of network with scale-free degree distributions was studied numerically. For both network types, the epidemic threshold λ_c is non-zero. However, the dependence of λ_c on network size and the effect of the spreading rate λ on ρ varied significantly between the two classes of network, even for networks with the same underlying degree distribution. These results demonstrate that it is

[4]The steady-state prevalence is the fraction of infected nodes in the steady state.

possible for two networks with the same degree distribution, but different local structures, to exhibit significantly different behaviours with respect to disease propagation.

The final observation in the previous paragraphs has motivated a number of authors to study classes of scale-free networks in which the degrees of neighbouring nodes are correlated. Such networks offer a more realistic picture of real social networks in which such correlation is common. In [53] the SIS model was studied on a class of highly-clustered scale-free networks. Numerical simulations indicated that the highly clustered networks behave in a qualitatively different manner than the usual scale-free models, both with respect to the dependence of steady-state prevalence ρ on spreading rate λ and to survival probability of the disease. Moreover, the authors of this paper argue that for this highly structured class of scale-free networks, there is a non-vanishing epidemic threshold even in the limit as the network size, n, tends to infinity. They further conjectured that the value of the threshold depends on the degree correlations within the network rather than on the degree distribution itself.

The relationship between the epidemic threshold and the degree correlations in a scale-free network has been further investigated in [19, 20]. In [19] the value of the epidemic threshold is related to the largest eigenvalue of the so-called connectivity matrix C, where $C_{kk'} = kP(k'|k)$. Here $P(k'|k)$ represents the probability that a given link emanating from a node of degree k connects to a node of degree k'. For networks with no higher order correlations, they demonstrate that the epidemic threshold is equal to the reciprocal of the largest eigenvalue of C. Based on these results, in [20] conditions for the absence of an epidemic threshold in scale-free networks with arbitrary two-point degree correlation functions $P(k'|k)$ and degree exponents in the range $2 < \gamma \leq 3$ were investigated. The principal result of this paper established that in this case, provided the network possesses no additional, higher order, structure, the epidemic threshold is again zero in the limit of infinite network size. We should also note here the work described in [185, 129] which further investigated the effects of degree correlations and local structure on the dynamics of disease spread in scale-free networks.

7.3 Containment Strategies on Heterogeneous Networks

One of the most fundamental issues in epidemiology is how to design effective strategies for containing the outbreak of an infectious disease. One simple strategy would be mass vaccination, in which (almost) every individual in the population is vaccinated against a disease, and hence immune to it. While this can be an effective strategy for containing infectious diseases, it is crude and operationally expensive. As a result, there is great interest in alternative strategies which, although perhaps slightly less effective, are much more economical in terms of resources and logistics. Recently, in [45, 144], the implications of power law degree distributions for the design of immunization programmes was investigated using mean-field approximations and numerical simulations. The first strategy considered was that of uniform random vaccination in which individuals are uniformly selected at random and vaccinated. However, while this strategy can work for homogeneous populations, it is known to be ineffective in the heterogeneous case [8]. The findings in [45, 144] suggest that for scale-free networks, and the SIS model of disease spread, considerable improvements over uniform vaccination can be achieved through targeting hub nodes within a network. In fact, two different approaches of this kind were suggested. In the first of these, nodes are vaccinated with probability proportional to their degree, so that a greater proportion of nodes of high degree are vaccinated than is the case for nodes of low degree. The second strategy aims to specifically target hub nodes by vaccinating all nodes in the network of degree higher than some threshold k_c. While this appears to be more cost effective, in terms of how many individuals need to be immunized in order to eventually eradicate the disease, it relies on a fairly complete knowledge of the network's topology. As mentioned before in the text, it is unrealistic to assume that we will have exact knowledge of each individual's degree within the network and the impact of sampling errors and inaccurate network data on vaccination schemes needs to be analysed more thoroughly.

A disease containment strategy, aimed at controlling outbreaks of smallpox was recently proposed in [58]. In this paper, the social networks through which disease spreads were modelled as bi-partite

graphs [47]. Such a graph has two distinct types of vertices, which correspond to locations and individuals respectively. The results of this paper suggest a containment strategy of targeted vaccination combined with early detection. Early detection could be accomplished by placing sensors at locations with high degree, that is, locations visited by many people, while efficient vaccination is effected by targeting long-distance travellers. The impact of factors such as targeted or mass vaccination schemes, withdrawing infected individuals to their homes, and delays in introducing containment measures, on the number of deaths caused by a smallpox outbreak was investigated by numerical simulation. The results suggested that the most significant factor was the early removal of infected individuals to their homes with the next most influential factor being the length of delay in implementing vaccination schemes.

In [15], motivated by the recent emergence of the SARS virus, several intervention strategies for epidemic containment were considered, and the impact of each strategy on the effective reproduction number was determined. In general, the results of the paper suggest that combining different strategies is a good idea, while the strategy of tracing and quarantining the contacts of diagnosed cases was found to be particularly effective. The model studied in this paper incorporated several realistic aspects of social structure. For instance, given that people tend to be more frequently in contact with individuals within their own household than with people from other households, a distinction was drawn between *within-household* transmission and *between-household* transmission. Furthermore, school-children and the rest of the population were considered separately. While the manner of counting secondary infections, and the reproduction number, used in this paper were somewhat non-standard, they have the advantage of being analytically tractable and, moreover, the number of "offspring" of a single infective, as counted in this paper, is independent of the size of the infective's own household. Parameter values pertaining to the distribution of household sizes were selected in accordance with given census data. Various control strategies were considered, including exposure avoidance, isolating cases at diagnosis, closing schools, quarantining affected household, and contact tracing. Apart from the efficacy of the above mentioned strategy of tracing and quarantining contacts of diagnosed cases, the results indicate that if an emerging infection were to enter a juvenile population, closing schools can reduce transmission significantly.

