DERLEME / *REVIEW*

The Hypothesis of a New Extrachromosomal Approach to Cancer Treatment

Kanser Tedavisine Yeni Bir Ekstrakromozomal Yaklaşım Hipotezi

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ABSTRACT

There are important unexplained points in cancer formation and its treatment with drugs. Some of the cancer drugs used in the treatment affect the genetic material of cancer cells. These types of drugs have a high toxicity. Despite important drugs used in cancer treatment have been developed today, there are serious problems in developing a drug with high specificity. This is because the pharmacological action points where drugs can affect cancerous cells and which are different from normal cells have not been fully determined. The current article reveals hypotheses about some extrachromosomal action points that may be beneficial in cancer treatment. Depending on the proposed hypothesis, information about the chemicals to be investigated is given.

Key Words: *antineoplastic drugs; Warburg hypothesis; aromatic amino acid; aromatic alpha-keto acid; Coenzyme Q10*

ÖZET

Kanserin oluşumu ve ilaçlarla tedavisinde aydınlatılmamış önemli noktalar bulunmaktadır. Tedavide kullanılan kanser ilaçlarının bir kısmı kanser hücrelerinin genetik materyaline etki etmektedir. Bu tip ilaçlar yüksek toksisiteye sahiptirler. Günümüzde kanser tedavisinde kullanılan önemli ilaçlar geliştirilmiş olmasına rağmen, henüz spesifitesi yüksek bir ilaç geliştirilmesi konusunda ciddi sorunlar vardır. Bunun nedeni kenserli hücrelere ilaçların etki edebileceği normal hücrelerden farklı farmakolojik etki noktalarının tam olarak tespit edilememiş olmasıdır. Bu makalede kanserin tedavisinde faydalı olabilecek bazı ekstrakromozomal etki noktaları ile ilgili hipotezler ileri sürülmüştür. Önerilen hipoteze dayanılarak araştırılması gereken kimyasal maddeler hakkında bilgiler verilmiştir.

Anahtar kelimeler: *antineoplastik ilaçlar; Warburg hipotezi; aromatik aminoasit; aromatik alfa-keto asit; Coenzyme Q10*

Introduction

Chemical carcinogens, known as an important cause of cancer (70%), develop tumors, shorten the emergence time of the tumor, and promote metastasis. Primary carcinogens directly initiate cancer, and secondary carcinogens initiate by reacting with the genetic structure via their metabolites. Co-carcinogens facilitate the promotion and progression of the tumor. Tumor cells, which form by the effect of chemical carcinogens, go through the stages of initiation, promotion, and progression[1,2].

Drugs used in cancer treatment may affect the tumor cell (internally: genetic and other functions) and the surrounding of the tumor (externally)[3].

Genetic Targets

Among the genetic targets of the drugs, mutant oncogenic genes and epigenetic and transcriptional conduction irregularities take place. RAS proteins coded[3] by the RAS gene (membrane-associated G-proteins: HRAS, KRAS, NRAS) are protooncogenic, and they have GTPaz activity. Its GPT-related form transfers the signals, stimulating cell division, from the cell membrane to the nucleus[4]. BRAF gene (Ras Associated Factor) codes a protein that controls cell division. Point mutations of the BRAF gene cause the activation of tyrosine kinases related to the membrane, such as MEK (MAPKK), ERK (MAPK), SRC, and BCR/ABL[4]. Mitogen-activated protein kinase (MAPK) has serine/treonine kinase (in the cytoplasm) activity that plays a role in the intracellular signal pathway (MAPKsi, AKT). Drugs such as vemurafenib, dabrafenib, trametinib, and cobimetinib inhibit BRAF mutation[3]. PI3K (phosphatidylinositol 3-kinase) is activated by the mutations of PIK3CA (phosphatidylinositol 3-kinase, catalytic, α-polypeptide). PI3K inhibitors are tried on PIK3CA-mutation cancers[3].

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The tumor is located between the suppressor genes Gatekeeper (APC, p53, Rb) and Caretaker (TP53). Point mutation, loss of heterozygosity (LOH), and increased methylation are seen in these genes in cancer[4]. Mutations causing loss of function in ubiquitin ligase CBL increase the signal on the thyrosine kinase pathway. The mutations in PTEN inactivating the phosphatase activity increase signal on the PI3K pathway. Drugs restoring the normal function of mutant TP53 have been studied. A bisindolylmaleimide-derivative drug has been discovered that regains the interaction between SMAD4 and SMAD3 mutations, which are cell growth-suppressing genes. Poly (ADP-ribose) polymerase (PARP) enzyme activity increases when BRCA1 and BRCA2, DNA repair and tumor suppressor genes, are mutated. Inhibition of these enzymes increases success in the treatment. Inhibition of CYCLOPS genes causes a loss in the number of copies. This induces death in the cells that do not contain the corresponding tumor suppressor gene (paralog)[3].

