Using physiological system networks to elaborate resilience across frailty states

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Abstract

Background: Aging is characterized by loss of resilience, the ability to resist or recover from stressors. Network analysis has shown promise in investigating dynamic relationships underlying resilience. We aimed to use network analysis to measure resilience in a longitudinal cohort of older adults and quantify whole-system vulnerabilities associated with frailty.

Methods: We used data from the Rugao Longitudinal Ageing Study, including 71 biomarkers from participants classified as robust, prefrail, or frail. We quantified biomarker correlations and topological parameters. Additionally, we proposed propagation models to simulate damage and recovery dynamics, investigating network resilience under various conditions.

Results: We classified 1754 individuals into robust (n=369), prefrail (n=1103), and frail (n=282) groups with 71 biomarkers. Several biomarkers were linked to frailty, including those related to blood pressure, ECG, kidney function, platelets, white blood cells. Each frailty stage was associated with increased network correlations. The frail network showed increased average degree and connectance, decreased average path length and diameter, and reduced modularity compared to robust and prefrail networks. Hub biomarkers, particularly β2-microglobulin and platelet count, played a significant role, potentially propagating dysfunction across physiological systems. Simulations revealed that damage to critical hubs led to longer recovery times in the frail network than robust and prefrail networks.

Conclusion: Network analysis could serve as a valuable tool for quantifying resilience and identifying vulnerabilities in older adults with frailty. Our findings contribute to understanding frailty-related physiological disturbances and offer potential for personalized healthcare interventions targeting resilience in older populations.

Keywords : Frailty, resilience, complex system, phenotype network, aging

Introduction

Older age is characterized by multimorbidity and frailty, with impairments across physiological, cognitive and physical capabilities (1,2). Resilience is the ability to resist or recover from the effects of a stressor, along a continuum from health to frailty (**Figure 1A**) (3,4,5). Quantifying resilience is essential if we are to target it as a therapeutic strategy to prevent adverse outcomes, disability, and mortality (6,7). Dynamical systems physiology is an attempt to bring together biological substrates linking resilience and frailty (8).

The measurement of resilience has been evolving. First, static indicators, such as survival following the onset of poor health (9) or the residual differences between age and frailty (distinguishing *adapters* and *prematurely frail*) (10), were used. While useful for population research, this approach is insufficient to capture any temporal dynamics. Secondly, stimulus-response experiments have been conducted to quantify resilience, including glucose tolerance tests (11), isometric exercises (12) and adrenocorticotropic hormone stimulation tests (13). These characterize resilience in an individual, particularly for specific organ systems, though they are more challenging to apply at scale. Thirdly, dynamical indicators of resilience (DIOR) based on variance and temporal autocorrelation have been applied to time-series data (5,14,15). In particular, longitudinal DIOR analysis of blood markers predicted mortality corresponding to a complete loss of resilience and frailty (14). Building on these ideas, establishing resilience operationally will lead to new insights into underlying causes of frailty, its treatment and prevention.

Network analysis has great potential to uncover some dynamic relationships underpinning resilience (16). Fundamentally, the technique starts by quantifying the correlations between a

range of physiological biomarkers (14,17). These conceptualize biomarkers as nodes, connected by any number of degrees (18,19,20). The number, strength, and paths throughout the network conform to topological parameters, and these characteristics can indicate network resilience (21). Networks with low resilience have mutually dependent increases in cross-correlations, making them vulnerable to whole-system failure (22,23). A general property of resilient networks is that they have scale-free attributes: the relationship between strongly connected hubs is exponentially related to the number of nodes with fewer connections regardless of network size (**Figure 1B**). As complex networks lose resilience, local clustering may become evident, and scale-free characteristics might start to reduce (**Figure 1B**). We recently applied this approach to describe internal correlation structures of physiological biomarkers underlying aging (24).

We set out to apply network analysis to data from a longitudinal study in which physiological biomarkers had been obtained in participants classified with different degrees of frailty. We wanted to quantify the number and degree of cross-correlations, any differences in the resulting network topology, and then simulate the effect of damage at any set of points in the network, and predict the potential for damage propagation or whole-system recovery under a range of conditions.

