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# Towards understanding topological features of protein interactome map of muscle cancer

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**ABSTRACT**

Muscle cancer despite being rare but it is important for more examination due to its high mortality risk. Here, applying protein-protein interaction PPI network, we analyzed proteins that are related to this neoplasm in more details. The String Database (db) as the source for network was used through Cytoscape 3.4. The centrality examination by Network Analyzer, Cytoscape plug-in is done. The findings indicate that there are some proteins with higher linkage to the disease and with central topological features. These hub-bottlenecks are TP53, AKT1, MYC, and KIT. The prioritized proteins in this analysis indicate that these molecules may have prominent role in interactome of muscle cancer; however, more investigation is required to establish this claim.

**Keywords**: Muscle cancer, Rhabdomyosarcomas, Protein-protein interaction (PPI) network analysis, Centrality analysis.

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## Introduction

Tumors that develop in soft tissues such as muscles are benign, tumor-like lesions and malignant neoplasms. The neoplasms are very rare but deadly. They are known as rhabdomyosarcomas. These types as mentioned are not very common and they are involved with two categories. Embryonal and alveolar kinds occur in children and adults, respectively. There is another sub-group of this malignancy (Pleomorphic rhabdomyosarcoma), which is mostly incident in elderly (1).

Rhabdomyosarcomas contains about 40% of soft tissue sarcomas (2). In some cases, metastasis from other organs develops to skeletal muscles. For instance, it may happen from lung cancer (3). Many biomarkers are assigned for rhabdomyosarcomas (4, 5). The identification of the tumor major proteins can be helpful to better understand the muscle cancer mechanisms.

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Revealing disease underlying mechanism can be effective for developing therapeutic methods (6). In fact, for this purpose there are many high-through put approaches such as microarray, proteomics, and bioinformatic analysis. Bioinformatics showed promising to accelerate this goal in many aspects. One of the novel aspects of bioinformatics usage is protein-protein interaction network analysis. Proteins do not act individually. They are in a complex interactome system that their normal function is required for a biological network integrity. The dysregulation of each component of the PPI network can lead to vast amount of dysfunctional behavior of an organism. In a serious condition, proteins that are central for the strength of a network are possibly dysregulated (7). Therefore, it is important evaluate this level of molecular context for disease interpretation. In this study, PPI network of muscle cancer is analyzed to provide a preliminary understanding of the disease interactome profile.

## Materials and Methods

For network construction, STRING Database integrated in Cytoscape 3.4 was applied (8, 9). String provides information from different sources. There are three options for query by String db, including disease query, PubMed query, and protein query. Here, disease query part is selected for muscle cancer. At first, the name of the disease was searched through disease query section of String db. The code for the disease is DOID: 4045. After that the parameters for the network construction was set as follows: number of nodes: 100 and the combined interaction score= 0.4 as the default option. The obtained network consists of many nodes and edges with different interpretations. The proteins with high disease score are those that have significant relationships with the disease (7). In addition, there is a cutoff score for interaction strength between nodes. This score is obtained from different data sources that String db combines these scores. Therefore, interactions with high scores are the proteins with high affinity. Following network construction, the Cytoscape algorithm, Network Analyzer is a suitable application for network central parameters evaluation. The centrality in a scale free network is considered as nodes with values likewise vital role in network integrity. Graph display explains about the behavior and distribution of nodes for different parameters (10). The selected parameters in this study are degree and betweenness centralities. Nodes that have highest connected links are assumed as hubs likewise, nodes that have larger numbers of shortest paths passing through them are bottlenecks (11). Proteins with both characteristics are hub-bottlenecks. Hub-bottlenecks are the strongest components of a PPI network (12). Visualization of centrality and disease score values was handled by style section of Cytoscape. In this part, the network can be adjusted to values distribution. The sub-network creation led to identification of some components of the network. The analysis was shown in this study for the first sub-network component that is a network with connected nodes and the isolated nodes were identified as small components of the query that was not considered.