Extragenic Targets

Tumor microenvironment, differentiation in the immune cells and stromal cells such as fibroblasts, abnormal endocrine signals, and microbiome occur in the environmental targets of antineoplastic drugs. Antiangiogenic treatments have been developed that target the vascular endothelial growth factor (VEGF), one of the external proteins of normal cells. It is thought that antibodies inhibiting immune checkpoints, such as PD-1/PD-L1 and CTLA-4, may effectively treat some types of cancer. Chimeric antigen receptor T-cell (CAR-T) infusion has been used in hematologic malignancy. Antibodies of multiple cell surface receptors (ex., Klgr1 and TIGIT) blocking metastatic colonization have been detected. Some antibodies are effective against proteins inhibiting the immune responses, such as programmed cell death protein 1 (PD-1)[3]. Mitosis inhibitors, hormones and antagonists, enzymes (asparaginase), tyrosine kinase blockers (imatinib), growth receptor inhibitors (trastuzumab), and vascular endothelial growth factor inhibitors (bevacizumab) are used in the treatment[1,2]. In addition, studies on the correlation between energy and synthesis have been conducted.

Warburg Effect

Otto Heinrich Warburg determined that the tumor cells convert glucose to lactic acid over pyruvic acid under aerobic conditions. Due to this finding, also known as Warburg effect, the researcher won the Nobel prize in 1924[5–8]. In healthy cells, pyruvic acid, composed of glucose, is transported to the mitochondria, converted to Acetyl CoA, and decomposed in the TCA cycle. Meanwhile, 36 ATP are produced from a glucose molecule (38 ATP in total). Tumor cells convert pyruvic acid into lactic acid via glycolysis in the cytoplasm under aerobic conditions and produce 2 ATP from one glucose molecule (4 ATP in total)[5,6,8–12].

Pyruvic acid is degraded with 95% oxidative phosphorylation and 5% aerobic glycolysis in healthy cells[5]. Cancer cells are degraded with approximately 85% aerobic glycolysis and 15% oxidative phosphorylation[11]. Oxidative phosphorylation in healthy cells is 19 times more than aerobic glycolysis (95/5). According to the rates mentioned above, healthy cells obtain 36.3 ATP from one mole of glucose, and cancer cells obtain a total of 9.1 ATP.

Reason for Cancer and Aerobic Glycolysis According to Warburg

According to Warburg, cancer is caused by anoxemia and function disorder in mitochondria[10,12]. Cells that adapt to low-oxygen environments are converted into cancer cells by fermentation. Non-adapted ones are subject to apoptosis[5]. As the mitochondria of cancer cells cannot function, apoptosis is not observed, and they continuously generate[8]. Fermentation developing in cancer is irreversible[5,6]. Pyruvic acid and alpha-ketoglutarate are converted into Acetyl CoA by dehydrogenases using thiamine pyrophosphate as co-enzymes[2]. This data supports Warburg effect.

Cancer cells are thought to retain their ability to ferment in the evolutionary process by inheriting from bacteria[6,9]. Besides cancer cells, fermentation is also seen in all the healthy cells (immune system cells in case of infection, healing wounds, embryonic cells) that generate rapidly[10]. Fermentation also occurs in the muscle cells as a result of excessive physical activity[6,10,13]. However, Warburg effect seen in normal cells is reversible[10,12].

Studies have indicated that most cancer cells do not impair mitochondrial function. Tumors can also be seen in lungs rich in an oxygenated environment. Thus, the idea that cancer arises from impaired aerobic respiration in mitochondria is not entirely accepted[11].

Metabolites, occurring due to Warburg effect, are required for angiogenesis required by the cancer cells[6]. Metabolites help the synthesis of agents such as

protein, lipid, and nucleic acid[10,11]. NADPH synthesis reproduces upon the Warburg effect. Glutamine, synthesized by transamination, contributes to NADPH production by catabolizing malate dehydrogenase into lactate[11]. NADPH acts as a reductant in the synthesis of fatty acids[10]. Modulation of pyruvate kinase facilitates re-directing pyruvic acid to pentose phosphate, nucleotide, and amino acid biosynthesis pathways[9–11]. The cells convert lactate, which is generated in the tissues by Cori cycle, back to glucose in the liver. In addition, they convert alanine back to glucose. One glucose molecule may provide 30 ATP and 2 NADPH if it enters the pentose phosphate pathway or 6 carbons for the synthesis of macromolecules if it does not enter the pathway[11]. If a glucose molecule degrades in TCA, it provides 5 times the ATP needed to generate a 16-carbon fatty acid chain. However, 7 glucose molecules are required to generate NADPH in the same synthesis[9,11]. Thus, aerobic glucose develops to provide elements to the cell rather than energy.