Methods

Study population and participants

The Rugao Longitudinal Ageing Study (RLAS) is a population-based study from Rugao, Jiangsu Province, China. Since 2014, the aging arm of RLAS has been following n=1788 participants aged 70-84 from a medium-sized township in Rugao (91% of the eligible population), collecting demographic, clinical, and laboratory measures (**Supplementary File 1)**. Waves 2, 3, and 4 (2016, 2017, 2019 respectively) collected data on frailty. Wave 4 also measured 71 biomarkers, classified into 17 physiological systems: anthropometry, blood pressure, electrocardiographic, lipids, glucose, endocrine, venous blood gas, electrolytes, inflammation, vascular function, liver function, heart function, kidney function, platelets, white blood cells, red blood cells, and reticulocytes (**Table S1**). Our principal cross-sectional analyses are at Wave 4: biomarker networks for each frailty category. Longitudinal analyses were for transitions between frailty states from Waves 2, 3, and 4 in relation to the biomarker networks derived at Wave 4 (details below).

Operationalization of frailty

The frailty phenotype was defined using five features: weight loss, exhaustion, low activity, weakness, and slowness. Weight loss, exhaustion and low activity were self-reported (*Have you lost more than 4.5 kg or 5% of your body weight in the past 12 months? Have you felt tired at least 3 or 4 days per week? Do you need help to walk?*). Weakness was defined as being <20th centile for Chinese adult population-standard norms in maximum handgrip strength using a dynamometer for three trials of each hand, or those unable to be assessed. Participants in the worst 20th centile for Chinese adult population-standard norms of the timed-up-and-go (taking

the longest time), or those who could not complete the task were categorized as having slowness. (**Supplementary File 1**) (25). Participants with three or more features were defined as *frail*, one or two as *prefrail* and none as *robust*.

Construction of biomarker networks

To create a network between pairs of biomarker nodes, we calculated the maximum information coefficient (MIC) using the R package *minerva* (26). Compared with Pearson, Spearman and Kendall correlation, the MIC could capture both linear and nonlinear correlation between biomarkers (24). Hubs are nodes with relatively more connections (degrees); these are not defined in absolute terms, and their identification is data-driven. To make the networks comparable, so we took equal random samples from each robust, prefrail, and frail group; across Waves 2 to 4, n=150, 150, 200 corresponded to the smallest, size-limiting group. We used the mean MIC values calculated separately for men and women. Subsampling was performed 100 times; we used the median of resampled MIC values for the final network. We applied hard thresholds from random matrices theory (RMT), effective at pruning networks, to filter out noise and spurious correlations. As hard thresholds of MIC can affect the number of edges in networks, the same procedures and thresholds were used for all three groups.

Network topology

Networks are described by several topological parameters (**Figure 1B**). These include: average degree, connectance, diameters, average path length, centralization betweenness, centralization degree, modularity, and clustering coefficient. In addition, we considered scale-free and small-world (local clustering) attributes.

Propagation models

We proposed propagation models to simulate the dynamics of spread on complex networks. Each node had two statuses: healthy or damaged. To simulate the perturbation caused by a stressor or stimulus, a proportion of nodes (α) were randomly set to be damaged. Damaged nodes could recover with fixed probability (u), and healthy nodes could become damaged due to connections with damaged nodes (r). To quantify network resilience, we simulated random damage to nodes and hubs, using the unit time for 50% of damaged nodes to recover (RT50) as the indicator. We considered four scenarios: where in respect of the damage rate (u), the recovery rate (r) was equal (Scenario 1), less (Scenario 2), or greater (Scenario 3). Scenario 4 fixed the recovery rate to decrease from robust to frail status (**Supplementary File 3)**.

Statistical analysis

Data analysis was conducted in R (version 4.0). We used *igraph* to visualize the network structure and compute the network topological parameters. To investigate parameter differences between *robust*, *prefrail* and *frail* networks (variable importance in projection, VIP value), we used partial least-squares discriminant analysis (PLS-DA, R package *ropls*), adjusted by age, sex ratio, education, smoking status and alcohol use.