7.4 Other Network Models and the General Theory of Disease Spread on Networks

In addition to the work discussed above on epidemic dynamics on scale-free network, a number of authors have considered the problem of disease spread on other network topologies. For instance, in [159] the impact of dynamically adding long-range links to regular one-dimensional lattices on the spread of disease was studied. Using the SIR model for disease spread, they have shown that the resulting small-world network [192] structure exhibits a shortcut-dependent epidemic threshold. An approximate expression for this threshold in terms of the effective spreading rate and the effective recovery rate was shown to be accurate over a large range of parameter values. The authors also acknowledged the fact, previously stated elsewhere [7, 123], that the basic reproduction number has limited use outside the homogeneous mixing paradigm. They argue that this is particularly true for small-world networks because "the effect of a secondary infection caused by nearest-neighbor transmission is different from the one caused by a long-range jump" [159]. Assuming a spreading probability of one, so that susceptibles in direct contact with infectives will become infected during the next iteration step, it was shown that the epidemic saturation time, i.e. the time it takes for 95% of the susceptible population to become infected, scales with $-log(n_0)$, where n_0 is the fraction of nodes initially infected. The scenario of spreading with near certainty would correspond to the onset of an epidemic, and is used by the authors to predict the final epidemic size as well the development of an epidemic from its beginning stages. The dynamics of the SIR model and the related *susceptible-exposed-infected-removed* (SEIR) model on small-world network were also investigated in the paper [183].

Recently, in [132] analytical techniques were developed which can be used to derive exact solutions for a large class of standard epidemiological models on a variety of networks. These techniques are based on generating functions and allow for great flexibility in terms of assumptions on network structure and degree correlations. Further they can accommodate heterogeneity in transmission rate and infectious period and allow for correlations between parameters such as transmission rate and node degree. The results derived in this paper include formulae for the epidemic threshold and average outbreak size for the network classes considered. More recently, the problem of epidemic spread on random graph models has been studied in a mathematically rigorous fashion within the framework of Markov processes in [66]. Here, the dependence of the final epidemic size and the lifetime of an outbreak on graph parameters such as the spectral radius of the network's adjacency matrix and the isoperimetric number of the network was investigated. Some general theorems as well as results for a variety of graph models including the ER and scale-free models were derived for the SIS and SIR models of disease spread.

The techniques developed in [132] were then applied in [7] in an effort to explain some puzzling aspects of the recent SARS outbreaks. Specifically, the question of why these outbreaks never led to an epidemic, given the relatively high estimates for the basic reproduction number, was considered. Using purely analytical tools, the authors derive expressions for the likelihood that a small outbreak results in an epidemic in, respectively, an urban network, a power-law network, and a Poisson network. It turns out that (and this is confirmed by numerical simulations) "outbreaks are consistently less likely to reach epidemic proportions in the power-law network than in the others". It should also be noted that it was shown that for all networks there is a nonzero probability that an outbreak does not become an epidemic, even when the spreading rate of a disease exceeds the epidemic threshold. By contrast, in the paradigm of homogeneous mixing, an epidemic will occur with certainty whenever the basic reproduction number is greater than unity. Other interesting findings are that:

(i) The likelihood of an outbreak is a monotonically increasing function of the degree of the first infective;

(ii) When the transmissibility of disease is far above the epidemic threshold, the risk of an epidemic is very high even for small initial outbreaks, at least in the case of urban networks.

Finally, we note that the evolution of diseases on local and global networks has been studied in [153]. The basic premise of this work was that different disease strains adapt to compete for resources (susceptible hosts). In the model proposed here, adaptation corresponds to a random mutation of both the transmission rate and the infectious period, which takes place whenever a new infection occurs. As the authors point out, in mean-field models this type of evolution would result in runaway behavior with selection for ever higher transmission rates and ever longer infectious periods. By contrast, both spatial heterogeneity in local networks and the presence of shortcuts in global networks appear to constrain the evolutionary dynamics, to the effect that the rate of adaptation is generally slower (in the case of a global network, the transmission rate even saturates at some finite value) and the variability (in the dynamics) higher than in mean-field models. Simulation results suggest that in networks with many long-distance connections and a low clustering coefficient, disease strains with conservative transmission rates and long infectious periods are most likely to survive. By comparison, for networks with strong local connectivity the fittest strains are those that have high transmission rates and relatively short infectious periods.

7.5 Summarizing Comments

(i) The structure of a social network can have a significant impact on the dynamics of disease propagation. In particular, it has been shown for scale-free networks, in the limiting case of infinitely many nodes, that the epidemic threshold is zero. This would mean that any non-zero spreading rate could lead to an epidemic.

(ii) The previous fact was initially established for uncorrelated scale-free networks of infinite size. For scale-free networks of finite size, the epidemic threshold is non-vanishing but considerably smaller than in the case of a homogeneously mixed population.

(iii) Results have recently been derived giving conditions under which the epidemic threshold will be zero for scale-free networks with degree correlations, in the limiting case of networks of infinite size.

(iv) The dynamical behaviour of epidemics on networks with heterogeneous degree distributions has implications for the design of strategies for containing outbreaks. In particular, the targeting of nodes, or individuals, of high degree can offer significant improvements over random immunization programmes.

8 Conclusions and Directions for Future Research

The need for a more systematic approach to the analysis of living organisms, alongside the availability of unprecedented amounts of data, has led to a considerable growth of activity in the theory and analysis of complex biological networks in recent years. Networks are ubiquitous in Biology, occurring at all levels from biochemical reactions within the cell up to the complex webs of social and sexual interactions that govern the dynamics of disease spread through human populations. Over the last few years, several core themes and questions in biological network analysis have arisen from pressing problems in Biology and Medicine. For instance, while the research on bio-molecular and neurological networks is still at a relatively early stage, a comprehensive understanding of these networks is needed to develop more sophisticated and effective treatment strategies for diseases such as Cancer and Schizophrenia. Other aspects of this line of research have been motivated by the need to determine the biological role of unannotated genes or proteins. On the other hand, at the level of social networks, future approaches to epidemic containment will need to take into account the interplay between network topology and dynamics.

Our aim in this article has been to provide as comprehensive an overview as possible of the uses of Graph Theory and Network Analysis within Biology, and to point out problems in Graph Theory that arise from the study of biological networks. Specifically, we concentrated on the following five broad topics.