Lactic acid, emerging due to the Warburg effect, reduces pH of the surrounding tissues. This facilitates the invasion of cancer[5,6,10]. Lactic acid increases the vascularization of the tissues and encourages metastasis. It is also efficient by developing the tissue fibers acting in metastasis, such as collagen[10].

The Use of Warburg Effect in the Diagnosis of Tumor

In Positron Emission Tomography (PET), used for tumor diagnosis, fluorodeoxyglucose (FDG), which is a radioactive fluorine (^{18}F) compound, is used. It was detected by PET that tumor cells use an excess amount of glucose[8–10]. Cancer cells use glucose approximately 20 times more than healthy cells[6,8].

Causes of Cancer and Use of Metabolic Differences in Cancer Treatment Today

The cause of cancer is explained by the activated oncogenic genes and inactivated tumor suppressor genes related to the mutations[10].

It can be considered that reduction of glucose uptake may be beneficial in the treatment of cancer (apoptosis). This study determined that cancer is less seen in approximately 30 thousand patients with Type 1 diabetes mellitus, followed up for 30 years, than in healthy people[6].

In normal cells, p53 gene inhibits excess glucose uptake, lipogenesis, NADPH synthesis, and cell proliferation. In cancer, this gene is mutated, thus it cannot function accordingly[10]. Expression of TIGAR gene in cancer inhibits phosphofructokinase. It directs glucose to the pentose phosphate pathway and generates NADPH in glutathione synthesis. In healthy cells, high oxidative phosphorylation and ROS generation induce apoptosis[11].

Cell growth and glucose metabolism are controlled by insulin-like growth factor (IGF)– Phosphoinositide 3-kinase/Protein kinase B (PI3K/AKT) pathway[9]. PI3K signal pathway promotes the amino acids enter in protein synthesis via mTOR (mammalian target of rapamycin). PI3K signal increases the expression of glucose-transporting molecules via AKT. It stimulates hexokinase and phosphofructokinase activity and increases the use of glucose. Thus, small molecules degrading PI3K signal cause the tumor to regress[11].

Cells using aerobic glycolysis have a high ATP/ADP and NADH/NAD+ ratio. If ATP decreases, the cells undergo apoptosis. Adenylate kinase contributes to ATP production by converting two ADPs into one ATP and one AMP. The resulting AMP activates the protein kinase (AMPK) that is activated with AMP. Protein kinase activates various structures in the cell via phosphorylation (tumor suppressor protein LKB1). Metformin and phenformin activate AMPK in the cell. It is reported that adding metformin in cancer treatments may be effective in the prevention and treatment of cancer[11].

Oncogenes stimulate thyrosine kinase (in the cytoplasm) enzyme acting in cell proliferation. Activated tyrosine kinase regulates glucose metabolism (not seen in normal cells)[9–12].

Proliferating cells express the M2 (PK-M2) isoform of pyruvate kinase, a glycolytic enzyme[11]. PK-M2 enzyme is only present in cancer and rapidly dividing cells. It is not present in other cells. This enzyme accelerates the conversion of glucose to pyruvic acid in cancer cells. When the synthesis of PK-M2 enzyme is inhibited, enzyme passes into another form. This may prevent growth in cancer cells[8].

Pyruvate dehydrogenase enzyme complex in the mitochondria consists of three enzymes; pyruvate dehydrogenase, dihydrolipoyl methylase, and dihydrolipoyl dehydrogenase[6]. Pyruvate dehydrogenase enzyme is converted into inactivated form via phosphorylation by pyruvate dehydrogenase. Phosphatase enzyme converts the inactivated pyruvate dehydrogenase into active form. Active pyruvate dehydrogenase converts pyruvate into Acetyl CoA and provides it to be degraded with oxidative phosphorylation in TCA. Pyruvate dehydrogenase

kinase inhibitors prevent pyruvate dehydrogenase from being converted into inactivated form. Among them, drugs such as SB-204990, 2-deoxy-D-glucose, 3-bromopyruvate, bromopyruvic acid, 3-bromo-2-oxopropyonate-1-propyl ester, 5-thioglucose, and dichloroacetic acid are developed. This lowers the aerobic glucose level in the cytoplasm of cancer cells[8].

It is asserted that cancer can also be seen depending on the insufficiency of enzyme (citrate synthase) that converts pyruvic acid into citric acid[6]. Isocitrate dehydrogenase-1 (IDH1) and Isocitrate dehydrogenase-2 (IDH2) enzymes convert the isocitrate in cytoplasm to alpha-ketoglutarate via NADP+/NADPH. Mutation was determined in the gene coding cytosolic IDH1 in 12% of glioblastoma multiforme[3,11]. D-2-hydroxyglutarate, emerging depending on mutation, inhibits TET2 protein function, which is efficient on epigenetic mechanisms. Suppression of TET2 activation causes abnormal DNA methylation[3,10].