## Results

The network construction can provide topological information of proteins in a whole interactome pattern. The related network to muscle cancer was constructed by 100 nodes (including 81 connected nodes and 19 isolated proteins) and 609 edges (see figure1). Network

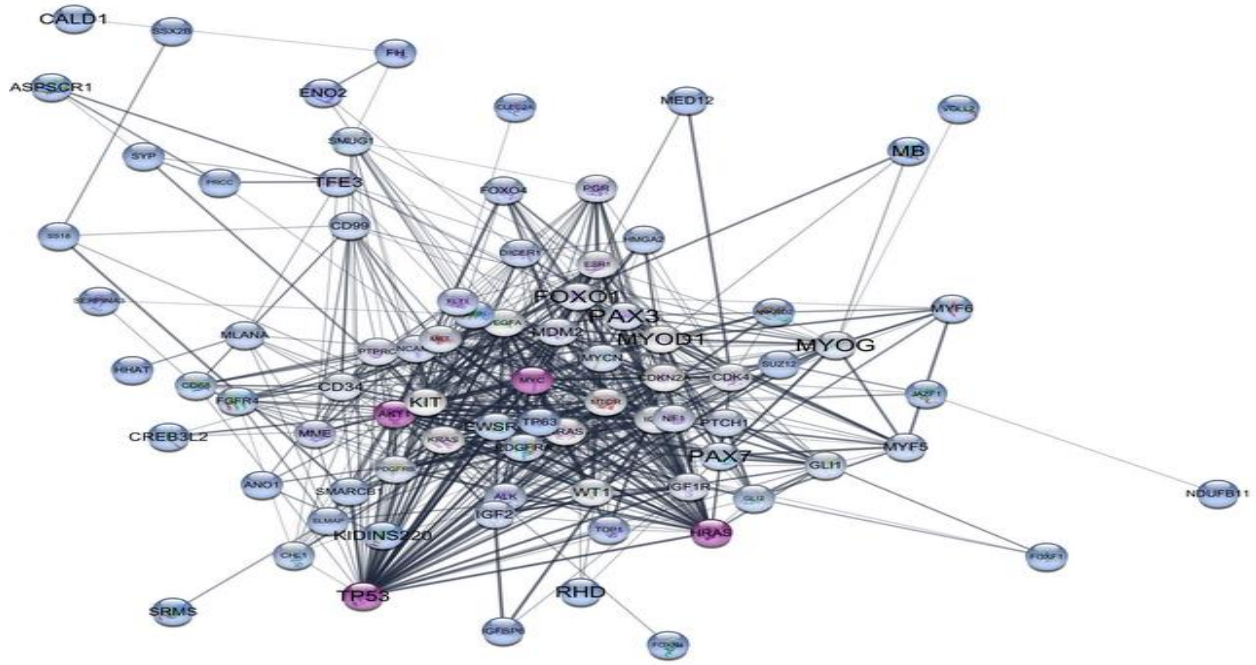



**Figure1.** The analyzed sub-network of muscle cancer with 81 nodes after removing 19 isolated nodes. The labels from small to big indicate the disease associations and the color changes from blue to purples shows degree increment.

**Table1.** Network general information obtained by Network Analyzer

| Network Nodes NO | Network Edges No | Network Isolated Nodes | Network Confidence Score | Highest Disease Score |
|---|---|---|---|---|
| 81 | 609 | 19 | 0.4 | 3.5 |

characteristic provides the overall information with the adjusted cutoff for its construction in table1. The distribution of central parameters; degree and betweenness centrality (BC) for the retrieved network is displayed as graphs in figure 2.

The nodes with highest centrality values are known as hub-bottleneck elements. The cutoff for the hub-bottleneck nodes selection was obtained by graph analysis in the figure 2, based on BC that is above 0.05. The network consists of proteins with different score values related to the disease (see table2).