(i) *Structural identification and modelling of bio-molecular networks*

Recent advances in high-throughput techniques have led to the construction of maps of protein-protein interaction, transcriptional regulatory and metabolic networks for a variety of organisms. Numerical investigations of the properties of these network maps, described in Section 3, indicate that they tend to have short characteristic path lengths, high clustering coefficients and scale-free degree distributions. Motivated by these observations, mathematical models such as the Barabasi-Albert scale-free network and Duplication-Divergence models have been proposed for protein interaction and genetic networks. However, the experimental techniques on which these network maps are based are prone to high rates of false positive errors, and typically only cover a fraction of the network's nodes. The development of more accurate and reliable experimental methodologies is of course of vital importance for future research on the structure of bio-molecular networks. On a more theoretical level, two of the most significant issues that need to be addressed in this area are the sampling properties of complex networks and the impact of data inaccuracies on the identification of network statistics such as the degree distribution.

(ii) *Centrality measures and essentiality in gene and protein networks*

Much of the research on applying centrality measures to bio-molecular networks has focussed on the prediction of gene or protein essentiality. In most of the studies discussed in Section 4, the centrality score of a node was found to be indicative of its likelihood to be essential. In

particular, this appears to be true for degree centrality, betweenness centrality and eigenvector centrality measures. However, there is no clear evidence that the more complex centrality measures perform any better than degree centrality. A major source of open problems in this area is the robustness of centrality measures to data inaccuracies. Once again, this issue is very important for the reliable application of these techniques to biological data.

(iii) *Motifs, modules and the hierarchical structure of bio-molecular networks*

The research on motifs described in Section 5 has helped to clarify the structural organisation of complex biological networks. Furthermore, the motifs of a network appear to characterise it in some sense, and motifs such as the feed-forward loop seem well suited to specific information processing tasks. However, while the motifs of a network represent statistically significant patterns, their precise biological significance and the mechanisms behind their emergence are only partially understood. To date much of the work on motifs has been numerical in nature, and the theoretical analysis of the motif profiles in mathematical network models is a potentially rich source of open and challenging problems. Moreover, analysis of this type will provide further insights into how accurately models such as Duplication-Divergence network describe real biological networks. A related area of research, discussed in Section 5, is the identification of functional modules and the prediction of protein function based on network topology. The latter problem is of considerable importance and the results discussed in the text indicate the potential of graph theoretical approaches to this question.

(iv) *Synchronization, network topology and neurological function*

Among the key issues in the study of complex networks is the question as to how indicative a network's topology is of its overall behaviour. Studies on systems of coupled oscillators suggest that, as far as fitness for synchronization is concerned, there are at least five structural properties that qualify as important indicators. These are: average betweenness centrality, average clustering coefficient, maximum degree, node degree variance, and characteristic path length.

The dynamics of synchronization on scale-free networks are characterized by a two-stage transition, initiated at low coupling strengths by the formation of distinct clusters oscillating at distinct frequencies, followed by a process of alignment at high coupling, during which the different clusters are tuned to a common frequency. A second characteristic property of scale-free networks is that, in the limiting case of infinitely many nodes, the threshold for the onset of synchronization vanishes.

At present, the majority of results on synchronization in complex networks appear to have been obtained using a combination of approximations and extensive simulations. As such, there is a clear need for a rigorous mathematical analysis to support, and underpin, the numerical findings. Also there is work to be done in applying the abstract mathematical models of phase-coupled oscillators to the analysis of experimental data.

(v) *Network structure and epidemic dynamics*

The interplay between epidemic dynamics and network structure is vital for understanding and containing the spread of infectious diseases. The numerical studies and mean-field analyses discussed in Section 7 have shown that a scale-free topology can significantly reduce the epidemic threshold, making the outbreak of epidemics more likely in networks with such a structure. Network topology also has an impact on the effectiveness of immunization schemes for containing epidemic outbreaks. In particular, for networks with a scale-free topology, the targeted immunization of nodes of high degree offers substantial improvements over uniform random immunization. Of course, the reliable identification of social network structure is vital for the practical implementation and interpretation of such results. One important direction for future research in this area is the extension of recent results to incorporate the effects of sampling and data noise on epidemic dynamics on networks and containment strategies.

To finish, it is our hope that this article will be of assistance to the broad community of researchers

working on the study of biological networks, by highlighting recent advances in the field, as well as significant issues and problems that still need to be addressed.

Acknowledgements

This work was partially supported by Science Foundation Ireland (SFI) grant 03/RP1/I382 and SFI grant 04/IN1/I478. Science Foundation Ireland is not responsible for any use of data appearing in this publication.

References

[1] J.A. Acebrón, L.L. Bonilla, C. J. Pérez Vicente, F. Ritort, and R. Spigler. The Kuramoto model: A simple paradigm for synchronization phenomena. *Reviews of Modern Physics*, 77:137–185, 2005.

[2] R. Albert and A. Barabasi. The statistical mechanics of complex networks. *Reviews of Modern Physics*, 74:47–97, 2002.

[3] R. Albert, H. Jeong, and A. Barabasi. Diameter of the world-wide web. *Nature*, 401:130–131, 1999.

[4] R. Albert, H. Jeong, and L. Barabasi. Error and attack tolerance of complex networks. *Nature*, 406:378–382, 2000.

[5] E. Alm and A. Arkin. Biological networks. *Current Opinion in Structural Biology*, 13:193–202, 2003.

[6] U. Alon. Biological networks: the tinkerer as engineer. *Science*, 301:1866–1867, 2003.

[7] L. Ancel Meyers et al. Network theory and SARS: predicting outbreak diversity. *Journal of Theoretical Biology*, 232:71–81, 2005.

[8] R. M. Anderson and R. M. May. *Infectious diseases of humans: dynamics and control*. Oxford University Press, 1991.

[9] Y. Artzy-Randrup et al. Comment on "network motifs, simple building blocks of complex networks" and "superfamilies of evolved and designed networks". *Science*, 305:1107c, 2004.

[10] M. Babu et al. Structure and evolution of transcriptional regulatory networks. *Current Opinion in Structural Biology*, 14:283–291, 2004.

