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The over expression of thioredoxin during malignancies

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ABSTRACT

Thioredoxin system comprised of thiorexin and NADPH dependent thiorexin reductase, is responsible for redox regulation of cells by controlling the apoptosis, proliferation and other vital processes of cells. The efficacy of thioredoxin system has been represented in a wide range of physiological and biological reactions in bacteria, yeast, plants, mammals and etc. including DNA synthesis, regulation of transcription factors, protein repairing, regulating the photosynthesis and controlling the apoptosis and preventing oxidative stresses, filamentous phage assembly, immune-modulating, neuronal survival, pregnancy and birth and many other physiological and biological functions. The up-regulation of thioredoxin has been observed in various malignancies, which was associated with tumor angiogenesis and development. In this regard, the thiordoxin system has become a putative target in new chemotherapeutic methods. In this study, we mentioned various features of thioredoxin system in malignant cells and reviewed the articles which have evaluated the expression rate of thioredoxin system in malignancies.

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Introduction

Thiredoxin system

Thioredoxins (Trx) are among redox proteins, which are firstly isolated from Escherichia coli in 1964. These proteins are widely distributed in nature from prokaryotes to eukaryotes. The thioredoxin system comprised of thioredoxin (Trx), flavoprotein thioredoxin reductase (TrxR) and NADPH, keeps the thioredoxin in its reduced state

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and acts as a high capacity hydrogen transport system. This system contributes in cellular functions and regulatory mechanisms through stabilizing disulfide bonds and facilitating the thiol/disulfide exchange reactions as antioxidants. The function of thioredoxin system is crucial in regulating the redox hemostasis, which results in the maintenance of the cellular viability and function. The expression of Trx has been detected during the fetal development that is the indication of its role in fetal cellular function (1). Trx is a multifunctional redox regulator with intracellular and extracellular functions. It would be secreted due to various stimuli (X-ray radiation, ultraviolet (UV) light, tumor necrosis factor α and etc.) to protect and regulate the cellular responses (2). The secretion of Trx have been identified in different mammalian cells such as placental, liver, secretory, leucocytes and keratinocytes of the skin due to various stimuli (3). TrxR is a selenoprotein, which is a flavoenzyme that reduces the Trx dependence on NADPH.

The efficacy of Trx system has been presented in a wide range of physiological and biological reactions in bacteria, yeast, plants, mammals and etc. including DNA synthesis, regulation of transcription factors, protein repairing, regulating the photosynthesis and controlling the apoptosis and preventing oxidative stresses, filamentous phage assembly, immune-modulating, neuronal survival, pregnancy and birth and many other functions. Peroxiredoxins and methionine sulfoxide reductases are known as two direct targets for the disulfide bond reduction of Trx isophorms (4).

Selenium (Se), which is a biologically important factor, is crucial for maintaining the optimum cellular function and could be found in proteins as selenocysteine. Although Se has shown some preventive effect on tumor development and invasion, its high concentrations might have cytotoxic effects. In the study of Erkhembayar et al. selenium-dependent increase of TrxR activity was observed in rat by administrating Se supplementation. In this study, it was supposed that TrxR might act as a carrier, which supported the supplementary Selenite to exert its effect.

Cytosolic isophorms (Trx1 and TrxR1), mitochondrial isophorms (Trx2 and TrxR2) and another third isophorm (TGR), all have been expressed in mammals with cells specific expression patterns (5,6). There are two kind of cytosolic Trx (Trx1 and Trx80), which have differentiated due to their immune-modulator functions regarding co-cytokines or chemokines. All the Trx proteins contain two catalytic cystine residues in their conserved active site with the (-Cys-Gly-Pro-Cys-) amino acid sequence (7).

Human thioredoxin-1 (Trx-1) is a protein with 104 amino acids and 12 k-Da molecular weight, which have been cloned from various mammalian species. In human, the Trx gene localization has been mapped to chromosome 9 at band 9q32 (8). Thioredoxin-2 (Trx-2) contains 166 amino acids with the 18 k-Da molecular weight, which have been cloned previously (9). In normal cells, the preventive effects of TrxR on the occurrence of malignancies have been shown in different ways that lead to the maintenance of DNA structure form mutations and oxidative damages and regulating the redox homeostasis. TrxR has shown supporting effect on the function of p53 (a well-known tumor suppressor) and the selenocycteine residue of the TrxR has also shown cancer inhibitory effects. These are among various pathways that TrxR involves in maintaining the redox status of the cells.