Mutations in enzymes involved in the TCA cycle, such as succinate dehydrogenase (converts succinate to fumarate) and fumarate hydratase (fumarate malate), are identified in human renal cancer. As a result of these mutations, Hif1α-mediated glucose use is activated[11].

In cancer cells, majority of the pyruvic acid is converted to lactic acid by the enzyme lactate dehydrogenase (LDHA) in the cytoplasm. In most human cancers, the LDHA amount is found to be higher than in healthy tissues. Inhibition of enzymes can be used in cancer treatment[6].

Cancer cells need glutamine, glycine, serine, and aspartate to synthesize the building blocks[3]. Cancer cells ferment glutamine, the most abundant amino acid in the blood, into lactate (ATP). It has been determined by the PET scans that some cancer cells use radioactive glutamine more than radioactive glucose (10 times more other amino acids)[9]. Human tumor cells, whose growth is directed by the MYC oncogene (mutated), are susceptible to glutamine deficiency[9,11]. The growth of tumors containing mutations in PIK3CA mostly depends on aspartate. Some tumors elevate the serine levels due to NRF2 and ATF4 signaling or oncogenic signaling as a result of the hypoxia responses. In this type of tumor, suppression of serine metabolism regresses the tumor. The tumorigenic potential of the cells initiating metastasis depends on their fatty acid uptake via CD36, which is the fatty acid receptor. Thus, lipid synthesis can be targeted in tumors containing high amounts of polyunsaturated lipids[3].

Hypothetical Points Recommended to be Investigated in Cancer Treatment

Phenylalanine is an essential aromatic amino acid[14,15]. It enters into the synthesis of protein and sympathetic amines in the body[1]. Tyrosine is chemically p-hydroxyphenylalanine[14]. Plants and bacteria can synthesize tyrosine (phenylalanine) from prephenate, released as an intermediate product in the shikimate pathway. Melanine, sympathetic amine, and thyroid hormones are synthesized from tyrosine in the body. Tryptophan is an essential aromatic amino acid, and it is glycogenic and ketogenic[14,15]. Plants and bacteria may synthesize tryptophane from shikimic acid or anthranilate[15]. Serotonin, niacin[14], kynurenine, and auxin are synthesized from tryptophane.

Phenylalanine, tyrosine, and tryptophan are absorbed from the digestive tract, used in protein synthesis, and metabolized. Phenylalanine is slowly and irreversibly converted to tyrosine by phenylalanine hydroxylase in the liver (75%)[1,14,16]. Phenylalanine is converted to phenylpyruvate via transamination. Phenylpyruvate is converted to either o-hydroxy phenylacetate or phenylactate by dehydrogenase enzyme[16]. Phenyllactic acid is oxidized in the body and is converted to phenylacetic acid. This product is conjugated with amino acids (glutamine, glycine) and excreted from the body[14,16,17]. Phenylalanine is converted to phenylethylamine by decarboxylation. This substance is converted to phenylacetylglutamate over phenylacetate and excreted in urine[16]. Benzene ring of phenylalanine is opened too little and then decomposed[18].

Tyrosine is decarboxylated (tyramine) and transaminated. Tyrosine transaminase enzyme that transfers the amine group in tyrosine to alpha-ketoglutarate converts tyrosine to p-hydroxyphenylpyruvate. This metabolite is converted to p-hydroxyphenyllactate and then to p-hydroxyphenylacetate. P-hydroxyacetate is excreted by conjugating with glycine and glutamine. In the digestive tract, tyrosine is converted to p-hydroxyphenylpyruvate by the deamination of its side chain under anaerobic conditions via the effect of intestinal bacteria. Then, p-hydroxyphenylacetic acid, p-cresol, and phenol are generated. Cresol (methylphenol) and phenols are absorbed by the intestines and then excreted by urine after being conjugated in the liver. Decomposition of the benzene structure of tyrosine is slow and rarely seen (fumarate, malate, acetoacetate)[14].

Tryptophan is converted to serotonin via hydroxylation and decarboxylation[14,18]. It is degraded by decomposing

to kynurenine, then to the products such as formic acid, alanine, xanthurenic acid, nicotinic acid, α-ketoadipic acid, Glutaryl-CoA, and Acetyl-CoA. Tryptophan is converted to indolepyruvate by oxidative deamination via tryptophanase enzyme secreted by *E. coli* in the intestine. Indolepyruvate is converted to indoleacetate and methylindol (scatol) by decarboxylation. Indole is formed from scatol in the liver. Indole is oxidized and converted to indoxyl and then to indoxyl sulphate and excreted by urine[14].