Results

Disturbances of networks in frailty: increased correlation between biomarkers

We were able to classify 1754 (98%) individuals in RLAS Wave 4 into robust (n=369), prefrail (n=1103) and frail (n=282) groups (**Table 1**). Many of the 71 biomarkers were associated with frailty (**Table S1**). Specifically, blood pressure (pulse pressure), ECG (dominant R wave in V5), kidney function (β2 microglobulin, cystatin C), platelets (plateletcrit, count), white blood cells

(monocyte count, neutrophil %, total leukocytes) were associated with the frailty phenotype. There were more abnormal biomarkers, with increasing MIC, across the stages of Wave 4 frailty (**Figure S1A**). From Wave 3 to Wave 4, individuals remaining robust (n=128) also had lower MIC compared with individuals who transitioned from non-frail into frail states (n=152) (**Figure S1B**); differences in MIC distributions of frail networks were already apparent at Wave 2, 3 clinical assessments (**Figure S1C-D**).

The networks for robust, prefrail and frail groups were filtered at a MIC threshold set at 0.28, determined by random matrix theory (**Figure S1E**). There were 125, 116 and 186 edges in the networks respectively, resulting from increased correlations between biomarkers in the frail group. (**Figure 2**). Correlations that were notably high were between: B-type natriuretic peptide (BNP, Heart function) and alanine aminotransferase (ALT, Liver function), folate (FOL, Vascular) and cystatin C (Cys.C, Kidney), high fluorescence reticulocyte (HFR, Reticulocyte) and white blood cell count (White Blood Cells), β2-microglobulin (β2.MG, Kidney function) and hematocrit (HCT, Red Blood Cells) (**Figure S2**).

Topological differences in networks across frailty status

Compared with networks of robust and prefrail groups, the average degree and connectance of the network in frail group increased while the average path length and diameter decreased (**Figure 3A**). This was driven by more connections in the frail network. The modularity also decreased in this network, suggesting that the greater number of connections in the frail network were made across physiological systems (**Figure 3A, Figure 2**). Centralization betweenness

(brokered connections) and centralization degree (direct connections) are measures of how critical certain nodes are in a network; the centralization degree slightly increased in the frail group while centralization betweenness decreased. Moreover, we evaluated small-world coefficients (σ) that described higher clustering coefficients and shorter average path lengths indicative of local clustering compared to a random network model (**Supplementary File 2**). The σ was 2.94, 2.80 and 1.74 in robust, prefrail and frail groups, respectively. In general, networks also had scale-free attributes where biomarker connections were characterized by a few but significant number of nodes (hubs), with an exponential decay of number of nodes with fewer connections (power-law distribution). However, this feature decreased from robust to frail groups, indicating frail networks would have more vulnerability to random failures (**Figure 3B**).

Disturbances in frail networks: emerging dysfunctional hubs

We focused on hubs with many connections; in keeping with scale-free attributes, most biomarkers had few connections (**Figure S3A**). The top three hub biomarkers were white blood cell count (WBC), triglyceride (TG), and uric acid (UA) in the robust group. They were body mass index (BMI), lymphocyte count (LY#) and QT interval (QT) in the prefrail group. Compared with the robust and prefrail group, the hub biomarkers such as β2-microglobulin (β2.MG), platelet count (PLT), hematocrit (HCT), B-type natriuretic peptide (BNP) and red blood cell distribution width (RDW) had increasing degree or betweenness centrality in the frail group (**Figure S3B**). In addition, the emerging hubs, such as β2.MG and PLT, were identified by PLSA-DA as being associated with frailty. The degree centrality of these biomarkers (**Figure**

S3C) was significantly correlated with VIP value from discriminant analysis (r=0.31, p=0.016), while the correlations were not significant in robust and prefrail groups. These changes in network centrality suggested that hub biomarkers were dysfunctional, and could propagate through interactions between physiological systems underlying frailty.

Simulations in frail networks lead to longer recovery

Testing the effect of network damage under different scenarios, we found random/hub damage led to longer recovery times in the frail network (**Figure S4**). Under conditions where the frail network had a higher damage rate, the RT50 recovery times were 19.5, 27.0 and 40.5 for robust, prefrail and frail networks respectively (**Supplemetary File 3**). When damage occurred in critical hubs, recovery time of the frail network was also greater than the other groups (**Figure 4**). Compared with random damage, the frail network was more vulnerable to hub damage. In this context, the median differences of RT50 were 1.5, 2.5, and 6 for robust, prefrail and frail groups.