Trx contributions in malignancies

According to literature, Trx is neither indicated as a direct oncogen nor contains

any mutagenic properties due to its expression in many normal cells and tissues. It has simulative and inhibitory effects on growth and apoptosis of normal cell, respectively and it would lead to development of the malignancy in cancerous cells.

Adult T-cell leukemia-derived factor (an interleukin-2 receptor inducer) is known as human thioredoxin homologue and can increase the proliferation of the malignant cells (10).

In various unusual situations such as cardiopulmonary bypass operations (11), rheumatoid arthritis, HIV and etc. the increased levels of plasma Trx system has been observed which make it as a putative target in tumor therapy (12,13).

It is proved that the presence of Trx is essential in tumorogenesis but the level of Trx involvement is different based on the type of the cancer. Based on different studies and using several techniques, mostly immunohistochemistry, the Trx expression has been evaluated in various types of human cancer cells such as lung, colorectal, cervix, gastric, hepatocellular and well-differentiated squamous cell carcinomas of skin in comparison with their related normal tissues. This has resulted in several fold increase of the Trx concentration in plasma and serum of cancer cells (14-18).

Based on these studies, it has been speculated that the Trx overexpression might be induced by the increased oxidative stress and hypoxic conditions resulted from the carcinoma cells. It is also concluded that tumor cell with overexpression of Trx showed higher proliferation and higher durability than tumor cells with no expression of Trx.

Miyazaki et al. revealed that-40 Cytokines such as interleukin-6 (IL-6) and interferon have revealed inducible role in increasing the Trx expression in condition such as HIV. During in-vivo situations, the lysis of red blood cells is considered an important source of elevating the Trx despite it is not known as the major cause of the increased level of plasma Trx. This is due to the high content of Trx in erythrocytes (21). Monocytes/macrophages and endothelial cells are the other sources of plasma Trx (22). Platelet dysfunction in Hermansky-Pudlak syndrome has resulted in elevated Trx concentrations (23).

Increasing the growth, angiogenesis and anti-apoptotic activities in tumor cells

In cancer cells, the overexpression of Trx might result in more aggressive phenotypes by accelerating the cell proliferation and decreasing the apoptosis or even modulating the cell genetic arrangements.

The Trx system might pose its growth promoting effects by altering the transcription factor expression and the consequent protein kinases activity cascades, modulating the integrin and adhesion molecules activity or even through the direct influence on the DNA synthesis (19). Based on immune-histochemistry studies, Trx would act as an autocrine growth promoting adult T-cell Leukemia-derived factor (ADF) in extracellular environment, or a stimulant which induces the growth of B cells which was indicated in hepatocellular carcinoma (10). In one study considering the Trx effects on breast cancer cells proliferation, it was suggested that Trx acted as a growth factor on cancerous cells and stimulated cells to produce their own growth factors (self-stimulating growth factor production) for more cell proliferation and tumor development (24). TrxR plays a major role in counteracting the processes of apoptosis in cancer cells including modulating through inhibition of the Ask-1 signaling cascade or supporting the antioxidant pathways such as peroxiredoxin activity (25).

TrxR links to the process of angiogenesis which is a vital phenomenon for successful tumor development. Based on the studies, this association might be through the increasing of hypoxia-inducible factor-1 (HIF-1) and

VEGF (Vascular endothelial growth factor) following the up-regulation of TrxR (26).

TrxR a putative chemotherapy target

Resistance to chemotherapy is another feature of cancer cells with high expression level of TrxR. Due to the prominent role of the TrxR in various levels of cancer development, TrxR has become an attractive target in chemotherapy methods and there were variety of studies, which aimed to discover new medicines for diminishing the up-regulated TrxR in every malignancy.

Methods in malignancy suppressing, which are associated to the TrxR, might affect the selenium part of the enzyme, directly knock down the TrxR or apply the medicines, which inhibit the TrxR activities.