Phenylalanine, tyrosine, and tryptophan are converted to keto-acids (phenylpyruvate, hydroxyphenylpyruvate, indole pyruvate, respectively) by deamination or transamination in the body and some of them are degraded in TCA[14]. However, since TCA cannot serve with full efficiency in the tumor cells, the degradation rates are low by this pathway.

Ammonia is combined mostly with glutamate (glutamine synthetase) and converted to glutamine in the blood. In the muscles, it is transferred to pyruvate (alanine aminotransferase) and alanine is synthesized. Glutamine and alanine come to the liver. Alanine transfers ammonia to alpha-ketoglutarate and converts it to glutamate (transamination) and is converted to glucose via pyruvate[14,19,20]. Glucose enters the pentose phosphate pathway (NADPH production) or is degraded again to pyruvate via glucose-6-phosphate. Thus, it is used in the synthesis of serine (glycine and cysteine from serine), alanine, valine leucine and isoleucine, glutamate (then glutamine, pyrroline, arginine) and aspartate (then asparagine, methionine, threonine, lysine)[19,20]. In the tissues other than the liver, glutamine is converted to glutamate by deamination. Glutamate is included in the cell. It is converted to alpha-ketoglutarate and aspartate by giving the amine group to oxaloacetate[20]. Glutamine, glycine, and aspartate enter the synthesis of purine, glutamine, and aspartate pyrimidine (folic acid is also important)[14,20].

Degradation of DNA of the cells initiates cancer. However, degradation of its metabolism is required for progress. Lactic acid formed from glucose in the cancer cells, is converted to alanine (transamination) in the liver. Alanine synthesizes with Acetyl CoA, and thus the fatty acids via lactic acid and pyruvic acid[20]. The conversion of alanine to glucose and aliphatic amino acids provides the need for genetic base, amino acid, fatty acid, and NADPH of the tumors. Aliphatic amino acids such as glycine are conjugated with metabolites such as phenylacetic acid (can cause loss of aromatic amino acids). Disruption of this cycle can suppress the growth of cancer cells (Fig. 1).

Aromatic amino acids are converted to aromatic a-keto acids. Aromatic keto acid reductase (80%) and lactate dehydrogenase (20%) enzymes take place in the reduction of phenylpyruvic acid and other alpha-keto acids to phenyllactic acid (and its derivatives)[21]. Part of phenylpyruvic acid is converted to phenylacetic acid and excreted from the organism by conjugating with aliphatic amino acids such as glutamine and glycine in the body. Aromatic alpha-keto acids can prevent the conversion of pyruvic acid to lactic acid in the tumor cells as they use lactate dehydrogenase enzyme, which converts pyruvic acid to lactic acid. Lactic acid and alanine cannot be synthesized from pyruvic acid. Adequate glucose cannot be formed in the liver due to decreased alanine. Alpha-keto acids lower aliphatic amino acids such as glutamine and glycine, which enter into DNA synthesis, as they cause their excretion by conjugation. As a result, they less convert to pyruvate, glucose, and aliphatic amino acids (glutamine, glycine, aspartate). This may cause tumorostatic effect by suppressing DNA synthesis and cell proliferation in cancer cells. Besides the degradation of DNA, the reason of cancer may be associated with the lack of aromatic amino acids or their alpha-ketoacid metabolites (which may also be transaminase or oxidative deaminase enzyme deficiency).

Low level of tyrosine may sufficiently prevent the synthesis of Coenzyme Q10. This may explain the low oxidative phosphorylation rate in cancer cells. In addition, tyrosine also participates in the structure of thyroid hormones that produce energy in mitochondria (vast ATP production). If mitochondria do not show activity, cells enter into apoptosis (cannot make aerobic respiration). Thus, aerobic glycolysis is required not to have apoptosis in cancer cells (less ATP and less free radicals).

The absorption and distribution of aromatic amino acids taken in certain rates together with food is regulated (abundance of one reduces the passage of the other through the membranes.) by other neutral amino acids (alanine, asparagine, glutamine, glycine, isoleucine, leucine, valine, serine, pyrroline, threonine, methionine, and cysteine). Thus, amino acids in food are taken to the body at a fixed rate. Metabolites of aromatic amino acids taken in the body via food at a fixed rate do not prevent the use of aliphatic amino acids, of which synthesis has increased in the cancer cells, DNA, and other building blocks. Thus, the hypothesis can be proposed stating that aromatic amino acids or their