We simulated the impact of different ratios of node recovery and damage (r/u) on the RT50 recovery rate. For random, compared with hub damage, the frail network had shorter RT50 with different ratios of recovery and damage rate (**Figure S4D**). With decreasing recovery : damage ratios, RT50 increased nonlinearly from robust to frail networks, and RT50 was even longer when critical hubs were damaged in the frail network (**Figure S4D**).

Discussion

Compared with robust and prefrail groups, the frail group demonstrated increased correlations and connections between biomarker networks, indicating enhanced dependence between physiological systems underlying frailty. Frail networks also showed topological disturbances, and dysfunctional hubs, reflecting a transition from modular organization to random and chaotic states. Dynamic simulations suggested the frail network had impaired ability to recover. These findings suggest that the collapse of nodes in the network of the frail group is more likely to lead to a loss of resilience for the entire system. Taken together, we showed network analysis can be usefully applied to longitudinal cohort data to give inferences into how to quantify whole-system vulnerability.

Our demonstration of increased cross-correlations in frailty, between organ (e.g., kidney, heart) and regulatory (e.g., autonomic, endocrine, immune) systems directly quantifies interconnections at risk (7,27,28,29). For example, excessive inflammatory cytokines could impact hypothalamic-pituitary axis activity and skeletal muscle metabolism after disruptive stressor events (3). Network topological changes, their predicted propagation dynamics, and their dependence on critical hubs suggest ways organ systems may become synergistically impaired (24,28) and how the number of dysregulated systems become exponentially associated with frailty (30). These emerging dysfunctional hub nodes, including β2-microglobulin (β2.MG) and platelet (PLT), could facilitate our understanding of the mechanisms of resilience and frailty. Previous studies have found that serum β2.MG was significantly associated with frailty (31,32,33). While these findings need to be replicated in other cohorts, the broader point

is that network analysis offers an opportunity to delineate potential vulnerabilities, and what might account for intra-individual and inter-individual variation.

A similar network model has been proposed in frailty, using theoretical deficits in the frailty index to explain vulnerability to damage propagation (20,34). Node damage and recovery rates were allowed to be dynamic and age-dependent, reflecting that prospective cohorts show differences in the rate at which subsystems and their interactions age (35). Our model was simplified with three parameters: the proportion of damage nodes, recovery rate and damage rate and could measure resilience as a recovery time metric. Our study extends this network approach, showing it could be practicable and efficient for cross-sectional data. While previous studies have reported increased connections on biomarker networks in diseases and frailty (22,36,37), we go beyond these observations to show the utility for networks to describe vulnerabilities in damage propagation.

There were some study limitations. Although 71 biomarkers were measured, these may not provide a comprehensive assessment of physiology and some important systems, such as autonomic nervous systems, were not well covered. The simulation analysis implemented a simple propagation model based on the damage onset and recovery assumptions, which may not reflect the true dynamics. Therefore, further work is needed to investigate the dynamic interactions between physiological subsystems. Considering the limited sample size of the RLAS, it would be necessary to validate the network analysis of resilience across frail states in other large cohorts. In addition, we used available physiological markers, but multiscale 'omics data might have given better mechanistic insights into frailty and resilience. Furthermore,

continuously collected data through wearable sensor devices or high-frequency longitudinal phenotyping would likely refine our models (4,17). Other simulation approaches have promising applications in human aging, such as predicting tipping points at which the system shifts abruptly from one state to another, though this requires detailed measurement of both stressor and any response (4,38).

In summary, we investigated the disturbances of physiological systems in frailty through network analysis and evaluated its effects on resilience. Specifically, we found that network disturbances through dynamic simulations were more apparent in the frail group. These results facilitate our understanding of resilience frailty from a complex network perspective. The approach provides insights into quantifying resilience at clinical and population levels, with the potential to promote personalized healthcare for older people (3,39,40).