[11] Z. Bar-Joseph et al. Computational discovery of gene modules and regulatory networks. *Nature Biotechnology*, 21(11):1337–1342, 2003.

[12] L. Barabasi and R. Albert. Emergence of scaling in random networks. *Science*, 286:509–512, 1999.

[13] L. Barabasi and Z. Oltvai. Network biology: understanding the cell's functional organization. *Nature Reviews - Genetics*, 5:101–113, 2004.

[14] M. Barthelemy et al. Dynamical patterns of epidemic outbreaks in complex heterogeneous networks. *Journal of Theoretical Biology*, 235:275–288, 2005.

[15] N. Becker et al. Controlling emerging infectious diseases like SARS. *Mathematical Biosciences*, 193:205–221, 2005.

[16] J. Berg, M. Lassig, and A. Wagner. Structure and evolution of protein interaction networks: a statistical model for link dynamics and gene duplications. *BMC Evolutionary Biology*, 4:51, 2004.

[17] A. Berman and R.J. Plemmon. *Non-negative matrices in the mathematical sciences*. SIAM classics in applied mathematics, 1994.

[18] A. Bhan, D. Galas, and T.G. Dewey. A duplication growth model of gene expression networks. *Bioinformatics*, 18(11):1486–1493, 2002.

[19] M. Boguna and R. Pastor-Satorras. Epidemic spreading in correlated complex networks. *Physical Review E*, 66:047104, 2002.

[20] M. Boguna, R. Pastor-Satorras, and A. Vespignani. Absence of epidemic threshold in scale-free networks with degree correlations. *Physical Review Letters*, 90(2):028701, 2003.

[21] B. Bollobas. *Random Graphs*. Cambridge University Press, 2001.

[22] B. Bollobas and O. Riordan. *Handbook of Graphs and Networks*, chapter "Mathematical results on scale-free graphs". Wiley, 2002.

[23] B. Bollobas and O. Riordan. Robustness and vulnerability of scale-free random graphs. *Internet Mathematics*, 1:1–35, 2003.

[24] B. Bollobas and O. Riordan. The diameter of a scale-free random graph. *Combinatorica*, 24:5–34, 2004.

[25] B. Bollobas et al. The degree-sequence of a scale-free random graph process. *Random Structures and Algorithms*, 18:279–290, 2001.

[26] P. Bonacich. Factoring and weighting approaches to status scores and clique identification. *Journal of Mathematical Sociology*, 2:113–120, 1972.

[27] P. Bonacich. Power and centrality: a family of measures. *American Journal of Sociology*, 92:1170–1182, 1987.

[28] P. Bonacich and P. Lloyd. Eigenvector-like measures of centrality for asymmetric relations. *Social Networks*, 23:191–201, 2001.

[29] S. Borgatti, K. Carley, and D. Krackhardt. On the robustness of centrality measures under conditions of imperfect data. *Social Networks*, 28:124–136, 2006.

[30] F. Brauer and C. Castillo-Chavez. *Mathematical Models in Population Biology and Epidemiology*. Springer-Verlag, 2000.

[31] D. Bray. Molecular networks: the top-down view. *Science*, 301:1864–1865, 2003.

[32] S. Brin and L. Page. The anatomy of a large-scale hypertextual web search engine. *Computer Networks and ISDN Systems*, 30(1-7):107–117, 1998.

[33] D. Bu et al. Topological structure analysis of the protein-protein interaction network in budding yeast. *Nucleic Acids Research*, 31(9):2443–2450, 2003.

[34] J. Buck. Synchronous rhythmic flashing of fireflies, ii. *The Quarterly Review of Biology*, 63(3):265–289, 1988.

[35] A. Capocci et al. Detecting communities in large networks. *Physica A*, 352:669–676, 2005.

[36] F. Chung and L. Lu. Coupling online and offline analyses for random power law graphs. *Internet Mathematics*, 1(4):409–461, 2004.

[37] F. Chung et al. Duplication models for biological networks. *Journal of Computational Biology*, 10(5):677–687, 2003.

[38] A. Clauset and C. Moore. Accuracy and scaling phenomena in Internet mapping. *Physical Review Letters*, 94:018701, 2005.

[39] A. Clauset, M. Newman, and C. Moore. Finding community structure in very large networks. *Physical Review E*, 70:066111, 2004.

[40] V. Colizza et al. Characterization and modeling of protein-protein interaction networks. *Physica A*, 352:1–27, 2005.

[41] G. Conant and A. Wagner. Convergent evolution of gene circuits. *Nature Genetics*, 34(3):264–266, 2003.

[42] E. Costander and T. Valente. The stability of centrality measures when networks are sampled. *Social Networks*, 25:283–307, 2003.

[43] M. Costanza et al. The yeast proteome database (ypd) and caenorhabditis elegans proteome database (wormpd): comprehensive resources for the organization and comparison of model organism protein information. *Nucleic Acids Research*, 28:73–76, 2000.

[44] Thomas M. Cover and Joy A. Thomas. *Elements of Information Theory*. John Wiley and Sons, Inc., 1991.

[45] Z. Dezso and A. Barabasi. Halting viruses in scale-free networks. *Physical Review E*, 65:055103, 2002.

[46] Z. Dezso, Z. Oltvai, and A. Barabasi. Bioinformatics analysis of experimentally determined protein complexes in the yeast saccaromyces cerevisiae. *Genome Research*, 13:2450–2454, 2003.

[47] R. Diestel. *Graph Theory*. Springer-Verlag, 2000.

[48] R. Dobrin et al. Aggregation of topological motifs in the *Escherichia coli* transcriptional regulatory network. *BMC Bioinformatics*, 5:10, 2004.

[49] L. Donetti and M. Munoz. Detecting network communities: a new systematic and efficient algorithm. *http://arXiv:cond-mat/0404652*, 2004.

[50] S. Dorogovstev, J. Mendes, and A. Samukhin. Structure of growing networks with preferential linking. *Physical Review Letters*, 85(21):4633–4636, 2000.

[51] S. Dorogovtsev and J. Mendes. Evolution of networks. *Advances in Physics*, 51:1079–1187, 2002.