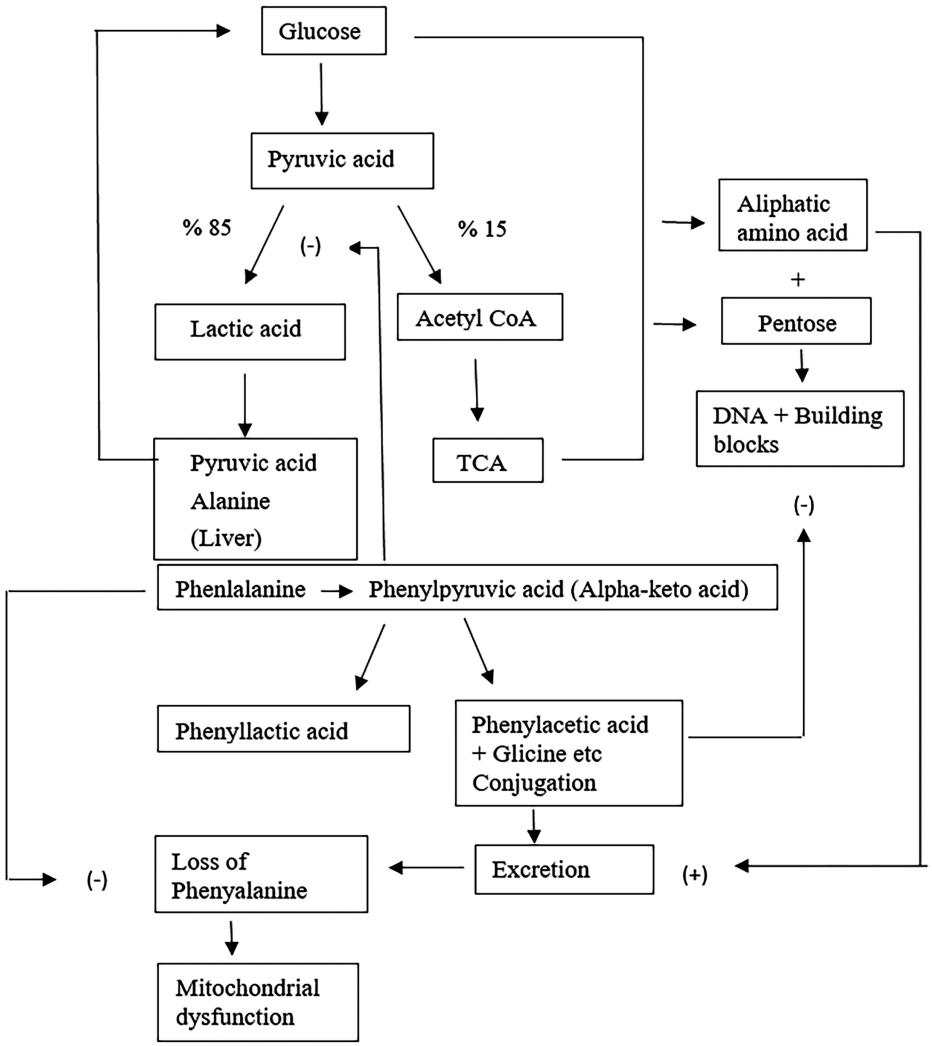

Figure 1. *Hypothetical action points of aromatic amino acids and aromatic alpha-keto acids (aromatic amino acid derivatives) in cancer cells.*

aromatic alpha-ketoside derivatives (including their analogs) should be administered externally for prophylactic or therapeutic purposes.

When aromatic amino acids are administered in high doses, they can also be efficient by decreasing the absorption of other neutral amino acids via gastrointestinal tract.

Sodium phenylpyruvate showed cytotoxic effect in lung (A549) and breast adenocarcinoma (MDA-MB-231) cell cultures (as far as is known, the hypothesis is passed for the first time)[22]. 4-Phenylbutyrate is a modulator that can cause differentiation or apoptosis in the cancer types such as prostate, ovarian, melanoma, glioma, and leukemia. 4-Pheynlbutyrate causes the excretion of glutamine from the body. Phenylbutyrate and phenylacetate are in the trial stage in treatment of Hodgkin's and non-Hodgkin's acute myeloid leukemia[23]. Phenylpyruvic acid is chemically similar to phenylbutyrate and is converted to phenylacetate. These studies support this hypothesis.

Fish are ammonotelic animals and they give the ammonia forming in their bodies directly into water[14]. The reason why cancer is not seen in sharks (not known in other fish) may be associated with the suppression of glutamine and DNA synthesis as a result of giving ammonia to the water. In such animals, cancer cells may not meet the increasing amine needs.

Body cells primarily subject the amino acids to deamination (keto acids are released). Bacteria primarily convert amino acids into polyamines by decarboxylation. Aerobic glycolysis of cancer cells is similar to bacteria.

Synthesis of serotonin, product of decarboyxlation, increases in carcinoid tumors[14]. This information is compatible with the hypothesis.

The substances recommended in this hypothesis are amino acids or the substances that can be converted into amino acids. Thus, their possibility of cell proliferation should also be considered.

Other hypothetical points to be investigated include aromatic amino acids and their analogs of alpha keto acid derivatives, prevention of pyruvate synthesis from lactic acid, prevention of alanine synthesis from lactic acid (lactic acid analogs), and supporting the mitochondrial functions.