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Conflict of interest

The authors declare that they have no relevant interests.

Compliance with ethical standards

Ethical approval This study was approved by the Human Ethics Committee of the School of Life Sciences of Fudan University. Informed consent was obtained from each participant.

Data Availability

The datasets for analysis in this study are available from the corresponding author on reasonable request.

Informed consent

Written informed consent was obtained from all participants prior to the study.

Author Contributions

Hao M, Zhang H, Li Y, Jin L, Wang XF: designed the research; Hao M, Zhang H, Li Y: conducted the research; Zhang H, Jiang XY, Sun XH, Wang JC collected the data; Hao M, Zhang H: developed the analysis plan and undertook the computational analyses. Hao M, Davis D, Zhang H: interpreted the data and wrote the first draft; Hao M, Jin L and Wang XF: had primary responsibility for the final content; and all authors read and approved the final manuscript.

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Table 1. Characteristics of study population stratified by frail status.

Demographic		Robust	Prefrail	Frailty	P-value
Sample Size	Wave 4 (2019)	369 (21.04)	1103 (62.89)	282 (16.08)	
(N, %)	Wave 3 (2017)	584 (31.91)	1044 (57.04)	202 (11.04)	
	Wave 2 (2016)	633 (36.28)	936 (53.64)	176 (10.09)	
Age	Wave 4	77.61 ± 4.43	78.56 ± 4.81	80.22 ± 4.74	< 0.001
M ± SD, years	Wave 3	76.92 ± 4.14	78.39 ± 4.48	79.38 ± 4.60	
	Wave 2	76.87 ± 4.34	77.04 ± 4.41	77.84 ± 4.42	
Gender	Wave 4	229 (62.06)	485 (43.97)	76 (26.95)	< 0.001
Male, N (%)	Wave 3	333 (57.02)	454 (43.49)	65 (32.18)	
	Wave 2	280 (44.23)	456 (48.72)	84 (47.73)	
Smoking ǂ	Wave 4	128 (34.69)	262 (23.75)	32 (11.35)	< 0.001
Yes, N (%)	Wave 3	138 (23.63)	193 (18.49)	35 (17.33)	
	Wave 2	136 (21.48)	257 (27.46)	44 (25.00)	
Alcohol Intake ǂ	Wave 4	172 (46.61)	395 (32.55)	51 (18.09)	< 0.001
Yes, N (%)	Wave 3	228 (39.04)	293 (28.07)	35 (17.33)	
	Wave 2	194 (30.65)	311 (33.23)	64 (36.36)	
Educational status ǂ	Wave 4	97 (26.29)	515 (46.69)	172 (60.99)	< 0.001
Illiteracy, N (%)	Wave 3	212 (36.30)	531 (50.86)	132 (65.35)	
	Wave 2	322 (50.87)	465 (49.68)	91 (51.70)	
Marital status ǂ	Wave 4	236 (63.96)	714 (64.73)	159 (56.38)	0.451
Married, N (%)	Wave 3	385 (60.82)	657 (62.93)	131 (64.85)	
	Wave 2	394 (62.24)	608 (64.96)	116 (65.91)	

M: mean, SD: standard deviation. Continuous and categorical variables were present as mean with SD and frequency (%), Group difference were analyzed by chi-square or ANOVA test. Marital status & including separated, divorced, never married or widowed.

Figure legends:

Figure 1. Human ageing processes with declined functions and decreased resilience on the ageing continuum of physiological reserve (A). The physiological system transited into frailty after the critical threshold and lost the capacity to cope with stressors. The topology of complex networks (B), including degree, connectance, diameters, betweenness, modularity, clustering coefficient, scale-free and small-world attributes (Supplementary File 2).

Figure 2. Phenotype networks of robust, prefrail and frail groups.

Figure 3. The topological parameters of phenotype networks (sample size = 100) in robust, prefrail and frail groups (A). The linear correlations between log frequency and log degree indicated scale-free attributes of the networks (B).

Figure 4. A representative example of recovery processes of phenotype networks in damage simulation for robust, prefrail and frail groups (hub damage, Scenario 4).