[52] R. Dunn, F. Dudbridge, and C. Sanderson. The use of edge-betweenness clustering to investigate biological function in protein interaction networks. *BMC Bioinformatics*, 6:39, 2005.

[53] V. Eguiluz and K. Klemm. Epidemic threshold in structured scale-free networks. *Physical Review Letters*, 89:108701, 2002.

[54] A.K. Engel and W. Singer. Temporal binding and the neural correlates of sensory awareness. *Trends in Cognitive Sciences*, 5(1):16–25, 2001.

[55] A. Enright, S. Van Dongen, and C. Ouzounis. An efficient algorithm for large-scale detection of protein families. *Nucleic Acids Research*, 30(7):1575–1584, 2002.

[56] E. Estrada. Subgraph centrality in complex networks. *Physical Review E*, 71:056103, 2005.

[57] E. Estrada. Virtual identification of essential proteins within the protein interaction network of yeast. *http://arxiv.org/abs/q-bio.MN/0505007*, 2005.

[58] S. Eubank et al. Modelling disease outbreaks in realistic urban social networks. *Nature*, 429:180–184, 2004.

[59] M. Faloutsos, P. Faloutsos, and C. Faloutsos. On power-law relatinships of the Internet topology. In *SIGCOMM*, 1999.

[60] D. Featherstone and K. Broadie. Wrestling with pleiotropy: genomic and topological analysis of the yeast gene expression network. *Bioessays*, 24:267–274, 2002.

[61] S. Fortunato, V. Latora, and M. Marchiori. Method to find community structures based on information centrality. *Physical Review E*, 70:056104, 2004.

[62] E. Fox Keller. Revisiting "scale-free" networks. *Bioessays*, 27:1060–1068, 2005.

[63] H. Fraser et al. Evolutionary rate in the protein interaction network. *Science*, 296:750–752, 2002.

[64] L. Freeman. A set of measures of centrality based on betweenness. *Sociometry*, 40:35–41, 1978.

[65] L. Freeman, S. Borgatti, and D. White. Centrality in valued graphs: a measure of betweenness based on network flow. *Social Networks*, 13:141–154, 1991.

[66] J. Ganesh, L. Massoulie, and D. Towsley. The effect of network topology on the spread of epidemics. In *IEEE Infocom*, 2005.

[67] L. Giot et al. A protein interaction map of drosophila melanogaster. *Science*, 302:1727–1736, 2003.

[68] M. Girvan and M. Newman. Community structure in social and biological networks. *Proceedings of the National Academy of Sciences*, 99(12):7821–7826, 2002.

[69] L. Glass. Synchronization and rhythmic processes in physiology. *Nature*, 410:277–284, 2001.

[70] K. Goh, B. Kahng, and D. Kim. Graph theoretic analysis of protein interaction networks of eukaryotes. *Physica A*, 357:501–512, 2005.

[71] D. Goldberg and F. Roth. Assessing experimentally derived interactions in a small world. *Proceedings of the National Academy of Sciences*, 100(8):4372–4376, 2003.

[72] D. Gomez et al. Centrality and power in social networks: a game theoretic approach. *Mathematical Social Sciences*, 46:27–54, 2003.

[73] B. Gong, L. Yang, and K. Yang. Synchronization on Erdös-Rényi networks. *Physical Review E*, 72:037101, 2005.

[74] N. Guelzim, S. Bottani, P. Bourgine, and F. Kepes. Topological and causal structure of the yeast transriptional regulatory network. *Nature Genetics*, 31:60–63, 2002.

[75] M. Hahn, G. Conant, and A. Wagner. Molecular evolution in large genetic networks: does connectivity equal constraint? *Journal of Molecular Evolution*, 58:203–211, 2004.

[76] J. Han et al. Effect of sampling on topology predictions of protein-protein interaction networks. *Nature Biotechnology*, 23(7):839–844, 2005.

[77] F. Hayot and C. Jayaprakash. A feedforward loop in transcriptional regulation: induction and repression. *Journal of Theoretical Biology*, 234:133–143, 2005.

[78] H. Hethcote. The mathematics of infectious diseases. *SIAM Review*, 42(4):599–653, 2000.

[79] A. Hirsh and H. Fraser. Protein dispensability and rate of evolution. *Nature*, 411:1046–1049, 2001.

[80] A. Hirsh and H. Fraser. Reply to communication on "protein dispensability and rate of evolution". *Nature*, 421:497–498, 2003.

[81] H. Hishigaki al. Assessment of prediction accuracy of protein function from protein-protein interaction data. *Yeast*, 18:523–531, 2001.

[82] P. Holmes, M. Huss, and H. Jeong. Subnetwork hierarchies of biochemical pathways. *Bioinformatics*, 19(4):532–538, 2003.

[83] H. Hong, M.Y. Choi, and B.J. Kim. Synchronization on small-world networks. *Physical Review E*, 65:026139, 2002.

[84] H. Hong, B.J. Kim, M.Y. Choi, and H. Park. Factors that predict better synchronizability on complex networks. *Physical Review E*, 69:067105, 2004.

[85] H. Hong, H. Park, and M.Y. Choi. Collective synchronization in spatially extended systems of coupled oscillators with random frequencies. *Physical Review E*, 72:036217, 2005.

[86] R. Horn and C. Johnson. *Matrix Analysis*. Cambridge University Press, 1985.

[87] J.M. Hurtado, L.L. Rubchinsky, and K.A. Sigvardt. Statistical method for detection of phase-locking episodes in neural oscillations. *Journal of Neurophysiology*, 91:1883–1898, 2004.

[88] D. Hwang et al. Thresholds for epidemic outbreaks in finite scale-free networks. *Mathematical Biosciences and Engineering*, 2(2):317–327, 2005.

[89] T. Ichinomiya. Frequency synchronization in a random oscillator network. *Physical Review E*, 70:026116, 2004.

[90] J. Ihmels et al. Revealing modular organization in the yeast transcriptional network. *Nature Genetics*, 31:370–377, 2002.