There are various synthetic and natural origin products, which have shown TrxR inhibitory effects. Some inhibitors used for cancer diminishing through inactivation of TrxR, are listed in Table 1.

The metal containing inhibitors have exerted their effects in a DNA-independent way and through inhibiting the TrxR and inserting cytotoxicity influence against cancer cells, which results in anti-proliferative effects (27).

Red drinking wine (contains containe myricetin and quercetin as flavonoids) is considered to induce the apoptosis due to its high content of antioxidants (28). Theaflavins are the polyphenols in black tea that cause its antioxidant and antitumor effects (29). Anthranoids obtained from plants and bacteria, Curcumin from Curcuma longa and Mansonone F obtained from Ulmus pumila, have shown antitumor properties (30,31).

In the study of Baker et al. administrating PX-12 (1-methylpropyl 2-imidazolyl disul-

Table 1. Different drugs used for suppressing the TrxR activity in cancer cells

Drugs			Evaluated cell line
Metal-containing inhibitors (metallodrugs)	Gold-containing inhibitors	Auranofin	Applied in human ovarian cancer cells
		gold-phosphine complexes	A549, MCF-7, HeLa, HL60
		gold-triphenylphosphane complexes	MCF-7, A549, A431
		gold-triazaphosphaadamantane complexes	MCF-7, A549, A431
		gold-NHC (N-heterocyclic carbene) complexes	MCF-7, HT-29, HCT-116, HEP-G2
		gold-thiosemicarbazones complexes	HL-60, MCF-7, HCT-116, Jurkat
	Ruthenium-containing inhibitors	PMRU20	HT29, MCF-7 and A549
		RUMA	
		NAMIA	
		PMRU27	
Naturally occurring products	Flavonoids and Polyphenols	Red drinking wine Tea (black tea)	HEK-293, H157, A549,HCT116
	Anthranoids	Hypericin (HYP) pseudohypericin (PHYP)	
	Curcumin		
	Mansonone F		
	Cinnamaldehyde		
	acylfulvene		
Newly emerged inhibitors	Indolequinones	ES936 and ES939	human pancreatic cancer cell lines
	Porphyrins	Rottlerin Protoporphyrin IX	
	Statins		human liver biopsies

fide) was proposed as a novel method in decreasing the plasma concentration of the TrxR through inhibiting the expression of TrxR and finally the VEGF of the tumor cells. In this study, PX-12 was administrated in patients with different malignancies such as colorectal and pancreatic cancers. The SELDI-TOF (Surface-enhanced laser desorption ionization time-of-flight) mass spectrometry method was applied for evaluating the plasma changes regarding the TrxR concentration. It was concluded that PX-12 reduce the plasma concentration of TrxR and VEGF, specifically in patients who had shown high plasma levels of these two proteins (32). In a randomized trial, PX-12 was administrated in patients with advanced pancreatic cancer, which did not result in noticeable antitumor activity (33).

It has been suggested that the apoptosis would be increased in tumor cells by targeting TrxR due to elevated oxidant susceptibility. It was also recommended that signaling pathways of ASK-1 and NF-αB would lose their regulation by diminishing the TrxR and finally it resulted in increased apoptosis due to the activation of JNK and p38 signaling cascades (34).

Manipulating through molecular approaches is another way to reduce the upregulation of TrxR and reverse the tumorogenic phenotype of the cancer cells to the normal cells.

In the study of Yoo et al. preventing the malignancy development was achieved by dirsct action on TrxR. In this study, a reduction in the TrxR level was observed as a result of knocked-down TrxR in lung cancerous cells via siRNA, which were based on the PhosphorImager method outcomes, by using 75Se for the labeling of the LLC1 (Lewis Lung Carcinoma) cell line of the mouse lung carcinoma. Further analysis in this study showed various phonotypical changes of cancer cells similar to the normal cells including a considerable reduced rate of cell proliferation and the monolayer growing of cells, which were tightly attached to the culture dish (35).

Conclusion

Although there are various studies regarding the effects of thioredoxin system in the process of cancer development, further investigations could increase our knowledge about the association between thioredoxin system upregulation and the cancerous cell functions. More comprehensive information improves the therapeutic techniques in cancer prevention.

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Conflict of Interest

The authors declare no conflict of interest.

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