Supporting Mitochondrial Functions

Coenzyme Q10 and aromatic amino acids such as phenylalanine and tyrosine, are involved in its synthesis, and unsaturated long chain fatty acids can support mitochondrial functions.

Coenzyme Q10

Coenzyme Q10 was primarily isolated from the mitochondria in bovine heart by Dr. Frederic Crane in 1957[7,24–26]. The ubiquinone that is known as Coenzyme Q10 is chemically 2.3-dimethoxy-5-methyl-6-decaprenyl-1.4-bensoquinone[25]. It is in the form of Coenzyme Q10 in humans, Coenzyme Q9 in mice, Coenzyme Q8 in E. coli, and Coenzyme Q6 in Saccharomyces cerevisiae[13].

The side chain of ubiquinone and cholesterol synthesis initiate from Acetyl-CoA and continue over mevalonic acid and isopentenyl diphosphate. Cholesterol, ubiquinone, vitamins A, E, K and terpenes are synthesized from isopentenyl diphosphate[27]. Aromatic ring of Coenzyme Q10 is synthesized from tyrosine. For its synthesis, B2, B3, B6, B12, C, folic acid, pantothenic acid, and some nutrient components are required[13,24–26]. It can be produced synthetically at a laboratory. Coenzyme Q10 can be produced in bacteria such as *Pseudomonas denitrificans* and *Agrobacterium tumefaciens* and numerous yeast media such as *Neurospora crassa and Aspergillus fumigatus*[24].

Coenzyme Q10 is found in sesame seeds, broccoli, soybeans, peanuts, cauliflower, oranges, strawberries, fish, meat, hearts, livers, kidneys, and eggs (all the animals)[24–26,28,29]. Food containing Coenzyme Q10 is divided in two as the ones containing more than 20 μg/g and less than 20 μg/g. Coenzyme Q10 is between 260–280 μg/g in pig heart, 8–200 μg/g in red meat and 4–64 μg/g in fish[24].

Danish people get 3–5 mg of Coenzyme Q10 from meat and poultry every day[7]. Swedish people get averagely 2–20 mg of Coenzyme Q10 every day, Japanese people get averagely 4.48 mg of Coenzyme Q10 every day in total. It is considered that men get 5.4 mg of Coenzyme Q10 and women get 3.8 mg of Coenzyme Q10 every day. It has been reported that frying calf heart decreases the amount of Co-enzyme Q10 by 30.58±1.37%, frying calf liver by 23.62±2.18% and boiling calf meat by 22.81±2.66%[24].

Aging, stress, and malnutrition reduce Coenzyme Q level in the body[7,24]. Coenzyme Q10 consumption increases by vigorous exercises, hypermetabolism, and acute shock situations. HMG CoA reductase inhibitors decrease the cholesterol and Coenzyme Q10 level[25].

In energy generation in mitochondria, enzymes such as Complex I (NADH-ubiquinon oxidoreductase: nicotinamide adenine dinucleotide dehydrogenase), Complex II (succinate-ubiquinon oxidoreductase: succinate dehydrogenase), Complex III (ubiquinol: ferrocytochrome C oxidoreductase: ubiquinon-cytochrome c reductase), Complex IV (ferrocytochrome c: oxygen oxidoreductase or cytochrome C oxidase), and Complex V (ATP synthase) take place[13,24]. Coenzyme Q10 is the co-enzyme of Complex I, II and III enzyme systems. These enzymes take part in electron transport and ATP synthesis[24,25]. Ubiquinone is converted to hydroxyquinon by receiving 2e in its quinone ring[24].

When Coenzyme Q10 is administered in pure form, it is absorbed orally at a low rate. When it is dissolved in fat and administered, its absorption rate increases. Its absorption occurs via passive diffusion[24,30]. Oral intake of ubiquinole oxidizes in ubiquinone in the stomach. After ubiquinone is reduced to ubiquinol in the intestines, it is absorbed from the small intestines in the form of chylomicrons and passes into the lymph circulation[13]. After it is converted to lipoproteins in the liver (VLDL, LDL), it mixes with blood[24,31]. When it is found mostly in the form of ubiquinone in the brain and lungs, it is found mostly in the form of ubiquinol in the blood (90–95%) and other tissues[13].

Fatty dispersions, emulsions, semi-emulsified systems, water-soluble powder formulations, and cyclodextrin complexes can be applied. It is known that the administration of Coenzyme Q10, in emulsified systems, and food increases bioavailability 3 times[24]. Oral

bioavailability of nanoparticles of Coenzyme Q10 is 4.28 times higher than its free form[30]. Also, cyclodextrins increase the oral bioavailability of Coenzyme Q10[24,30].