## Discussion

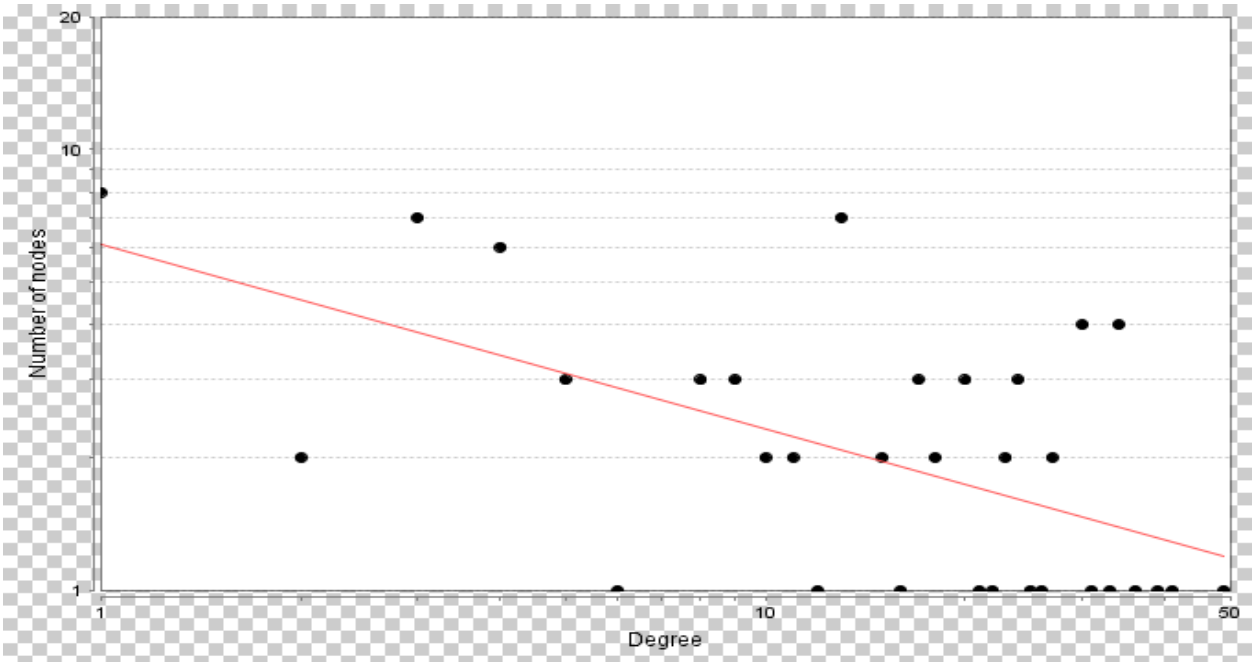
Molecular investigation of muscle cancer is important to provide more information about disease mechanisms. One of the novel ways in this regard is the application of PPI network (13). PPI network analysis can present more knowledge about the disease in the terms of interaction basis (14). In fact, different functions of a cell are by the active systematic interactions of protein components. Interaction examination identifies topological features of a disease that among them are some central agents that plays crucial role in the network strength. It means that without normal functions of these elements,

**Table2.** Centrality analyses for muscle cancer disease, the nodes are listed based on degree values. The asterisk signs indicate the hub-bottlenecks

| Row | Gene Name | Disease Score | Degree | BC |
|---|---|---|---|---|
| 1 | TP53* | 2.83535 | 49 | 0.087231 |
| 2 | AKT1* | 1.726686 | 41 | 0.098457 |
| 3 | MYC* | 1.540923 | 39 | 0.068452 |
| 4 | HRAS | 1.968372 | 36 | 0.015563 |
| 5 | KIT* | 2.651509 | 34 | 0.071001 |
| 6 | CDKN2A | 1.681523 | 34 | 0.033233 |
| 7 | KRAS | 1.440961 | 34 | 0.015934 |
| 8 | NRAS | 1.683613 | 34 | 0.016271 |
| 9 | MYOD1 | 3.196026 | 33 | 0.037328 |
| 10 | VEGFA | 1.32088 | 31 | 0.021464 |