Figure 1

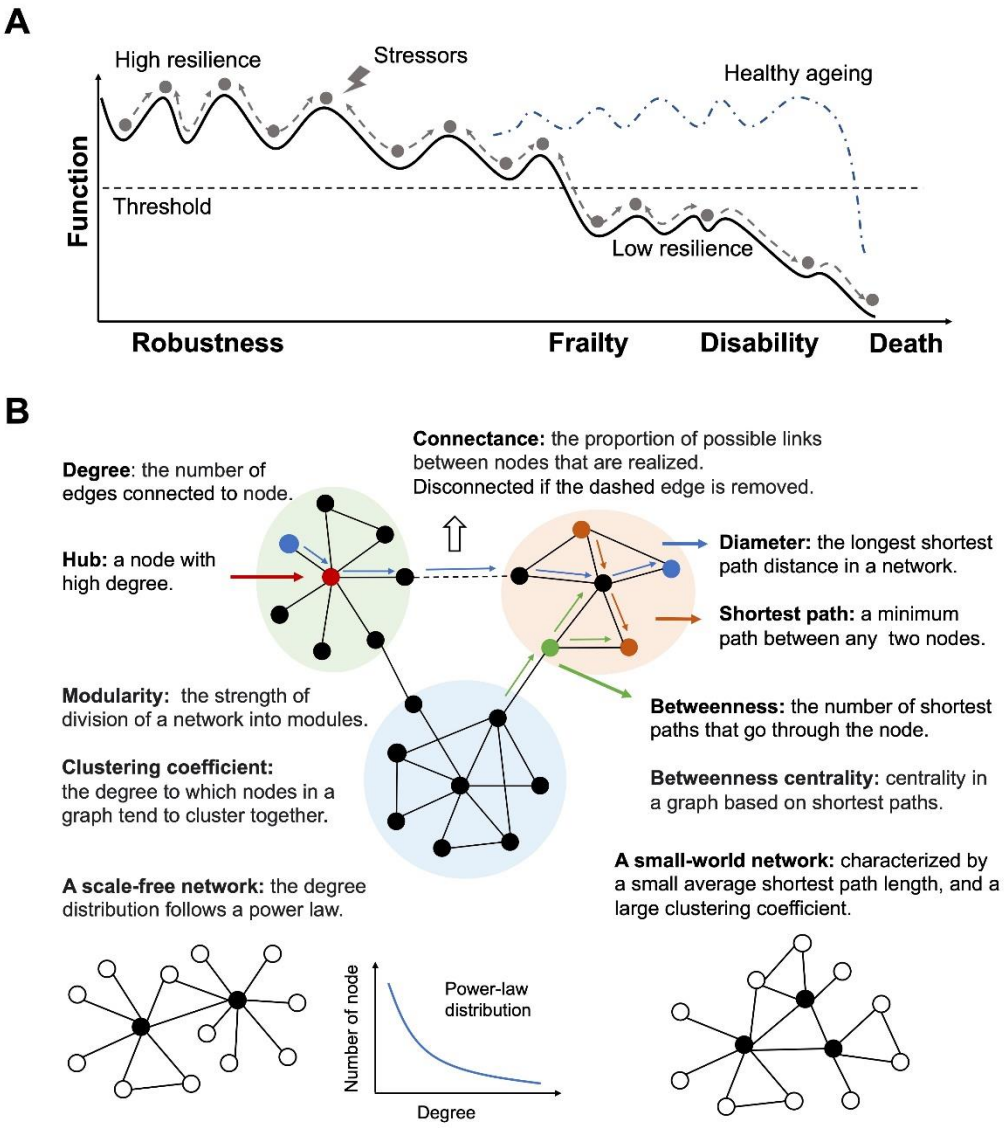

Figure 2