Coenzyme Q10 is efficient as an antioxidant[25,28]. Coenzyme Q10 inhibits lipid peroxidation by binding oxygen-derived radicals. Unstable free radicals become stable by receiving an electron from ubiquinol[24]. They act in membrane stability, cell signal, gene expression, cell proliferation, and control of apoptosis[13,24]. It slows down the aging process. It is protective against geriatric brain diseases. It modulates the gene expression of Coenzyme Q10 and repair DNA damage and mutations[28,29]. It strengthens the musculature and immune system[24]. It has been shown that Coenzyme Q10 reduces oxidative stress in diabetic patients, provides glycemic control, and decreases the HbA1 c levels. It is also reported that it prevents cardiovascular and neurodegenerative diseases and supports their treatment[24,25]. Coenzyme Q10 decreases the frequency of migraine attacks[26]. It is effective in mitochondrial encephalopathies[25] and Alzheimer[24]. It provides recovery in the symptoms of Parkinsonism[24,25,30].

In the patients with breast, lung, prostate, cervix, colon, rectal, and stomach cancer, it is reported that plasma-tissue Coenzyme Q10 level is low[7,29,32,33]. Low Coenzyme Q10 in the body emerges before malignancy[29]. The studies have revealed that Coenzyme Q10 supplementation increases the survival rate in breast, lung, ovary, kidney, brain, esophagus, stomach, and pancreas cancer[30]. In a survey conducted by naturopathic doctors in North America in 2002, it was reported that Coenzyme Q10 treated 77% of breast cancers. Coenzyme Q10 increases tumor regression and survival rate when taken together with food in addition to oral tamoxifen treatment[7]. Complete regression of tumor was observed after the administration of 90 mg/day Coenzyme Q10 to the patients with breast cancer[31]. Coenzyme Q10 with a dose of 600 mg daily was shown to be beneficial for older women with breast cancer. It needs to be clarified whether or not it is beneficial in the patients with terminal prostate cancer[34]. It was shown that the level of Coenzyme Q10 lowered in the blood of mice infected with the leukemia virus, and the mortality rate in mice treated with Coenzyme Q10 was 50% less than in untreated mice[32]. Coenzyme Q administered with diet to mice with dibenzpyrene-induced tumors delayed tumor formation and reduced tumor size and mortality rate[32,35]. It was found that co-administration of coenzyme Q10 and tamoxifen to rats with dimethylbenzanthracene (DMBA)-induced breast carcinoma increased antioxidant activity and decreased the risk of cancer recurrence and metastasis[31]. Coenzyme Q10 decreases MMP-2 activity, a metastatic protein, proportional to the dose and suppresses metastasis[36].

Compared to non-malignant cells, Coenzyme Q increases free radical production in malignant cells[30]. It has been shown that Bleomycin (BLM) treatment increases the content of Coenzyme Q10 in normal liver tissues of hamsters. This partially supports the idea that it may effectively protect against BLM-induced cytotoxicity[37].

100 mg Coenzyme Q10 was administered twice daily to 10 of 20 children with acute lymphoblastic leukemia or Hodgkin's lymphoma (180 mg/m^2 in total) and their electrocardiograms were examined. According to the study results, it is suggested that Coenzyme Q10 may protect myocardial toxicity[33]. It is thought that the agents such as Coenzyme Q10 may reduce tumor-related damage by inhibiting the effects of cytokines promoting tumor development[32]. Coenzyme Q10 strengthens the immune system. It is also revealed that it may increase immunoglobulin G and T4/T8 lymphocytes[33]. Coenzyme Q10 is beneficial in reproductive diseases[30]. Experimental data on the effects of co-enzymeQ10 in cancer treatment are important. Despite these data, its use in cancer treatment is unclear. It has the potential to be used as a preventive and supportive treatment.

Coenzyme Q10 may be administered via oral and intravenous route[7]. It is recommended to administer a dose of 10–100 mg per day[33]. When 100 mg Coenzyme Q10 is taken daily, this increases 1 μg/ml, which is its normal level in the blood, to 2 μg/ml[13,33]. It is reported that high toxicity will occur when 300 mg is administered per day[33]. However, some authors have reported that the toxicity of Coenzyme Q10 is low and doses up to 400–500 mg/day do not cause any clinically visible side effect[13,32]. Among the side effects, dizziness, headache, reflux, nausea, insomnia, fatigue, irritability, mild sensitivity, abdominal pain, and elevated liver enzymes can be listed[7]. Warfarin may reduce its anticoagulant effects[33]. Its effect in pregnant and breastfeeding women has not been known yet[25].

Conclusion

It is recommended to investigate whether the aromatic amino acids and their alpha-keto acid derivatives (including their analogs), as asserted in the hypothesis, have antitumoral effects through in vitro and in vivo methods.

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