many abnormal processes can be triggered and the whole phenotype of a cell changes (10). The vigorous dysregulation of proteins with high centrality may lead to disease onset and development (15). Here, the important components of muscle cancer are evaluated. In figure1, a PPI network constructed of 100 related proteins with muscle cancer is built. However, 19 proteins are excluded and isolated. As it is shown in table 1 there are 81 nodes and 609 links or 7.5 links per each node. Though, the network as a PPI map behaves differently and the distribution of linkage for the nodes are not similar known as a scale free network which can be interpreted in figure2. This network consists of many features that can be discussed in details in the following figures and tables. The disease score relations and degree values changes are visualized in the network. The overall information of the disease network is listed in the table1. This table shows that the network contains a big sub-network and small isolated components. Further analysis via sub-network creation approved the result that the network contains a big sub-network of proteins and therefore the rest of the analysis was for the sub-network. Distribution of degree and betweenness centrality is shown in figure 2. Only small numbers of nodes show large and high values of the mentioned centrality parameters. In table2, the centrality analysis is resulted in identification of some central components. The list of top ten of nodes corresponding to degree and BC is mentioned. However, only four of these showed the highest values among others due to established cutoffs for this selection. The four key proteins in our study are TP53, AKT1, MYC and KIT that possess high disease relations. These proteins are previously reported for muscle tumours (16-19). It can be concluded that, while there are many reported proteins for this malignancy (20-22), among them, there are four important ones in terms of interaction levels. TP53 gene encodes tumour protein p53. This protein is a tumour suppressor protein and inhibits cell proliferation (23). AKT1 gene encodes serine-threonine protein kinase. This enzyme is involved in metabolism regulation, cell proliferation and division and angiogenesis processes (24, 25). MYC encodes a multifunctional protein that is involved in the processes such as cell cycle (26). KIT as like the other three mentioned genes is involved in cancer processes (27).

A




B

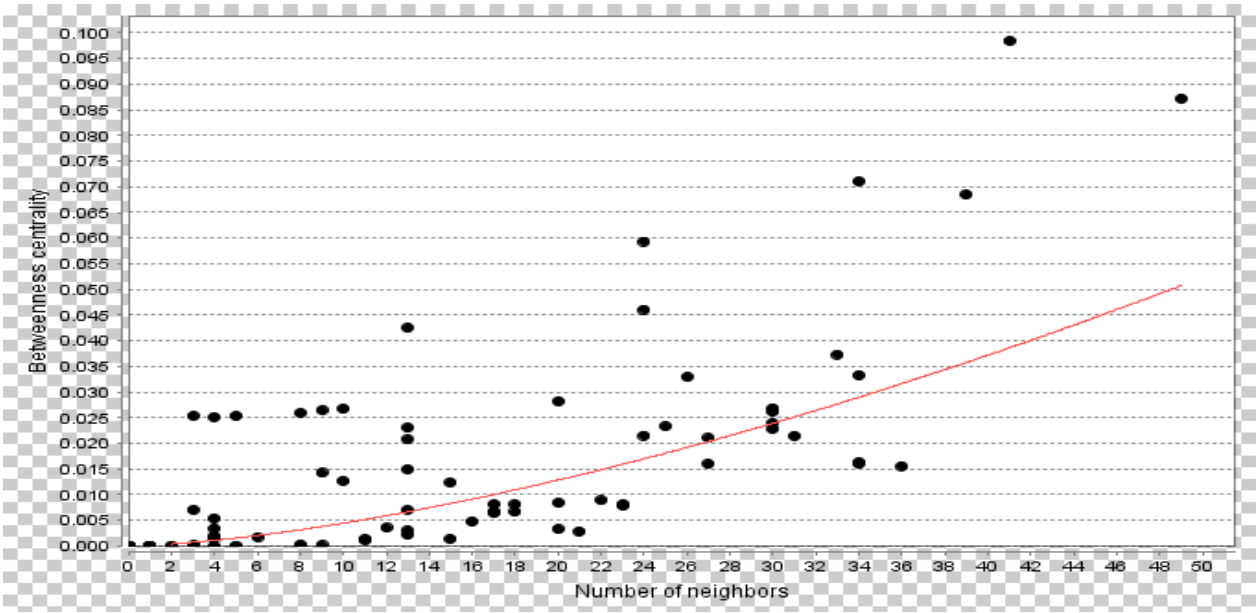



**Figure2.** Degree and BC graphs of the query network indicate a scale free network. Nodes are shown as black circles A: degree distribution that is between 0-49, Correlation= 0.6 and R-squared= 0.3 B: Betweenness centrality distribution that is between 0-0.1, Correlation= 0.6 and R-squared= 0.4.

## Conclusion

The finding revealed among 100 related proteins, there are four crucial ones including TP53, AKT1, MYC and KI that closely are involved with muscle cancer. It can be concluded that this biomarker panel can be improve for

discrimination of muscle cancer from the other diseases with similar signs.

## Conflict of Interest

The author Declare no conflict of interest.

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