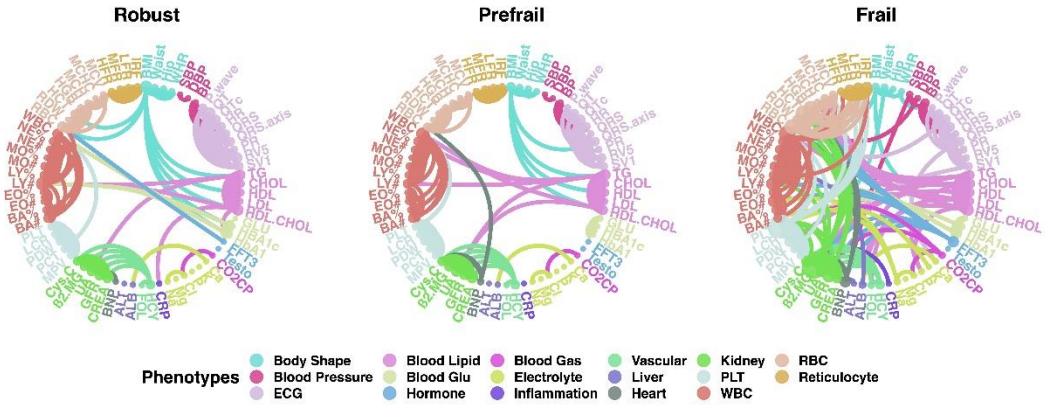

Figure 3

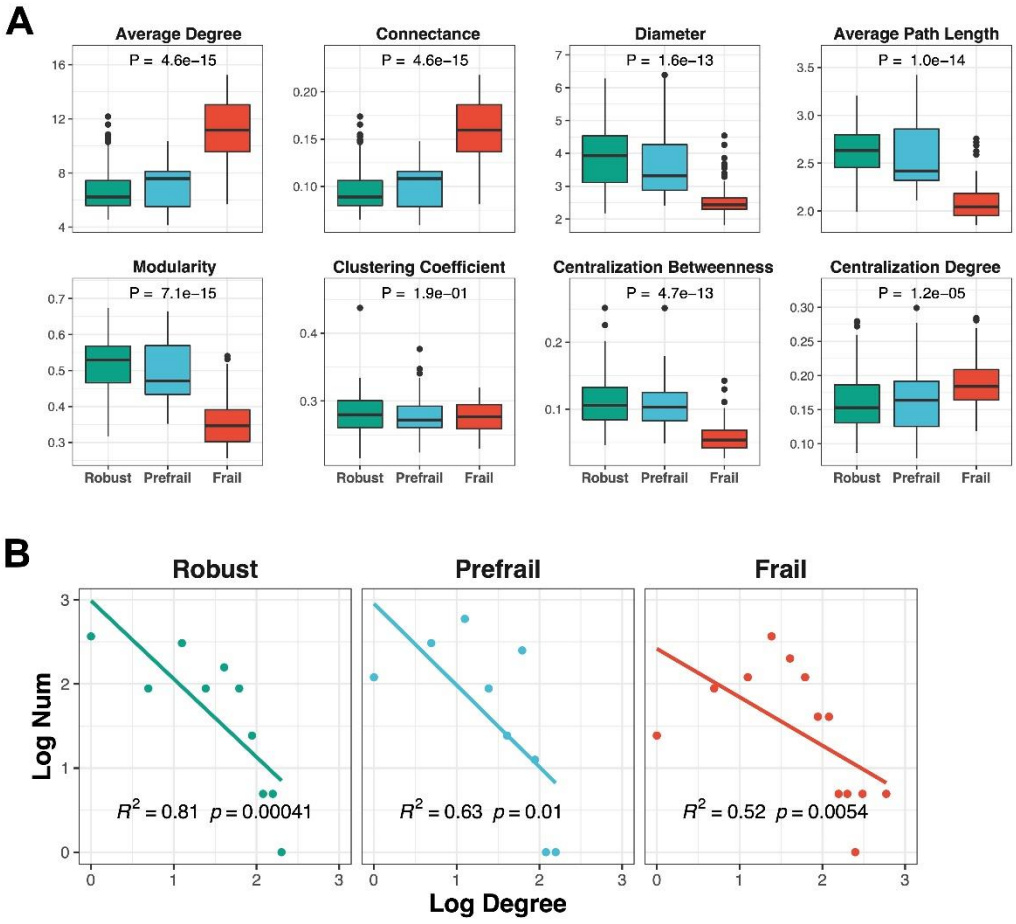

Figure 4