[91] A.A. Ioannides, V. Poghosyan, J. Dammers, and M. Streit. Real-time neural activity and connectivity in healthy individuals and schizophrenia patients. *NeuroImage*, 23:473–482, 2004.

[92] T. Ito et al. A comprehensive two-hybrid analysis to explore the yeast protein interactome. *Proceedings of the National Academy of Sciences*, 98(8):4569–4574, 2001.

[93] S. Itzkovitz and U. Alon. Subgraphs and network motifs in geometric networks. *Physical Review E*, 71:026117, 2005.

[94] A. Jadbabaie, N. Motee, and M. Marahona. On the stability of the kuramoto model of coupled nonlinear oscillators. In *Proceedings of the American Control Conference, Boston, Massachusetts*, 2004.

[95] H. Jeong, S. Mason, A. Barabasi, and Z. Oltvai. Lethality and centrality in protein networks. *Nature*, 411:41–42, 2001.

[96] H. Jeong, Z. Oltvai, and A. Barabasi. Prediction of protein essentiality based on genomic data. *ComPlexUs*, 1:19–28, 2003.

[97] H. Jeong et al. The large-scale organization of metabolic networks. *Nature*, 407:651–654, 2000.

[98] I. Jordan et al. Essential genes are more evolutionarily conserved than are nonessential genes in bacteria. *Genome Research*, 12:962–968, 2002.

[99] M. Joy et al. High-betweenness proteins in the yeast protein interaction network. *Journal of Biomedicine and Biotechnology*, 2:96–103, 2005.

[100] P. Peebles Jr. *Probability, Random Variables, and Random Signal Principles*. McGraw-Hill, 2000.

[101] E. R. Kandel, J. H. Schwarz, and T. M. Jessell, editors. *Principles of Neural Science*. McGraw-Hill, 2000.

[102] U. Karaoz et al. Whole-genome annotation by using evidence integration in functional-linkage networks. *Proceedings of the National Academy of Sciences*, 101:2888–2893, 2004.

[103] N. Kashtan et al. Topological generalizations of network motifs. *Physical Review E*, 70:031909:1–12, 2004.

[104] J. Keener and J. Sneyd. *Mathematical Physiology*. Interdisciplinary Applied Mathematics. Springer-Verlag, New York, Inc., 1998.

[105] J. Kleinberg. Authoritative sources in a hyperlinked environment. In *9th ACM-SIAM Symposium on Discrete Algorithms*, 1998.

[106] P. Krapivsky, S. Redner, and F. Leyvraz. Connectivity of growing random networks. *Physical Review Letters*, 85(21):4629–4632, 2000.

[107] Y. Kuramoto. In H. Arakai, editor, *International Symposium on Mathematical Problems in Theoretical Physics*, volume 39 of *Lecture Notes in Physics*. Springer, New York, 1975.

[108] Y. Kuramoto and I. Nishikawa. Statistical macrodynamics of large dynamical systems: case of a phase transition in oscillator communities. *Journal of Statistical Physics*, 49:569–605, 1987.

[109] J.-P. Lachaux, E. Rodriguez, J. Martinerie, and F.J. Varela. Measuring phase synchrony in brain signals. *Human Brain Mapping*, 8:194–208, 1999.

[110] G. Lahav et al. Dynamics of the p53-mdm2 feednack loop in individual cells. *Nature Genetics*, 36(2):147–150, 2004.

[111] A. Langville and C. Meyer. A survey of eigenvector methods for web information retrieval. *SIAM Review*, 47(1):135–161, 2005.

[112] M. Lappe and L. Holm. Unraveling protein interaction networks with near-optimal efficiency. *Nature Biotechnology*, 22(1):98–103, 2004.

[113] V. Latora and M. Marchiori. A measure of centrality based on the network efficiency. *http://arxiv.org/abs/cond-mat/0402050*, 2004.

[114] T. Lee et al. Transcriptional regulatory networks in saccharomyces cerevisiae. *Science*, 298:799–804, 2002.

[115] N. Lemke et al. Essentiality and damage in metabolic networks. *Bioinformatics*, 20(1):115–119, 2004.

[116] S. Letovsky and S. Kasif. Predicting protein function from protein/protein interaction data: a probabilistic approach. *Bioinformatics*, 19:i197–i204, 2003.

[117] S. Li et al. A map of the interactome network of the metazoan *C. elegans*. *Science*, 303:540–543, 2004.

[118] F. Liljeros et al. The web of human sexual contacts. *Nature*, 411:907, 2001.

[119] H. Ma, J. Buer, and A. Zeng. Hierarchical structure and modules in the *Escherichia coli* transcriptional regulatory network revealed by a new top-down approach. *BMC Bioinformatics*, 5:199, 2004.

[120] S. Mangan and U. Alon. Structure and function of the feed-forward loop network motif. *Proceedings of the National Academy of Sciences*, 100(21):11980–11985, 2003.

[121] S. Mangan, A. Zaslaver, and U. Alon. The coherent feedforward loop serves as a sign-sensitive delay element in transcription networks. *Journal of Molecular Biology*, 334:197–204, 2003.

[122] S. Maslov and K. Sneppen. Specificity and stability in topology of protein networks. *Science*, 296:910–913, 2002.

[123] R.M. May and A.L. Lloyd. Infection dynamics on scale-free networks. *Physical Review E*, 64:066112, 2001.

[124] P.N. McGraw and M. Metzinger. Clustering and the synchronization of oscillator networks. *Physical Review E*, 72, 2005.

[125] H. Mewes et al. MIPS: a database for genomes and protein sequences. *Nucleic Acids Research*, 30(1):31–34, 2002.

[126] R. Milo et al. Network motifs: simple building blocks of complex networks. *Science*, 298:824–827, 2002.

[127] R. Milo et al. Response to comment on "Network motifs, simple building blocks of complex networks" and "Superfamilies of evolved and designed networks". *Science*, 305:1107d, 2004.

[128] R. Milo et al. Superfamilies of evolved and designed networks. *Science*, 303:1538–1542, 2004.

[129] Y. Moreno, J. Gomez, and A. Pacheco. Epidemic incidence in correlated complex networks. *Physical Review E*, 68:035103, 2003.

[130] J. D. Murray. *Mathematical Biology, Volume 1.* Springer-Verlag, 2002.

[131] E. Nabieva et al. Whole-proteome prediction of protein function via graph-theoretic analysis of interaction maps. *Bioinformatics*, 21:i302–i310, 2005.

[132] M. Newman. Spread of epidemic disease on networks. *Physical Review E*, 66:016128, 2002.

[133] M. Newman. The structure and function of complex networks. *SIAM Review*, 45(2):167–256, 2003.

[134] M. Newman. Detecting community structure in networks. *European Physics Journal B*, 38:321–330, 2004.

[135] M. Newman. Fast algorithm for detecting community structure in networks. *Physical Review E*, 69:066133, 2004.

[136] M. Newman. A measure of betweenness centrality based on random walks. *Social Networks*, 27:39–54, 2005.

[137] M. Newman and M. Girvan. Finding and evaluating community structure in networks. *Physical Review E*, 69:026113, 2004.

[138] T. Nishikawa, A.E. Motter, Y.-C. Lai, and F.C. Hoppensteadt. Heterogeneity in oscillator networks: Are smaller worlds easier to synchronize? *Physical Review Letters*, 91:014101, 2003.

[139] B.F. O'Donnell, M.A. Wilt, C.A. Brenner, T.A. Busey, and J.S. Kwon. EEG synchronization deficits in schizophrenia spectrum disorders. *International Congress Series*, 1232:697–703, 2002.

[140] R. Overbeek et al. Wit: integrated system for high-throughput genome sequence analysis and metabolic reconstruction. *Nucleic Acids Research*, 28(1):123–125, 2000.

[141] C. Pal, B. Papp, and D. Hurst. Communication on "protein dispensability and rate of evolution". *Nature*, 421:496–497, 2003.

[142] R. Pastor-Satorras and A. Vespignani. Epidemic spreading in scale-free networks. *Physical Review Letters*, 86(14):3200–3203, 2001.

[143] R. Pastor-Satorras and A. Vespignani. Epidemic dynamics in finite size scale-free networks. *Physical Review E*, 65:035108, 2002.

[144] R. Pastor-Satorras and A. Vespignani. Immunization of complex networks. *Physical Review E*, 65:036104, 2002.

[145] M. Penrose. *Geometric Random Graphs*. Oxford University Press, 2003.

[146] J. Pereira-Leal, A. Enright, and C. Ouzounis. Detection of functional modules from protein interaction networks. *PROTEINS: Structure, Function and Bioinformatics*, 54:49–57, 2004.

[147] A. Pikovsky, M. Rosenblum, and J. Kurths. *Synchronization: a universal concept in nonlinear sciences*, volume 12 of *Cambridge Nonlinear Sciences*. Cambridge University Press, 2001.

[148] N. Przulj, D. Corneil, and I. Jurisica. Modeling interactome: scale-free or geometric. *Bioinformatics*, 20(18):3508–3515, 2004.

[149] N. Przulj, D. Wigle, and I. Jurisica. Functional topology in a network of protein interactions. *Bioinformatics*, 20(3):340–348, 2004.

[150] F. Radicchi et al. Defining and identifying communities in networks. *Proceedings of the National Academy of Sciences*, 101:2658–2663, 2004.

[151] J. Rain et al. The protein-protein interaction map of Heliobacter Pylori. *Nature*, 409:211–215, 2001.

[152] E. Ravasz et al. Hierarchical organization of modularity in metabolic networks. *Science*, 297:1551–1555, 2002.

[153] J. Read and M. Keeling. Disease evolution on networks: the role of contact structure. *Proceedings of the Royal Society - B*, 270:699–708, 2003.

[154] J.G. Restrepo, E. Ott, and B.R. Hunt. Onset of synchronization in large networks of coupled oscillators. *Physical Review E*, 71:036151, 2005.

[155] E. Rodriguez, N. George, J.-P. Lachaux, J. Martinerie, B. Renault, and F. Varela. Perception's shadow: long-distance synchronization of human brain activity. *Nature*, 397:430–433, 1999.

[156] P.R. Roelfsema, A.K. Engel, P. König, and W. Singer. Visuomotor integration is associated with zero time-lag synchronization among cortical areas. *Nature*, 385:157–161, 1997.

[157] N. Rosenfeld, M. Elowitz, and U. Alon. Negative autoregulation speeds the response times of transcription networks. *Journal of Molecular Biology*, 323:785–793, 2002.

[158] M. Samanta and S. Liang. Predicting protein functions from redundancies in large-scale protein interaction networks. *Proceedings of the National Academy of Sciences*, 100(22):12579–12583, 2003.

[159] J. Saramaki and K. Kaski. Modeling development of epidemics with dynamic small-world networks. *Journal of Theoretical Biology*, 234:413–421, 2005.

[160] A. Sawa and S.H. Snyder. Schizophrenia: diverse approaches to a complex disease. *Science*, 296:692–695, 2002.

[161] J. Schmith et al. Damage, connectivity and essentiality in protein-protein interaction networks. *Physica A*, 349:675–684, 2005.

[162] A. Schnitzler and J. Gross. Normal and pathological oscillatory communication in the brain. *Nature Reviews - Neuroscience*, 6:285–295, 2005.

[163] J. Scholz, M. Dejori, M. Stetter, and M. Greiner. Noisy scale-free networks. *Physica A*, 350:622–642, 2005.

[164] B. Schwikowski, P. Uetz, and S. Fields. A network of protein-protein interactions in yeast. *Nature Biotechnology*, 18:1257–1261, 2000.

[165] E. Segal et al. Genome-wide discovery of transcriptional modules from DNA sequence and gene expression. *Bioinformatics*, 19(Supp 1):i273–i282, 2003.

[166] E. Segal et al. Module networks: identifying regulatory modules and their condition-specific regulators from gene expression data. *Nature Genetics*, 34(2):166–176, 2003.

[167] D. Segre, A. De Luna, G. Church, and R. Kishony. Modular epistasis in yeast metabolism. *Nature Genetics*, 37(1):77–83, 2005.

[168] T. Seidenbecher, T. Rao Laxmi, O. Stork, and H.-C. Pape. Amygdalar and hippocampal theta rhythm synchronization during memory retrieval. *Science*, 301:846–850, 2003.

[169] S. Shen-Orr, R. Milo, S. Mangan, and U. Alon. Network motifs in the transcriptional regulatory network of escherichia coli. *Nature Genetics*, 31:64–68, 2002.

[170] R. Sole et al. A model of large scale proteome evolution. *Advances in Complex Systems*, 5:43–54, 2002.

[171] K.M. Spencer, P.G. Nestor, M.A. Niznikiewicz, D.F. Salisbury, M.E. Shenton, and R.W. McCarley. Abnormal neural synchrony in schizophrenia. *Journal of Neuroscience*, 23(19):7407–7411, 2003.

[172] K. Stephenson and M. Zelen. Rethinking centrality: methods and examples. *Social Networks*, 11:1–37, 1989.

[173] S.H. Strogatz. From Kuramoto to Crawford: exploring the onset of synchronization in populations of coupled oscillators. *Physica D*, 143:1–20, 2000.

[174] S.H. Strogatz. Arthur Winfree (obituary). *SIAM news*, 36(1), 2003.

[175] M. Stumpf, C. Wiuf, and R. May. Subnets of scale-free networks are not scale-free: Sampling properties of networks. *Proceedings of the National Academy of Sciences*, 102(12):4221–4224, 2005.

[176] A. Thomas et al. On the structure of protein-protein interaction networks. *Transactions of the Biochemical Society*, 31(6):1491–1496, 2003.

[177] A. Tong et al. Global mapping of the yeast genetic interaction network. *Science*, 303:808–813, 2004.

[178] P. Uetz et al. A comprehensive analysis of protein-protein interactions in Saccharomyces cerevisiae. *Nature*, 403:623–627, 2000.

[179] F. Varela, J.-P. Lachaux, E. Rodriguez, and J. Martinerie. The brainweb: phase synchronization and large-scale integration. *Nature Reviews Neuroscience*, 2:229–239, 2001.

[180] A. Vazquez et al. Global protein function prediction from protein-protein interaction networks. *Nature Biotechnology*, 21(6):697–700, 2003.

[181] A. Vazquez et al. Modeling of protein interaction networks. *ComPlexUs*, 1:38–46, 2003.

[182] Y.M. Moreno Vega, M. Vàsquez-Prada, and A.F. Pacheco. Fitness for synchronization of network motifs. *Physica A*, 343:279–287, 2004.

[183] J. Verdasca et al. Recurrent epidemics in small-world networks. *Journal of Theoretical Biology*, 233:553–561, 2005.

[184] B. Vogelstein, D. Lane, and A. Levine. Surfing the p53 network. *Nature*, 408:307–310, 2000.

[185] D. Volchenkov, L. Volchenkova, and Ph. Blanchard. Epidemic spreading in a variety of scale-free networks. *Physical Review E*, 66:046137, 2002.

[186] C. Von Mering et al. Comparative assessment of large-scale data sets of protein-protein interactions. *Nature*, 417:399–403, 2002.

[187] A. Wagner. The yeast protein interaction network evolves rapidly and contains few redundant duplicate genes. *Molecular Biology and Evolution*, 18(7):1283–1292, 2001.

[188] A. Wagner. How the global structure of protein interaction networks evolves. *Proceedings of the Royal Society - B*, 270:457–466, 2002.

[189] A. Wagner and D. Fell. The small world inside large metabolic networks. *Proceedings of the Royal Society - B*, 268:1803–1810, 2001.

[190] W. Wang and S. Ruan. Simulating the SARS outbreak in Beijing with limited data. *Journal of Theoretical Biology*, 227:369–379, 2004.

[191] S. Wasserman and K. Faust. *Social Network Analysis: Methods and Applications*. Cambridge University Press, 1994.

[192] D. Watts and S. Strogatz. Collective dynamics of small-world networks. *Nature*, 393:440–442, 1998.

[193] A.T. Winfree. Biological rhythms and the behavior of populations of coupled oscillators. *Journal of Theoretical Biology*, 16:15–42, 1967.

[194] S. Wuchty. Interaction and domain networks of yeast. *Proteomics*, 2:1715–1723, 2002.

[195] S. Wuchty, Z. Oltvai, and A. Barabasi. Evolutionary conservation of motif constituents in the yeast protein interaction network. *Nature Genetics*, 35(2):176–179, 2003.

[196] S. Wuchty and P. Stadler. Centers of complex networks. *Journal of Theoretical Biology*, 223:45–53, 2003.

[197] I. Xenarios et al. DIP: the database of interacting proteins. *Nucleic Acids Research*, 28(1):289–291, 2000.

[198] J. Yang, Z. Gu, and W. Li. Rate of protein evolution versus fitness effect of gene deletion. *Molecular Biology and Evolution*, 20(5):772–774, 2003.

[199] E. Yeger-Lotem et al. Network motifs in integrated cellular networks of transcription-regulation and protein-protein interaction. *Proceedings of the National Academy of Sciences*, 101(16):5934–5939, 2004.

[200] S. Yook, Z. Oltvai, and A. Barabasi. Functional and topological characterization of protein interaction networks. *Proteomics*, 4:928–942, 2004.

[201] H. Yu et al. Genomic analysis of essentiality within protein networks. *Trends in Genetics*, 20(6):227–231, 2004.

[202] B. Zemljic and V. Hleben. Reliability of measures of centrality and prominence. *Social Networks*, 27:73–88, 2005.

[203] L. Zhang et al. Motifs, themes and thematic maps of an integrated saccharomyces cerevisiae interaction network. *BMC Journal of Biology*, 4:6, 2005.

[204] E. Ziv, M. Middendorf, and C. Wiggins. An information-theoretic approach to network modularity. *Physical Review E*, 71:046117, 2005.

[205] E. Ziv et al. Systematic identification of statistically significant network measures. *Physical Review E*, 71:016110:1–8, 2005.